# MICRO 2020 Fate and impact of microplastics: Knowledge and responsibilities.

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# MICBO2020 International conference

23-27 NOVEMBER 2020 LANZAROTE AND BEYOND\*

FATE AND IMPACTS OF MICROPLASTICS: KNOWLEDGE AND RESPONSIBILITIES



*MICRO\** is a biannual international conference; a collaborative process coordinated by the scientific community and stakeholders.

MICRO 2020 is hosted by the Marine Sciences For Society researchers' network and the World Network of Island and Coastal Biosphere Reserves, under the patronage of UNESCO.

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### Under COVID-19 conditions, MICRO 2020 was substantially online-based.

This 3rd edition of MICRO was a chance to contribute to fostering a collaborative effort among our continuously expanding MICRO community.

For this edition, **Local Nodes** played an invaluable role, they included:

Aalborg, Banyuls, Barcelona, Bastia, Bayreuth, Bouguenais, Boulogne sur mer, Braunschweig, Brussels, Bühl, Copenhagen, Créteil, Crozon, Exeter, Gran Canaria, Guyancourt, Helgoland, Ithaca, La Trinité sur Mer, Lanzarote, Leipzig, Madrid, Mallorca, Marne la Vallée, Mazatlán, Menorca, Oostende, Palavas, Plouzané, Plymouth, Rostock, Siena, Toronto, Trondheim, Urdaibai, Utrecht, Vigo and Wageningen.

We were more than 2000 authors thankful to be part of such an intense week. Hope the way we supported each other and our pursuits during the conference extends 'MICRO 2020: Lanzarote and Beyond' to 'MICRO 2022: OUTSIDE'

**Authors, Speakers, Chairpersons, Scientific Committee, Technical Crew, Core Group, Nodes and ALL participants**: SUCH A GREAT WEEK, accompanied throughout by Elinor Ostrom, Donella Meadows and Lynn Margulis.

THANKS Gaia !

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\_\_\_\_\_MICRO 2020. Fate and Impact of Microplastics: Knowledge and Responsibilities. 23-27 November 2020\_\_\_\_\_

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# **Core Group and Scientific Committee**

The Core Group for MICRO 2020 was constituted by a team of 11 people who guaranteed the steps along the road to a successful MICRO 2020 in connection with the Scientific Committee. In alphabetical order they are:

### Ana Carrasco, Andy Booth, Aquilino Miguelez, Arnaud Huvet, Bethany Jorgensen, Eva Cardona, Juan Baztan, Maria-Cristina Fossi, Melanie Bergmann, Richard C. Thompson and Sabine Pahl.

The Scientific Committee for MICRO 2020 was composed of 55 representatives from 37 institutions. They guaranteed the steps of the peer review process and supported the scientific quality and legitimacy of the work shared during the conference. In alphabetical order they are:

**Amy Uhrin,** National Oceanic and Atmospheric Administration; Ana Carrasco, Cabildo de Lanzarote; Ana Liria, Universidad de Las Palmas de Gran Canaria; Ana-Carolina Ruiz, Universidad Nacional Autónoma de México; Andreas Greiner, Universität Bayreuth; Andy Booth, Stiftelsen for industriell og teknisk forskning; Annika Jahnke, Helmholtz Centre for Environmental Research; Aquilino Miguelez, Cabildo de Lanzarote; **Arnaud Huvet,** Institut Français de Recherche pour l'Exploitation de la Mer; Bart Koelmans, Wageningen University; Bethany Jorgensen, Cornell University; Bruno Tassin, École des Ponts ParisTech; **Carlie Herring**, National Oceanic and Atmospheric Administration; **Carolin Völker,** Institute for Social-Ecological Research; Céline Surette, Université de Moncton; Chelsea M. Rochman, University of Toronto; Christian Laforsch, Universität Bayreuth; Christophe Maes, Institut de Recherche pour le Développement; Cristina Panti, Università degli Studi di Siena; **Elodie Martinez,** Institut de Recherche pour le Développement; Erik van Sebille, Utrecht University; Fabienne Lagarde, Université du Maine; François Galgani, Institut Français de Recherche pour l'Exploitation de la Mer; **Gunnar Gerdts,** Alfred Wegener Institute; Heidi Savelli-Soderberg, United Nations Environment Programme; Ika Paul-Pont, French National Centre for Scientific Research; Jesús Gago, Instituto Español de Oceanografía; Johan Robbens, Instituut voor Landbouw-en Visserijonderzoek; Johnny Gasperi, Université Gustave Eiffel; Juan Baztan, Marine Sciences For Society, CEARC; Juliana-A. Ivar-do-Sul, Leibniz Institute for Baltic Sea Research; Julie Forges, Université de Moncton; **Lisa Devriese,** Flanders Marine Institute: Lucía Viñas, Instituto Español de Oceanografía;

Maria-Cristina Fossi, Università degli Studi di Siena; Maria-Helena Costa, Universidade Nova de Lisboa; Marianne Krasny, Cornell University; Martin Wagner, Norwegian University of Science and Technology. Mateo Cordier, Université Paris-Saclay; Matthew Cole, Plymouth Marine Laboratory; Matthias Labrenz, Leibniz Institute for Baltic Sea Research; Melanie Bergmann, Alfred Wegener Institute; Monica Ferreira-da-Costa, Universidade Federal de Pernambuco; Nanna Hartmann, Technical University of Denmark; Nikolai Maximenko, University of Hawaii; **Omer Chouinard,** Université de Moncton; Patricia Ostiategui, Universidad de Las Palmas de Gran Canaria; Paula Sobral, Universidade Nova de Lisboa; **Pennie Lindegue,** Plymouth Marine Laboratory; Raimonds Ernsteins, Latvijas Universitate; Richard C. Thompson, Plymouth University; Sabine Pahl, Plymouth University: Salud Deudero, Instituto Español de Oceanografía; **Sonja Oberbeckmann,** Leibniz Institute for Baltic Sea Research; Thierry Huck, French National Centre for Scientific Research; Yong-Sik Ok, Korea University.



**Under the patronage of UNESCO**
## Chairpersons

The Chairpersons for MICRO 2020 were essential during the week-long conference, from facilitating smooth presentations to connecting the audience with the speakers. A very demanding week! In alphabetical order they are:

Adam Porter, Exeter; Alex Cortada. Menorca: Ana Liria, Gran Canaria; Ana-Carolina Ruiz, Mazatlán; Andy Booth, Trondheim; Annika Jahnke, Leipzig; Aquilino Miguelez, Lanzarote; Arnaud Huvet, Plouzané; Bart Koelmans, Wageningen; Bénédicte Morin, Palavas; Bethany Jorgensen, Ithaca: Bruno Tassin, Marne la Vallée; Isabelle Schulz, Brussels; Chelsea M. Rochman, Toronto; Chris Walkinshaw, Plymouth; Christian Laforsch, Bayreuth; Christophe Maes, Plouzané; Cristina Panti, Siena; Elke Brandes, Braunschweig; Erik van Sebille, Utrecht; Eva Cardona, Menorca; Fan Liu, Aalborg; Florence Parker-Jurd, Plymouth; François Galgani, Bastia; Guillaume Duflos, Boulogne sur mer; Gunnar Gerdts, Helgoland; Ignacio de Sobrino, Lanzarote; Ika Paul-Pont, Plouzané; Irene Brandts, Barcelona; Jean-François Ghiglione, Banyuls; Jesús Gago, Vigo; Johnny Gasperi, Bouguenais;

Juan Baztan, Crozon; Juliana-A. Ivar-do-Sul, Rostock; Lisa Devriese, Oostende: Manuel Monge-Ganuzas, Urdaibai; Maria Cristina Fossi, Siena; Maria Murcia, Menorca; Marta Sales, Menorca; Martin Loeder, Bayreuth; Martin Wagner, Trondheim; Mateo Cordier, Guyancourt; Matthew Cole, Plymouth; Matthias Völkl, Bayreuth; Melanie Pöhlmann, Bayreuth; Miguel Tamayo, Madrid; Miriam Weber, Bühl; Patricia Ostiategui, Gran Canaria; **Pennie Lindeque**, Plymouth; Rachel Coppock, Plymouth; Rachid Dris, Créteil; Richard C. Thompson, Plymouth; Sabine Pahl, Plymouth; Salud Deudero, Mallorca; Sascha Müller, Copenhagen; Seema Agarwal, Bayreuth; Sonja Oberbeckmann, Rostock; Stefan Peiffer, Bayreuth; Sven Frei, Bayreuth; Thierry Huck, Plouzané; Winnie Courtene-Jones, Plymouth; Xavier Cousin, Palavas; Yannick Lerat, La Trinité sur Mer; Zara Botterell, Plymouth.

Day 1/	'5, Mono	lay 23 <sup>rd</sup> ]	November	2020
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Day 1, Monday 23rd. November 2020					
8h30-8h45	Conference Launch Event				
9h-10h	<b>23.1_O</b>	23.1_Me	23.1_Ma		
10h-10h15	23.1_Gaia: 3 sessions brief				
10h30-11h30	23.2_0	23.2_Me	23.2_Ma		
11h30-11h45	23.2_Gaia: 3 sessions brief				
12h-13h	23.3_O	23.3_Me	23.3_Ma		
13h-13h15	23.3_Gaia: 3 sessions brief				
14h-15h	23.4_O	23.4_Me	23.4_Ma		
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15h30-16h30	23.5_O	23.5_Me	23.5_Ma		
16h30-16h45	23.5_Gaia: 3 sessions brief				
17h-18h	23.6_O	23.6_Me	23.6_Ma		
18h-18h15	23.6_Gaia: 3 sessions brief				
18h30-19h27	Highlight from Plymouth. Everywhere: Life in a littered world.				
19h30-20h	Poster.23.7_O	Poster.23.7_Me	Poster.23.7_Ma		
20h-20h30	Poster.23.8_O	Poster.23.8_Me	Poster.23.8_Ma		
20h30-21h	Poster.23.9_O	Poster.23.9_Me	Poster.23.9_Ma		

### Session 23.1\_O. Chaired by Andy Booth, Trondheim

## Measuring particle size distribution and mass concentration of microplastics and nanoplastics: opening an analytical challenge in the sub-micron size range

Caputo Fanny, Vogel Robert, Savage John, Vella Giacomo, Alice Law, Della Camera Giacomo, Hannon Gary, Peacock Ben, Mehn Dora, Ponti Jessica, Geiss Otmar, Aubert Dimitri, Prinamello Adriele, Calzolai Luigi. Paper number 332829

## An environmentally-friendly method for the identification of microplastics using density analysis

Barnett Symiah, Quintana Belén, Pietroluongo Guido, Miliou Anastasia. Paper number 334050

## Fate and removal rates of microplastics in multiple WWTP's utilizing different Biological Active filters (BAF)

Iordachescu Lucian, Papacharalampos Konstantinos, Denieul Marie-Pierre, Barritaud Lauriane, Baratto Gilles, Ingrand Valerie, Plessis Emmanuel, Vollertsen Jes. Paper number 334212

### Microplastic sample preparation methods for wastewater samples: Their applicability for submicro- & nano-plastic and the challenges ahead

*Al-Azzawi Mohammed S. M, Knoop Oliver, Drewes Jörg E.*. Paper number 334254

## Measuring particle size distribution and mass concentration of microplastics and nanoplastics: opening an analytical challenge in the sub-micron size range

Caputo Fanny, Vogel Robert, Savage John, Vella Giacomo, Alice Law, Della Camera Giacomo, Hannon Gary, Peacock Ben, Mehn Dora, Ponti Jessica, Geiss Otmar, Aubert Dimitri, Prinamello Adriele, Calzolai Luigi.

Are existing analytical methods for the characterization of nanoplastic (NP) and microplastic (MP) suitable for meeting the requirements coming from impeding legislation? In particular, the implementation of the proposal from the European Chemical Agency (ECHA) to restrict the NP and MP use in consumer products will require reliable methods to perform size and mass-based concentration measurements. Analytical challenges arise: (i) with highly polydisperse samples in the nanometric range, and also (ii) at the sub-micron interface, e.g. between 800 nm and 5 µm, where techniques applicable at the nanometre scale reach their upper limit of applicability and analytical approaches applicable at the micrometre scale have to be pushed to their lower limits of detection. In this work we compared the performances of nine analytical techniques by measuring the particle size distribution and mass-based concentration of quadrimodal polystyrene nanoparticle mixtures of 100-250 nm and of bimodal mixtures containing both nano and microparticles. We tested which analytical approaches can accurately and reliably measure highly polydisperse samples and bridge the gap at the sub-micrometre interface, with the educational aim to underline advantages, disadvantages and limitations of each technique. The results show that widely used light scattering based measurements, such as dynamic light scattering (DLS), do not have the resolution power to distinguish multiple populations in very complex samples. Nanoparticle tracking analysis (NTA), nano flow cytometry (nFCM) and asymmetric flow field flow fractionation coupled to static light scattering (AF4-MALS) measure the sample accurately in the nano-range (<1000 nm) but cannot measure particles in the micrometre range. Staticlight scattering (SLS) is not able to accurately detect particles below 200 nm, and simi-larly to transmission electron microscopy (TEM) and flow cytometry (FCM), is not suitable for accurate mass-based concentration measurements. Alternatives both for high-resolution particle sizing and mass-based concentration measurements between 100 nm and 5µm are tunable resistive pulse sensing (TRPS) and centrifugal liquid sedimentation (CLS). However, the combination of different instrumental set-ups, a priory knowledge of particle properties or advance data post-processing may be required for accurate and reliable measurements. Therefore, the combination of at least two complementary methods is suggested, to be selected on a case-by case basis on sample specific considerations. Despite the samples used in this study may not be considered truly representative of the complexity of secondary mi-cro(nano)plastic particles extracted from environmental matrices, this work could also give some indications of what analytical techniques may be suitable for sizing analysis of such complex samples in a defined size range.

Keywords : ECHA restriction , microplastic , Nanoplastic , particle concentration , particle size distribution , regulation , risk assessment.

# An environmentally-friendly method for the identification of microplastics using density analysis.

Barnett Symiah, Quintana Belén, Pietroluongo Guido, Miliou Anastasia.

The main objective of this study is to identify the type of polymer of microplastics using density analysis, utilising an affordable and accessible methodology. Current methodologies for microplastic polymer identification are not practical in field work especially in remote areas. The methodology here proposed offers a cost-effective, time-efficient and field alternative, useful to separate and identify plastics using their density. A considerable amount of literature has been published on the separation and identification of plastics using their density Morét-Ferguson et al., (2010) placed a piece of plastic in distilled water, if the plastic sank concentrated CaCl2 or SrCl2 was added until the plastic was neutrally buoyant. However, if the plastic floated, ethanol was added until the plastic was neutrally buoyant. Similarly, to identify unknown plastics, Kolb and Kolb (1991) created solutions with varying densities and then recorded which plastics floated or sank. Syakti (2017) extracted microplastics from environmental mediums based on the principle that different polymers have different densities. This study differs from other research since the proposed methodology successfully distinguished eight types of microplastics using their densities, only using water, sucrose and ethanol and gave a precise protocol for the preparation of the solutions. Most importantly, this method is suitable for micoroplastics. In this method, solutions of 8 different known densities were prepared following precise protocols. Microplastic particles are disposed in different test tubes containing solutions of different densities. Since different types of plastics have different densities, they will sink or float depending on the solution they are in. The buoyancy of the microplastics in the solutions is noted and used to identify the microplastics' polymer type.

Keywords : density analysis , environmentally friendly , identification , marine , microplastics , pollution.

# Fate and removal rates of microplastics in multiple WWTP's utilizing different Biological Active filters (BAF)

Iordachescu Lucian, Papacharalampos Konstantinos, Denieul Marie-Pierre, Barritaud Lauriane, Baratto Gilles, Ingrand Valerie, Plessis Emmanuel, Vollertsen Jes.

In recent years, wastewater treatment plants have been in focus as potential sources of microplastic (MP) pollution in receiving waters. However, several studies show that WWTPs remove between 80-99 % of synthetic particles from the influent. Due to discrepancies between sampling methods, sample preparation and characterization of MP, comparison between MP studies on WWTP's is though challenging. In this study, the distribution of MP in four WWTPs, three discharging into the Mediterranean Sea, is investigated applying identical sampling strategies and analytical methods, allowing direct comparison of the obtained results. The key features of the four WWTPs are: WWTP1 - Downflow BAF (Media 1) WWTP2 - Upflow BAF (Media 2) WWTP3 - Upflow BAF (Media 3) WWTP4 - Activated sludge/clarifier. Samples were taken in duplicates of the plant inlet, sludge, outlet and before the biofilters. Treated wastewater, was sampled by a filtration system where large amounts of water – typically some 1000 L – were filtered on 10 µm steel filters and the retentate then processed in the lab. Concentrated matrices, e.g. raw wastewater, were taken with an autosampler over 24 hours. The results represent samples collected during dry weather. Prior to analysis, the samples underwent extensive treatment. Subsequently, the concentrated samples were analyzed with a state-of-the-art FPA-µFTIR instrument in transmission mode, where MP's were quantified in terms of particle number and mass, polymer composition, size distribution in size range from  $10 - 500 \,\mu\text{m}$ . This technology was combined with an automated analysis of the vast amounts of data that such analysis produces, removing much of the human bias otherwise occurring during data analysis. Particles of 500 – 5000 µm were quantified by ATR-FTIR. The investigated WWTP's showed overall MP removal rates of 91-99.93 %, while the removal rates of the biofilters varied between 29.69 and 68.5 %.

Keywords : Activated sludge , biofilters , FTIR , Mediterranean , microplastics , Polymer type , Sample preparation , Sampling , Sludge , Treated wastewater , Wastewater , Wastewater treatment plants ,  $\mu FTIR$ 

## **Microplastic sample preparation methods for wastewater samples: Their applicability for sub-micro- & nano-plastic and the challenges ahead** Al-Azzawi Mohammed S. M, Knoop Oliver, Drewes Jörg E..

Environmental samples are complex and a quantification of microplastics often cannot be performed unless the plastic particles are separated from the accompanying natural matrix. Unfortunately, sample preparation methods microplastic analysis are not standardized vet. Furthermore, the various sample preparation methods in literature often lack a comprehensive validation to ensure that the microplastics are not affected by the sample preparation methods. Additionally, nanoplastics, on account of their smaller size, are more difficult to quantify than microplastics. In a previous study, common sample preparation methods for microplastic analysis were validated, and two suitable methods were presented; Fenton reaction and hydrogen peroxide. The two methods were shown to be effective at removing organic matter from sludge matrices without affecting the microplastic characteristics. These methods were validated for microplastics with sizes 80-330 µm. However, there is a concern that smaller microplastics and especially nanoplastics, due to their larger surface area to volume ratios, are more vulnerable to sample preparation. Thus, their characteristics, especially size, could be adversely affected during sample preparation. Therefore, the current study was designed for smaller microplastics (; 10 µm) to investigate the effects of sample preparation methods on sub-micro and nano-plastics. The main concern is the change of size distribution of these particles, as they can be lost due to sample preparation. First experiments were performed on nano-PS spheres (107 nm, 78 nm) using Nanoparticle Tracking Analysis (NTA). The results show a decrease in the average size of particles of about 10-15 % after exposure to a treatment with hydrogen peroxide. Fenton analysis was not possible as the reaction generates nano-iron precipitates, which impeded the measurement of the size distribution. Thus, solutions are being investigated to deal with this problem and to increase the accuracy of the measurement to allow an accurate reporting on size changes for both treatments.

Keywords : Digestion methods , Fenton , Microplastics , Peroxide

### Session 23.1\_Me. Chaired by Annika Jahnke, Leipzig

Effect of simulated treatments and weathering processes on the aging of land-based microplastics

Miranda Mariana N., Sampaio Maria J., Silva Adrián M. T., Pereira M. Fernando R.. Paper number 333899

**Are chemicals from aged plastic collected on beaches toxic for human cells?** *Le Bihanic Florane, Clérandeau Christelle, Cachot Jérôme, Morin Bénédicte.* Paper number 334289

**Induction of in-vitro toxicity by UV-weathered microplastic leachates** *Klein Kristina*. Paper number 334463

**Clothing laundry and wearing as sources of microfibre release to water and air** *De Falco Francesca, Cocca Mariacristina, Avella Maurizio, Thompson Richard C.*. Paper number 334487

### Effect of simulated treatments and weathering processes on the aging of landbased microplastics

Miranda Mariana N., Sampaio Maria J., Silva Adrián M. T., Pereira M. Fernando R..

After being released to the environment, microplastics can go through different aging processes that change their physical and chemical properties. These modifications are overlooked in many publications that use virgin microplastics in sorption and/or toxicity studies. Therefore, the main goal of this investigation is to test the degree of microplastics aging resulting from their exposure to ozone, UV light and simulated solar radiation, mimicking different effluent treatment processes or degradation due to environmental agents. In parallel, a three-month experiment was conducted for which the microplastics were exposed to real conditions of weathering, regarding temperature, sunlight, and humidity variations. All these experiments were performed for three polymer powders: low-density polyethylene (LDPE) with a mean particle size of around 500 µm, and polyethylene terephthalate (PET) and unplasticized polyvinylchloride (uPVC), both with a mean particle size of around 150 µm. The microplastic samples (virgin and modified) were analyzed by FTIR spectroscopy to infer about the intensity of oxidation, XRD for crystallinity characterization, and SEM to identify surface morphological changes. The Carbonyl Index, well described in the literature as a metric for the oxidation of polymers, was used here to quantify the changes in the chemical structure of the material. This allowed to estimate the aging evolution through exposure time, since samples were periodically collected for FTIR analysis. The results show that the three polymers are differently affected by the treatments tested. Furthermore, it can help us to understand what are the main aging mechanisms that can affect microplastics in urban environments and what modifications can result from those stressors, which can potentially lead to different impacts in the ecosystems and human health from those that are expected for virgin microplastics.

Keywords : LDPE , ozone , PET , solar radiation , uPVC , UV light

**Are chemicals from aged plastic collected on beaches toxic for human cells?** Le Bihanic Florane, Clérandeau Christelle, Cachot Jérôme, Morin Bénédicte.

Although there is increasing knowledge about toxicity of weathered plastic to aquatic organisms, really poor data are available for human health. Human can be exposed to plastic debris via consumption of contaminated seafood. As an example we have detected microplastics, including fibers, in marine food web of the South-West Atlantic coast up to  $0.66 \pm 0.96$  particle/individual in tissues of cultivated oyster (N = 40), and  $0.91 \pm 1.15$  particle/individual in seabass fish meat (N = 35). To investigate the chemical risk of plastic ingestion via contaminated seafood we have analyzed the level of cytoxicity, genotoxicity and oxidative stress induced by representative plastic debris collected on a South-West Atlantic beach. Hepatic cell line Hep G2 and intestinal cell line Caco-2 were exposed to DMSO extracts prepared from individual debris of PET, PVC, HDPE, PP and Nylon. No cytotoxicity of PET, HDPE, PP and Nylon has been observed up to 1 % (v/v) DMSO extracts. Only the 1 % PVC DMSO extract induced cytotoxicity to both Hep G2 and Caco-2 cells. DMSO extracts of 0.3 % Nylon and 0.3 % PP induced significant oxidative stress production to Hep G2 cells and 0.3 % Nylon to Caco-2 cells. Consequently, the comet assay revealed for both cell lines a significant induction of DNA damage after exposure to 0.3 % Nylon DMSO extract. PET DMSO extract at 0.3 % concentration also induced significant DNA damage to Hep G2 cells as well as 0.3 % HDPE and PVC to Caco-2 cells. Further analyses will be performed to identify the chemical fingerprints of the plastic debris that were extracted by the DMSO and could explained the observed toxicity. These results are of a high concern for the potential chemical risk of plastic ingestion to human health. This study is part of the ARPLASTIC research project.

Keywords : environmental aged plastic , genotoxicity , human cell line , oxidative stress

#### **Induction of in-vitro toxicity by UV-weathered microplastic leachates** Klein Kristina.

Microplastics are degraded continuously in the aquatic environment by abiotic and biotic factors such as UV-irradiation, temperature, mechanical abrasion, hydrolysis and bacteria. For this reason, chemicals that are typically incorporated into the polymer matrix can leach into the surrounding medium more readily. Although chemical effects of plastics have been examined recently with invitro bioassays, only a few studies have studied the impact of weathering on plastic leaching. Therefore, we investigated in-vitro effects of 12 different UV-weathered microplastic (pellets<5 mm) leachates, divided into virgin and recycled types of conventional plastics as well asbio-based and biodegradable plastics. Atmospheric weathering with subsequent leaching wasconducted with two irradiation types (UV-C and UV-A/B) and compared to a no irradiationtreatment, i.e., dark control (DC). In order to account for volatile substances, aquatic weath-ering with simultaneous leaching (UV-A/Bag) was performed. We further used nontargetanalysis to detect the number of leached chemicals. The oxidative stress response (AREc32 assay) was induced by the highest number of plasticleachates. Here, nine plastic leachates caused an elevated induction rate after UV-C and UV-A/Baq weathering. With regard to baseline toxicity (Mikrotox assay), seven UV-C irradiated plastic leachates induced unspecific toxicity. Anti-estrogenicity and anti-androgenicity activ-ities (yeasbased reporter gene assays) were almost congruent, with seven UV-C irradiatedleachates inhibiting the respective receptor. Following UV-A/Baq treatment, we detected 12340 chemical components in total. This study demonstrates that UV light led to the release of high chemical counts, of which UV-C most frequently induced toxicities. However, UV-A/Baq weathering was not generallyharmless due to high incidences and in some cases even higher activities when compared toUV-C irradiated samples. Finally, a recycled and bio-based plastic type induced very highin-vitro toxicities, although both forms are believed to be of beneficial use.

Keywords : antiandrogenicity , antiestrogenicity , Baseline toxicity , degradation , oxidative stress , weathering

**Clothing laundry and wearing as sources of microfibre release to water and air** De Falco Francesca, Cocca Mariacristina, Avella Maurizio, Thompson Richard C..

In the latest years, much attention have been raised on the contribution of textile washing processes to the global release of microplastics to marine environments. However, microplastics of fibrous shape have been found ubiquitous beyond just seas and oceans, but also in freshwater systems, soil and atmospheric deposition. This environmental problem is not only related to synthetic textiles like polyester, polyamides and acrylic, since also natural or artificial fibres like cotton and rayon have been detected in environmental samples. Therefore, the widespread occurrence of microfibres (synthetic, artificial or natural fibres ; 5 mm) in different environmental compartments, calls for more researches on the sources of this pollution and on parameters of influence. In this scenario, the present work aims to investigate the release of microfibres from garments to water by laundering and to air by wearing. In order to understand the influence of textile parameters on the release, polyester garments with different material compositions, fabric structure, varn twist, fiber type, and hairiness, were characterized and analysed. Tests to assess the release to water were performed by using a commercial washing machine and a multistep filtration procedure of the related wastewater. Tests to evaluate the release to air were carried out by involving volunteers wearing the selected garments in a controlled clean room and using passive samplers. Results showed how the release to both media is influenced by textile parameters and that the release of microfibres to air could be as important as that to water. Mitigation actions at the textile design stage should be considered and assessed to reduce the release of microfibres to both water and air.

Keywords : microfibres , microplastics , polyester , textile design , textiles

### Session 23.1\_Ma. Chaired by Arnaud Huvet, Plouzané

## Monitoring microplastics in central Great Barrier Reef surface waters: A temporal case study at the Yongala Shipwreck

*Miller Michaela, Kroon Frederieke, Hamann Mark.* Paper number 334185

## Bioavailability of steroid hormones sorbed on microplastics for aquatic organisms through biological fluids

Siri Cécilia, Masset Thibault, Liu Yang, Breider Florian. Paper number 334242

## Experimental evidence of physiological and behavioral effects of microplastics ingestion in Sparus aurata

Rios-Fuster Beatriz, Arechavala-Lopez Pablo, García-Marcos Karlos, Alomar Carme, Compa Montserrat, Alvarez Elvira, Deudero Salud. Paper number 334262

# Monitoring microplastics in central Great Barrier Reef surface waters: A temporal case study at the Yongala Shipwreck

Miller Michaela, Kroon Frederieke, Hamann Mark.

Plastic pollution, and specifically microplastics (; 5 mm), are ubiquitous within the marine environment, including surface waters, water column and benthic sediments. Marine plastic contamination is expected to increase if future projections of increased plastic production eventuate. In this context, scientists are increasingly interested in monitoring status of trends of microplastic pollution in the marine environment. In this study, we present results for the first three years of an on-going monitoring program to quantify microplastic contamination in surface waters of the central Great Barrier Reef, Australia. Specifically, surface seawater samples (n = 68) collected between September 2016 and September 2019 were processed for microplastic identification and quantification. Processing involved density separation followed by filtration, visual identification and sizing of putative microplastics using stereomicroscopy and ImageJ software, and chemical characterisation using Fourier transform infrared spectroscopy. A total of 845 putative microplastics were identified across all tows, consisting of fragments and fibres, with preliminary analyses indicating that polypropylene and polyethylene are the most common polymers. While microplastics were detected in every single tow, a clear temporal pattern in contamination has not been detected. This study serves as the first temporal assessment of microplastic contamination in surface waters of the Great Barrier Reef.

Keywords : Great Barrier Reef , Monitoring , Temporal Trends

# Bioavailability of steroid hormones sorbed on microplastics for aquatic organisms through biological fluids

Siri Cécilia, Masset Thibault, Liu Yang, Breider Florian.

Many studies have reported that hydrophobic organic contaminants have the ability to sorb on microplastics in aquatic ecosystem via different mechanisms of sorption. However, studies investigating desorption of such contaminants in organism fluids are lacking. The bioavailability of potential risk-compounds for aquatic organisms has to be assessed. It has been largely reported that adsorption onto microplastics is directly linked with hydrophobicity property of contaminants; the more hydrophobic the pollutant, the higher the sorption capacity. However, it is expected that the least hydrophobic molecules desorb more easily from microplastics. In our study, steroid hormones with different hydrophobicities, namely progesterone and testosterone, are used and their respective desorption from different types of microplastics is investigated in gastric and gut fluids. The idea is to discern if the ingestion of microplastics by organisms might promote desorption of potential sorbed micropollutants and therefore their bioavailability for such species. By using more complex composition of gastric and intestinal fluids separately and then sequentially, the purposes are to reproduce realistic digestive conditions and furthermore determine the organ which is the most prompt to enhance bioavailability of pollutants. Our results show that a higher desorption efficiency (58.6-73.0%, depending on plastic types) is reached in intestinal fluid than in freshwater (31.1-53.6%) or in gastric fluid (18.7-43.9%). Bile salts in intestinal fluid form micelles able to promote the solubilization of pollutants. Adsorption of pepsin onto microplastics has also been revealed, suggesting a competition between pollutant and pepsin for sorption sites and a potent reduction of pollutant solubilization. Our results indicate that progesterone daily intake via microplastics exposure is equal to 16.0µg/kg/day upon intestinal digestion, which represents about 13% of daily intake via water exposure. This observation suggests that ingestion is an additional route of exposure of pollutants and therefore emphasizes pollutants bioavailability for aquatic organisms.

Keywords : Desorption , Pollutants , Bioavailability , Biological fluids

# **Experimental evidence of physiological and behavioral effects of microplastics ingestion in Sparus aurata**

Rios-Fuster Beatriz, Arechavala-Lopez Pablo, García-Marcos Karlos, Alomar Carme, Compa Montserrat, Alvarez Elvira, Deudero Salud.

Increasing global research has identified microplastics to be impacting marine organisms. Here, we investigate the physiological and behavioral effects of thirty-six juvenile Sparus aurata exposed to microplastic enriched diets during a 21-day period. Physiological effects were assessed in liver and brain using the following biomarkers: activities of the antioxidant enzymes catalase (CAT), superoxide dismutase (SOD), glutathione peroxidase (GPx) and glutathione reductase (GRd), the detoxifying enzyme glutathione S-transferase (GST) and malondialdehyde (MDA) as indicative of lipid peroxidation. Individuals were recorded for behavior analysis (i.e. social interactions and feeding behavior). Results revealed an increased cellular stress from control to marinatedmicroplastic group, with virgin-microplastic group showing intermediate levels in all quantified biomarkers. Significant differences were found in liver for all biomarkers except for MDA, which suggests that the time of exposure to enriched plastic diets of this experiment is long enough to trigger the activation of antioxidant enzymes but not to produce cell damage by lipid peroxidation. In brain samples, the marinated-microplastic group presented significantly higher values for CAT and SOD, highlighting its function as primary antioxidants. Regarding behavioral effects, virgin and marinated groups displayed significantly more aggressive actions, such as biting and avoiding than control groups. In conclusion, a short period of microplastic exposure causes biomarker responses along with behavioral changes in fish. Nevertheless, further research is needed to assess long-term effects of microplastic ingestion and its potential consequences on fish populations.

Keywords : biomarkers , ecotoxicology , feeding behavior , plastic pollution , social interactions

### Session 23.2\_O. Chaired by Sven Frei, Bayreuth

**Laboratory testing of the behavior of microplastics in lake systems** *Elagami Hassan.* Paper number 333268

**Accumulation of microplastic in a Lower Rhine alluvial floodplain in Germany** *Rolf Markus, Laermanns Hannes, Steininger Florian, Löder Martin, Möller Julia, Bogner Christina.* Paper number 334325

### Loads, spatial patterns and potential toxicity of microplastics emitted from wastewater treatment plant effluents in rivers in Germany *Schmidt Christian, Kumar Rohini, Yang Soohyun, Büttner Olaf.* Paper number 334690

### Laboratory testing of the behavior of microplastics in lake systems

Elagami Hassan.

Plastics are among the most widespread contaminants on Earth. They build up with high concentrations in the different environmental compartments before reaching fresh water bodies. In thermally stratified lakes, microplastics can migrate between epilimnion, metalimnion and hypolimnion. This increases the probability of that microplastic can be ingested by filter feeders allowing microplastics to migrate through different trophic levels. In this study, the transport of microplastics in lakes is presented through systematic laboratory experiments. The settling velocities of various biodegradable and non-biodegradable particles with various shapes and sizes were measured in the settling column under laminar conditions using particle image velocimetry. The particles sizes ranged between 150 to 2400 µm. The experimental results presented that shape, size and density of the particle are the key parameters controlling the sedimentation behavior of the particles. The measured settling velocities ranged between 0.4 to 50 mm/s. Subsequently, the same particles used in the first lab experiments were incubated in a pond at the University of Bayreuth for 6, 8 and 10 weeks. The biofilm formation on the incubated particles was investigated using confocal laser scanning microscopy. Also, the effect of biofouling of microplastics on the physical properties and on the settling velocity of the particles was investigated experimentally. It was observed that biofilm-building organisms has only colonized few spots on the surface of MPs. The changes in shape, size and density of the incubated were negligible. Also, after 6, 8 and 10 weeks of incubation, no significant change in the settling velocity of the incubated particles was observed. The decrease in the settling velocity varied between 3 to 7 %.

Keywords : biofouling , lakes , microplastic transport , particle image velocimetry

### Accumulation of microplastic in a Lower Rhine alluvial floodplain in Germany

Rolf Markus, Laermanns Hannes, Steininger Florian, Löder Martin, Möller Julia, Bogner Christina.

River systems are major pathways for the transport of microplastics (MPs). The Rhine River is among the biggest rivers with respect to basin size and discharge in northwestern Europe. It is highly impacted by human activities with a multitude of different land use forms in its basin. Studies have documented the presence of MPs in the Rhine, along its course through Germany, as well as in several of its tributaries. However, the highly dynamic and ecologically sensitive alluvial floodplains of the Rhine River have not been investigated in detail so far. Knowledge on the state of MP contamination and on potential entry pathways of MP into meadows of the Rhine is essentially important for an ecological risk assessment. In this study we analysed the contamination level and distribution of MPs in the meadow soils of the Rhine. We investigated a study site in the northern periphery of Cologne in the nature reserve Merkenich-Langel. We hypothesize that the main entrance pathway of MPs into the meadow soil is via fluvial transport and flooding. Indeed, the site was regularly flooded in the past and agricultural fields, which could be another major source of MP input via surface runoff, are not present in the vicinity of the sampling site. We chose three sampling transects with increasing distance to the river water level located within the past flooded area. Three mixed samples from those transects were taken and analysed for MP concentrations via ATR-FTIR and µ-FPA-FTIR spectroscopy after density separation and enzymatic-oxidative purification. We found an increase of MP concentration per kg of dry soil in the depth 5-20 cm with increasing distance to the river (ML1: 25.612 particles/kg, ML2: 50.776 particles/kg & ML3: 85.076 particles/kg). We attribute this increase to differences in intensity and frequency of flooding.

Keywords : floodplain soils , Rhine , terrestial enviroments

# Loads, spatial patterns and potential toxicity of microplastics emitted from wastewater treatment plant effluents in rivers in Germany

Schmidt Christian, Kumar Rohini, Yang Soohyun, Büttner Olaf.

Wastewater treatment plant (WWTP) effluents are considered to be source of microplastics for rivers. Based on the a review of 19 studies, observed microplastic concentrations (size range between 10 and 5000 um) in 79 WWTP effluents ranged between 4x10<sup>o</sup> and 4.5x10<sup>5</sup> items/m<sup>3</sup> with a median of around 6400 items/m<sup>3</sup>. Combined with a Germany-wide data set of WWTPs, the resulting total, median microplastic load emitted by 9000 WWTPs in Germany is 7x10<sup>1</sup>2 items/year. Microplastic concentrations were simulated using a graph-based river network model which accounts for the spatial organization of the river network and the position of WWTPs. Microplastic transport was assumed to be without losses by sedimentation, entanglement or degradation. On average the simulated microplastic concentrations tended to increase with increasing stream order suggesting that WWTP effluents accumulate in the river network with a higher rate than river discharge. The resulting, simulated WWTP-derived in-stream concentrations are higher than observed in-stream concentrations which encompass all sources of microplastic, not only those emitted from WWTPs. This highlights that there is a need for a better understanding retention and remobilization of microplastics in rivers. Microplastic concentrations were compared to a species sensitivity distribution (SSD) of 8 freshwater species based derived from no-observed-effect concentrations and lowest-observed-effect concentrations. Both, observed microplastic concentrations as well as the considerably higher simulated, WWTP-derived microplastic concentration are approximately one order of magnitude below currently known toxic effect levels. It should be noted that differences between particles in terms of polymer type, shape and degree of ageing used for toxicity tests and those occurring in environmental samples may limit the comparability based on concentrations alone.

Keywords : river , river network , Wastewater treatment plant

### Session 23.2\_Me. Chaired by Bruno Tassin, Marne la Vallée

## Microplastics in a deep, dimictic lake of the North German Plain with special regard to vertical distribution patterns

*Tamminga Matthias, Fischer Elke.* Paper number 328229

## Exposure of nanoplastics to freeze-thaw leads to aggregation and reduced transport in model groundwater environments

Alimi Olubukola, Farner Jeffrey, Tufenkji Nathalie. Paper number 331662

#### **Microplastic concentration in active combined sewage overflow events** *Forrest Shaun, Vermaire Jesse, Mcmahonc Darryl, Adams Bill.* Paper number 332090

## Which parameters influence the infiltration behaviour of microplastic particles into fluvial sediment?

*Waldschläger Kryss.* Paper number 332418

# Microplastics in a deep, dimictic lake of the North German Plain with special regard to vertical distribution patterns

Tamminga Matthias, Fischer Elke.

Investigations for microplastics within the aquatic phase mainly concentrated on the surface of the world's oceans. However, this surface layer comprises only a small percentage of the global water volume and is unlikely representative of the total water body. While the prevalence of microplastics in the oceans has been investigated for several decades, the global freshwater systems have only recently received rising attention. Considering that lakes constitute 87 % of the world's liquid surface freshwater, it is necessary to quantify microplastics in limnic systems. Knowing the distribution of microplastics along all three spatial dimensions, including depth profiles, is crucial in this regard. As for marine investigations, investigations on microplastic pollution in freshwater have almost exclusively dealt with the surface water layer inducing considerable uncertainties. Heterogeneous sampling, purification, and identification strategies add to this uncertainty. To close this gap, we investigated the vertical profile of microplastic pollution in a deep, dimictic lake of the North German Plain. The present investigation is part of the project 'MICROLIM' that aims to budget microplastics within the model catchment area of Lake Tollense (Mecklenburg Western-Pomerania). Sampling was conducted at eight locations along a SW-NE transect (Fig. 1). At each location, samples comprising 1,000 l were taken at three depths (near-surface, 7 m and 10 m) by in situ pumpfiltration including particles between 63-5,000 µm in size. Three field campaigns (Sep. 2018 and Mar. 2019 and 2020) were carried out to assess seasonal differences (total n=72). Moreover, water temperature and conductivity were recorded to investigate the influence of water stratification on microplastic abundance. Thereby, we estimated the inventory of microplastics within the lake's water column.

Keywords : freshwater , lake , Nile Red staining , stratification , water column

# Exposure of nanoplastics to freeze-thaw leads to aggregation and reduced transport in model groundwater environments

Alimi Olubukola, Farner Jeffrey, Tufenkji Nathalie.

Plastic pollution presents a significant environmental challenge given the potential risks associated with freshwater contamination. Nanoplastics released into aquatic systems are affected by various physical, chemical and biological weathering processes that will influence their mobility in saturated porous media. Currently, however, little is known regarding the impacts that environmental conditions such as temperature and climate instability will have on nanoplastic transport and fate in the environment. To improve our understanding of nanoplastic mobility in groundwater following exposure to temperature cycles of freezing and thawing, controlled laboratory scale columns were used to investigate the transport potential of polystyrene nanoplastics exposed to either constant (10°C) temperatures or freeze-thaw (FT) cycles ( -10°C to 10°C) in water saturated quartz sand. The stability and transport of nanoplastic suspensions were examined both in the presence and absence of 5 mg/L natural organic matter (NOM) over a range of solution ionic strengths (IS) (3 - 100 mM NaCl). Under all conditions, exposure to 10 FT cycles led to significant aggregation and reduced mobility compared to nanoplastics held at 10 °C. This was especially apparent at low IS (3 and 10 mM NaCl) in the absence of NOM. Although the presence of NOM increased nanoplastic mobility, it did not prevent the aggregation of nanoplastics exposed to FT. Even though previous studies show that nanoplastics and nanomaterials are more mobile in cold temperatures, our findings illustrate that nanoplastics are more likely to be associated with soils and less likely to undergo long range transport in groundwater in colder climates following freezing temperatures. This highlights the need to account for climate and temperature changes when assessing the risks associated with nanoplastic release in aquatic systems.

Keywords : Cold climate , Nanoplastic fate in freshwaters , Nanoplastic transport , Plastic pollution , Porous media , Soil , Temperature , Weathering

#### Microplastic concentration in active combined sewage overflow events

Forrest Shaun, Vermaire Jesse, Mcmahonc Darryl, Adams Bill.

Preliminary results from an active combined sewage overflow event in the City of Ottawa, discharging to the Ottawa River, suggest microplastic concentrations are approximately 400 times greater during these active discharge events. The city of Ottawa Ontario and Gatineau Quebec have over 50 combined sewage overflow locations that direct sewage overflows into the Gatineau and Ottawa Rivers. One overflow location in downtown Ottawa has an indication signal of active overflow events. Baseline microplastic concentrations were measured at this location in addition to concentrations during an active overflow event. The baseline concentrations were 0.41 microplastics per cubic meter, with concentrations increasing to 167 microplastics per cubic metre during the overflow event. This demonstrates that sewage overflows are a significant conduit of microplastics to river environments and presents the necessity for more detailed investigations and analysis on this pathway of microplastics to freshwater systems.

Keywords : freshwater , microplastics , overflow , rivers , sewage , wastewater

# Which parameters influence the infiltration behaviour of microplastic particles into fluvial sediment?

Waldschläger Kryss.

To answer this question, an experimental test series was conducted and the results were compared to principles of classical sediment transport. An infiltration column was filled with glass spheres with different diameters (1.5 - 11 mm), in either uniform or bimodal packings. Subsequently, 21 microplastic particles with different properties (density, shape, diameter) were applied to the top of the glass spheres and a constant water flow was simulated for one hour. Finally, maximum and average infiltration depths of the individual microplastic particles were recorded. The infiltration depth of the particles increased with decreasing particle diameter (dMP) and with increasing diameter of the glass spheres (dGS). An examination of the differences in infiltration depth due to particle shape (pellets, fragments, fibres) has shown that spherical particles infiltrate deepest and that for fibres, a thinner diameter leads to a deeper infiltration. Density does not seem to influence the infiltration depth of microplastics. Additionally, it was assessed to what extent the ratio of the diameters of microplastics and glass spheres influences the infiltration depth. It was shown that the boundary values from fine sediment infiltration (dMP/dGS ¿ 0.32 = fine surface sealing; 0.11 ¿ dMP/dGS ¿ 0.32 = finite depth infiltration; dMP/dGS ; 0.11 = unimpeded static percolation) can also be applied to microplastics. Formulas were developed which can be used for defining the necessary depth of a sediment sampling, so that a comprehensive representation of microplastic contamination can be generated. The study therefore offers a first insight into microplastic infiltration in sediment. In the future, more studies should be carried out to determine depth-variable microplastic concentrations in sediment. In addition, studies should pay attention to the grain size of the sampled sediment, which has a strong influence on the infiltration depths of microplastics and thus strongly influences the microplastic concentrations in sediment.

Keywords : fine sediment infiltration , infiltration behaviour , microplastics , particle properties

### Session 23.2\_Ma. Chaired by Mateo Cordier, Guyancourt

Anthropogenic litter and microplastics in sediments from Lake Tollense, Germany *Hengstmann Elena*, *Fischer Elke*. Paper number 328311

**Cigarette butts: spatial and seasonal characterization in a sandy beach in South Brazil** *Conceição Gisele, Ramos Bruna, Lacerda Ana Luzia, Proietti Maíra.* Paper number 334507

Spatial and seasonal characterization of litter at a sandy beach in south Brazil: types, materials and sources

Ramos Bruna, Lameiro Fábio Rodrigues, Alencar Melanie Vianna, Lacerda Ana Luzia De Figueiredo, Proietti Maira. Paper number 334518

### Anthropogenic litter and microplastics in sediments from Lake Tollense, Germany

Hengstmann Elena, Fischer Elke.

Microplastic and especially macroplastic pollution data in freshwater systems is rare compared to marine systems. However, freshwater systems deserve equal consideration, as they are pathways of plastic to the ocean. Limnic basins may act as (temporary) sinks for plastics on this way. Aiming to identify sources for plastics and influences on its distribution in a limnic environment, anthropogenic litter (¿5 mm) was monitored semi-annually over a three-year period at Lake Tollense in Mecklenburg-Western Pomerania, Germany. Four sandy bank border segments were selected, representing various expositions and levels of anthropogenic influence. Additionally, beach and lakebed sediment samples were taken semi-annually over a two-year period to analyze microplastic occurrence. The total mean of macrolitter abundance is  $0.26 \pm 0.17$  items/m<sup>2</sup> and, always, macrolitter is predominately composed of plastic (75%). Cigarette butts are the most found items at all beaches investigated. Higher abundances were found in September compared to March showing the influence of anthropogenic activity on litter abundance. Furthermore, the transport of litter into the lake via tributaries seems to play an important role. Anthropogenic litter distribution was analyzed via images taken by unmanned aerial vehicles. These showed that litter did not accumulate at the strandline but rather at the edges of sandy beach areas where low vegetation is occurring. For the microplastic analysis, samples were processed in the laboratory by a first step of digestion of organic matter (H2O2 30%, HCl 10% and NaClO 6-14%) followed by wet sieving and the subsequent final separation via elutriation. Microplastics were identified using the Nile Red staining method and for a subset, polymers were identified using Raman spectroscopy. Results on microplastic occurrence will be presented comparing different sampling times as well as sampling points. The influence of anthropogenic activity, tributaries, and wind will be analyzed according to the macroplastic abundance at Lake Tollense.

Keywords : anthropogenic litter , beach , lake , microplastics , sediments

### Cigarette butts: spatial and seasonal characterization in a sandy beach in South Brazil

Conceição Gisele, Ramos Bruna, Lacerda Ana Luzia, Proietti Maíra.

Cigarette butts are the most common type of litter found in beaches worldwide: in the 2018 World Cleanup Day, Ocean Conservancy counted 5.7 million cigarette butts collected from beaches from 122 countries. Over 20 thousand of these butts were from the Brazilian coast, where they also represented the most abundant item. Cassino beach is an extensive sandy beach located in south Brazil, and has intense portuary, fishery and touristic activities. Beach use, especially tourism, is seasonal and concentrated near the urban area of the Cassino neighborhood. This study aimed to map the spatial and seasonal patterns of the amounts of cigarette butt at Cassino beach. During 27 months (between 2016 and 2019), through 200m2 transects (in triplicate) of the sand strip, we collected a total of 4,002 cigarette butts, representing 14.7% of total items. There was a direct relationship between human activities and the number of cigarette butts. During the austral summer, when beach use increases, the number of cigarette butts also increases: 2,709 were found in summer, compared with 80 in the winter. We also observed a higher concentration of cigarette butts at the dune base (7.80 items.100m2) than at the wrack line (1.70 items.100m2). Cigarette butts are an indicator of how beach activities are related to beach pollution, and they had a higher frequency in sites with more beach users when compared to areas with lower urban occupation. Discarded cigarette butts contain plastic filters and numerous toxic chemicals that can affect environmental and human health. To solve this problem, it is important to develop science-based public policies, with management actions focused on environmental education activities and an increase in the number of receptacles for the proper disposal of cigarette butts.

Keywords : chemicals , cigarettes , public policy , Solid waste

# Spatial and seasonal characterization of litter at a sandy beach in south Brazil: types, materials and sources

Ramos Bruna, Lameiro Fábio Rodrigues, Alencar Melanie Vianna, Lacerda Ana Luzia De Figueiredo, Proietti Maira.

Litter at sea is an ecological, economic, and social problem that impacts marine environments around the world. To create prevention and mitigation measures to solve this issue, it is necessary to understand the amounts and potential sources of this kind of pollution. Cassino Beach is an extensive (220km) sandy beach in Southern Brazil that presents multiple uses, with intense touristic, portuary and fishery activities. A peculiarity of this beach is that motor vehicles can transit on the sand strip, increasing beach use and possibly litter disposal. Seasonal touristic activities and the large extension of the beach allow comparisons of litter amounts, composition and possible sources between seasons and sampling sites. Sampling was done over 27 months at four points at the beach (2 urban, 2 nonurban); at each site, litter (¿2.5cm) present in three 200m2 transects was collected and evaluated. A total of 19,457 items were collected, and plastic composed 88% of litter. Paper, metal, and cloth items were also present, but in low amounts. In the plastic category, fragments and cigarette butts were the major types of litter, with abundances of 28.4% and 17.0% respectively. Urban sites presented higher amounts of litter than non-urban sites, and beach user-related litter source was more common in urban sites, emphasizing the contribution of beach use in marine litter deposition. Corroborating this, the summer season presented the highest litter density, and undetermined and/or beach use related sources were dominant in all sites and seasons. Mapping the predominant material, types and sources of marine litter creates important baseline data that can contribute not only to beach monitoring, but also to the development of litter reduction strategies.

Keywords : cigarette butts , coastal zone , debris , fragments , plastic pollution , seasonality , tourism

### Session 23.3\_O. Chaired by Martin Loeder, Bayreuth

Impact of microplastic on bacterial communities in soil and their impact on microplastic degradation capacity

Rohrbach Stephan, Gkoutselis Gerasimos Makis, Weig Alfons, Obst Martin, Rambold Gerhard, Horn Marcus A..

Paper number 333663

## Extracting microplastics from complex environmental samples dos, don'ts, and validation

Möller Julia, Heisel Ingrid, Satzger Anna, Oster Simon Jakob, Agarwal Seema, Löder Martin, Laforsch Christian. Paper number 334187

## Digging in the dirt: Does soil pollution affect colony founding in the black garden ant Lasius niger?

*Seidenath Dimitri, Otti Oliver, Holzinger Anja, Lücker Darleen, Kemnitz Klara, Feldhaar Heike.* Paper number 334412

# Impact of microplastic on bacterial communities in soil and their impact on microplastic degradation capacity

Rohrbach Stephan, Gkoutselis Gerasimos Makis, Weig Alfons, Obst Martin, Rambold Gerhard, Horn Marcus A..

High hydrophobicity, molecular weight and crystallinity are the main reasons for the longevity of petrol-based plastics. Plastic surfaces are colonizable by microbes, thus exerting a selection pressure on communities in the so-called 'plastisphere'. Microbes recruited into the 'plastisphere' might have a higher likelihood for being capable of plastic surface modification or even biodegradation. Certain prokaryotes inhere the ability to degrade hydrophobic and crystalline biopolymers and hydrophobic molecules such as lignin and wax. Indeed, few plastic-modifying or degrading strains have been discovered during the last decade, demonstrating that 'non-biodegradable' plastics can be subject to microbial conversion. Thus, our objectives were to determine the effect of polymer type on the plastisphere microbial community and to isolate microbes able to modify surfaces and/or biodegrade plastics. Habitats impacted by large amounts of these respective polymers like landfill soils might represent environmental enrichments of plastic colonizers and degraders. To address this hypothesis, we incubated various polymers packed in mesh-bags buried in a landfill soil, and determined bacterial community structure by 16S rRNA gene amplicon sequencing, biofilm formation by fluorescent staining of the extracellular matrix coupled to confocal laser scanning microscopy, and isolated plastic surface oxidizing Alphaproteobacteria. 16S rRNA gene amplicon sequencing and biofilm analysis demonstrated a polymer specific microbial community in tested samples. The acquired knowledge will help us to gain a deeper understanding of how bacterial communities interact with microplastic and associated potential impacts on soil functions. These insights might likewise open possibilities to develop microbial community-based risk management strategies and environmental-friendly systems to recycle plastic waste.

Keywords : Microbial Community , Plastic Biodegradation

# Extracting microplastics from complex environmental samples - dos, don'ts, and validation

Möller Julia, Heisel Ingrid, Satzger Anna, Oster Simon Jakob, Agarwal Seema, Löder Martin, Laforsch Christian.

Research on microplastics has been developing since 2004, with improved methods enabling the detection and characterization of ever-smaller microplastics. Initially, the field was limited to the marine environment, later expanding to freshwater systems and finally to the terrestrial environment. Soils are assumed to be a significant sink for microplastic pollution, but the extent of the contamination in soils is still widely unknown. This is mainly due to a lack of suitable methods capable of detecting a variety of small, particulate synthetic polymers in a complex particulate environmental matrix. We have developed a method for soil sample purification using a combination of sieving, density separation, and a novel sequential enzymatic digestion in order to purify soil samples of around 250g to such an extent that a particle analysis with focal plane array based µ-FTIR spectroscopy becomes possible. Overall, the protocol enables the removal of 299.9% (DW) of the bulk mineral mass and another average reduction of 77%(DW) of the lightweight organic fraction. Furthermore, the visual and molecular integrity of small particles in the size range of 100-400µm made of PE, PET, PA, PVC, and the biodegradable PLA were analyzed before and after undergoing the purification protocol. The presentation will also address the pitfalls we have encountered while developing the method. Furthermore, the importance of conducting validation for all methods and media used in microplastic research, as well as the significance of validating with chemically characterized spiking material, will be discussed. With this critical appraisal of how methods are developed, this talk aims to encourage a discussion targeted towards the final goal of devising harmonized procedures for more comparability within microplastic research.

Keywords : Analytical methods , Enzymatic digestion , FTIR spectroscopy , Purification protocol , Soil , Validation ,  $\mu$ 

# Digging in the dirt: Does soil pollution affect colony founding in the black garden ant *Lasius niger*?

Seidenath Dimitri, Otti Oliver, Holzinger Anja, Lücker Darleen, Kemnitz Klara, Feldhaar Heike.

Uncovering the various causes of the global decline in biodiversity is a major task of ecological research. One main driver are changes in land use, including intensification of agriculture and urbanization. Extensive research in this area has identified different factors, such as pesticides and nitrogen deposition, that negatively impact biodiversity. Other anthropogenic factors, such as plastic waste and air pollution, have only recently attracted increased research interest. Thus, it is largely unknown if pollutants that accumulate in the soil, such as manure, plastic and airborne particulate matter, pose a threat to soil dwelling organisms like insects. Moreover, the combinatorial effect of multiple stressors remains to be investigated. Here we exposed black garden ant Lasius niger queens at the colony founding stage to five pollutants in two concentrations. We compared their single effects to multiple stressor environments made from different combinations of the pollutants. When founding a colony, mated queens bury themselves into the ground and lay their brood on the bare soil. At this very sensitive stage small disruptions can prevent the successful establishment of a colony. To simulate soil contamination we used manure, brake dust, soot, microplastic particles and microplastic fibres. We measured the development time of the different life stages and queen survival. Once workers were present, we assessed the brood weight and number of offspring as a surrogate for colony founding success. Even though we expect to find negative effects of single pollutants, it might well be that significant effects only manifest when ant queens or brood are exposed to a combination of pollutants. Organisms might be able to compensate single effects but will be overstrained when facing multiple stressors. Our research aims to understand the importance of a range of pollutants on soil organisms and which role they play in the global biodiversity loss.

Keywords : insect decline , microplastic , multiple stressors , particulate matter , soil pollution

### Session 23.3\_Me. Chaired by Gunnar Gerdts, Helgoland

#### Developments in the identification of microplastics: the road ahead

Prata Joana C., Da Costa João P., Fernandes António José Silva, Da Costa Florinda Mendes, Duarte Armando C., Rocha-Santos Teresa. Paper number 334245

## Microplastics accumulation in surface waters of the Wadden Sea: The role of biogenic enrichment in marine foam

*Hernando Morales Victor, Ndhlovu Rachel T., Mol Anne, Vaksmaa Annika, Niemann Helge.* Paper number 334587

### Developments in the identification of microplastics: the road ahead

Prata Joana C., Da Costa João P., Fernandes António José Silva, Da Costa Florinda Mendes, Duarte Armando C., Rocha-Santos Teresa.

Microplastics are widespread contaminants that still lack standardized methods for their assessment. While most research includes a visual method of identification, the challenge increases when sampling smaller microplastics. Staining dyes, such as Nile Red, have become a popular objective criterion to identify small microplastics. Nile Red stains lipophilic particles and confers fluorescence under specific excitation wavelengths. However, Nile Red can stain natural particles and requires a precise sample preparation which may vary between matrices. This preparation is often based on organic matter removal using H2O2, for eliminating plant tissues, or KOH, for animal tissues. Observation can be made directly or under a microscope illuminated by a 470 nm light, ideally emitting  $\geq$ 1600 lux, and observed through an orange filter. Quantification should be made rapidly due to the risk of losing fluorescence and particles, with an average loss of 74% under 2 months. Despite the convenience of Nile Red, namely cheap and fast quantification of microplastics, it does not provide chemical characterization. In this case, spectroscopy methods capable of identifying particles down to a few µm are required, such as micro-Raman spectroscopy. These methods also require sample preparation to reduce interference and improve throughput. It is recommended that at least some particles should be characterized by spectroscopy, such as by characterizing all particles within 1 mm2 squares in the filter. However, these methods are also limited by a high cost, low availability, and low throughput. Therefore, a combination of identification methods is required for the quantification and characterization of microplastics to allow higher throughputs and the detection of smaller particles.

Keywords : Method development , Nile Red , Raman spectroscopy , Staining dyes

# Microplastics accumulation in surface waters of the Wadden Sea: The role of biogenic enrichment in marine foam

Hernando Morales Victor, Ndhlovu Rachel T., Mol Anne, Vaksmaa Annika, Niemann Helge.

Plastic pollution is a worldwide environmental problem with adverse consequences to marine life and human health. One of the characteristics that makes plastic useful and highly demanded compared with other materials is its lightness. Due to the low density, many plastics float making rivers a perfect route for millions of tons of plastic to seep into the ocean every year. However, a discrepancy exists between those vast quantities and the amounts of floating plastics traced back at the sea surface (1% of all plastic debris in the environment). This mismatch could be the result because a substantial fraction might have sunk to the ocean floor or deposited along the coast. Besides, the distribution of floating plastic is heterogeneous, which complicates abundance estimations. We have investigated aggregation of floating plastics in marine foam in Dutch waters as a result of Langmuir and other convergent currents. These currents are visible at the sea surface as streaks/lines of marine foam, which is mainly derived from extracellular polymers and other biogenic products. Foam and marine snow can influence the fate of macro- and microplastics in the ocean through emulsification, aggregation, biofilm formation, and/or sedimentation. However, studies on microplastics in these biogenic motorways are limited. We sampled a total of 12 parallel surface trawl transects at two locations of the Wadden Sea, inside and just adjacent but outside of the foam. Preliminary results of visible plastics on the supernatants showed microplastic abundances ranging from 0 to 0.140 pieces per m2. We found a significant difference between transects with 25 fold higher concentrations of microplastic particles in samples recovered from the foam. Our results show the need for an integrated effort in order to understand the effect of localized microplastic accumulation through currents and entrapment in foam on microplastics abundance and distribution in our oceans.

Keywords : distribution , extracellular polymeric substances , marine foam , microplastics , microplastics abundance
### Session 23.3\_Ma. Chaired by Juliana-A. Ivar-do-Sul, Rostock

**Geology of plastics and microplastics** *Ivar Do Sul Juliana*. Paper number 332827

**Historical microplastic contamination in sediments of Mirim Lagoon, RS-Brazil** *Alves Felipe, Veloso Carolina, Pinheiro Lara, Rodriguez Felipe, Agostini Vanessa, Pinho Grasiela.* Paper number 334302

## Microplastics history contamination in the Guarapiranga Reservoir (São Paulo, Brazil) coincides with the anthropocene

Landim Marina, Almeida Juliana B. S., Gerolin Cristiano R., Gonçalves Norberto S., Otero Vanessa, Melo Maria João, Labuto Geórgia, Semensatto Décio. Paper number 334489

#### Microplasticrusts on the remote Trindade Island, Brazil

Santos Fernanda, Fernandino Gerson, De Souza Maria Cristina, Angulo Rodolfo, Guedes Carlos. Paper number 334594

#### **Geology of plastics and microplastics**

Ivar Do Sul Juliana.

As the scientific literature on plastics and microplastics keeps increasing more than any other environmental contaminant of emerging concern, new avenues start to be explored by researchers which include diverse and complementary research fields. One of these new topics is the geology of plastics and microplastics, a multidisciplinary topic that will be explored on this section. Because plastics, mainly microplastics, are widespread within sedimentary compartments, and because they are not degraded in the environment, they might be preserved in sediment records as technofossils. Plastic polymers can then be explored as recent (ca. 70 years) anthropogenic markers that we have deliberately launched in terrestrial and aquatic environments. One ultimate use of microplastic as technofossils, once their preservation in the geological time scale is confirmed, is to feed discussion on the onset and potential formalization of the Anthropocene Epoch. In this section we welcome scientific works on macroplastics. mesoplastics and microplastics and their constituint polymers in consolidated or unconsolidated sediments and rock records from terrestrial and marine ecosystems, whether a range of disciplines such as paleoecology, sedimentology and archaeology can be explored in the context of plastic contaminants.

Keywords : Anthropocene Epoch , anthropogenic marker , preservation , sedimentary bodies , sink , technofossils

**Historical microplastic contamination in sediments of Mirim Lagoon, RS-Brazil** Alves Felipe, Veloso Carolina, Pinheiro Lara, Rodriguez Felipe, Agostini Vanessa, Pinho Grasiela.

The inappropriate disposal of plastics causes negative impacts on the environment. Despite being resistant, plastic is subject to degradation and fragmentation processes which result in the formation of microplastics (MPs, i.e. particles ; 5 mm). Currently, MPs are considered emerging contaminants and are virtually present in all aquatic ecosystems. The Mirim Lagoon is the second largest lagoon in South America, located on the border with Uruguay and its hydrographic basin is economically important for activities focused mainly on agriculture, fishing, irrigation and human supply. Despite of these anthropogenic activities, there are no research papers on plastic contamination in this environment. In order to characterize the potential sediment contamination with MPs of Mirim Lagoon, a gravity corer was used to sampling the corer in December 2018. In the laboratory, the core was sagittally opened. The sediment for MPs evaluation was carried out every 2 cm along the core. Then, sediments were dried in an oven at 40°C until constant weight. The isolation of potential MPs from the sediment was carried out using a saline flotation method using a NaCl solution (density 1.2 g.cm<sup>-3</sup>), followed by filtration of the supernatant using a filter paper (0.7 µm pore). The filters were analyzed under a stereomicroscope attached to a camera. Potential MPs were visually identified and classified according to type (fiber, fragment, pellet) and color. A total of 41 potential MPs were recorded in the top 80 cm of sedimentary sequence, being mostly fibers and blue in color. MPs distribution showed no contamination in the basal section of the core. On the 20 cm layer, a distinct increase in contamination was detected. Further work will involve determination of the polymer types and sediment age in order to stablish an Anthropocene marker for a better understanding of MP contamination in South America.

Keywords : Anthropocene, Paleoceanography, Plastics, Water Pollution

## Microplastics history contamination in the guarapiranga reservoir (são paulo, brazil) coincides with the anthropocene

Landim Marina, Almeida Juliana B. S., Gerolin Cristiano R., Gonçalves Norberto S., Otero Vanessa, Melo Maria João, Labuto Geórgia, Semensatto Décio.

Paper number 334489

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#### **Microplasticrusts on the remote Trindade Island - Brazil**

Santos Fernanda, Fernandino Gerson, De Souza Maria Cristina, Angulo Rodolfo, Guedes Carlos.

The global microplastic pollution achieved alarming levels posing challenges and affecting not only ecological or chemical systems, but also geological systems. In this scenario, recent studies have indicated the relationship between plastics and other marine litter items and geology, resulting in new terms such as plastiglomerates, pyroplastics, anthropoquinas and plasticrusts, being the latter described as partially melted plastic fragments encrusting to rock surfaces. A sample of remobilized sandstone pebble with a micro fraction of plasticrusts, we termed them as microplasticrusts, has been found on the remote Trindade Island (Brazilian offshore territory on the South Atlantic Ocean) at the Tartarugas (Turtles) Parcel beach. The microplasticrusts of the sampled pebble has been quantified and characterized according to size, color and overall texture.

Keywords : Geological System , Microplastic , Plastic litter

## Session 23.4\_O. Chaired by Ika Paul-Pont, Plouzané

Retrospective analysis of microplastics in *Mytilus edulis* over the last decades

Halbach Maurits, Vogel Miriam, Tammen Juliane, Rüdel Heinz, Koschorreck Jan, Scholz-Böttcher Barbara.

Paper number 334166

## **Evaluation of ecotoxicological consequences of photodegraded microplastics exposure in planktonic species**

Silva Luana, Silva Jerusa, Soroldoni Sanye, Agostini Vanessa, Pinho Grasiela. Paper number 334225

## Monthly survey of microplastic beaching in the Arcachon Bay (France) and relationship with environmental factors

Lefebvre Charlotte, Clerandeau Christelle, Le Bihanic Florane, Jalón Rojas Isabel, Morin Bénédicte, Lecomte Sophie, Cachot Jérome. Paper number 334395

### Physiological effects of microfiber ingestion by oyster larvae (Crassostrea virginica)

*Knauss Christine, Mcfarland Katherine.* Paper number 334586 **Retrospective analysis of microplastics in** *Mytilus edulis* **over the last decades** Halbach Maurits, Vogel Miriam, Tammen Juliane, Rüdel Heinz, Koschorreck Jan, Scholz-Böttcher Barbara.

Microplastic (MP) primarily originate from degradation of plastic litter is a ubiquitous environmental contaminant. Its bioavailability and a so far insufficiently predictable environmental hazard potential require both, current and retrospective investigations. Blue mussels (Mytilus spp.) are an established indicator organism for monitoring environmental pollutants. As sediment filtering organisms they are in direct interaction with natural and anthropogenic deposition. Further they serve amongst others as a human food source. Therefore, they reflect environmental pollution and have a link to food safety and human health aspects. The bioavailability of MP for blue mussels has been shown and make them ideal indicators for retrospective studies. The German Environmental Specimen Bank is sampling and archiving blue mussels on a regular basis at Eckwarderhörne (North Sea) and at Darsser Ort (Baltic Sea). Samples of these sites are available from 1986 (Eckwarderhörne) and 1992 (Darsser Ort) up to now. Blue mussel subsamples of the available time series were pooled into duplicates. Samples were processed through an enzymatic and chemically oxidative digestion for MP extraction. Purified samples were analyzed mass-quantitatively for 9 polymers by pyrolysis gaschromatography-mass spectrometry (Py-GCMS) [1, 2]. Resulting data reflect temporal variations of 7 polymers in blues mussels at both locations. The most prominent polymers are PVC, PET and PS. This is independent from the respective site, although the samples from Eckwarderhörne show slightly higher contents for PVC and PET. The elevated abundance of high-density polymers like PET and PVC supposed to sink in marine waters might indicate their enrichment in the sediment and thus the blue mussel habitat. This study presents the first mass-quantitative MP data in biota over recent decades and allows a first insight into trends of microplastics contamination. [1] M. Fischer, B.M. Scholz-Böttcher, Environmental science & technology 51, 5052 (2017). [2] M. Fischer, B.M. Scholz-Böttcher, Anal. Methods 11, 2489 (2019).

Keywords : Baltic Sea , mytilus edulis , North Sea , timeseries

## **Evaluation of ecotoxicological consequences of photodegraded microplastics exposure in planktonic species**

Silva Luana, Silva Jerusa, Soroldoni Sanye, Agostini Vanessa, Pinho Grasiela.

Plastics can be found in 90% of marine ecosystems. This problem increases with formation of microplastics, particles with diameter less than 5 mm. In aquatic environments, microplastics degrade by environmental factors, being UV radiation the most relevant among them. Furthermore, degradation can influence the plastic toxicity, releasing chemical additives. Therefore, the objective of this study was to analyse the microplastic toxicological potential in aquatic organisms, considering the polymer type and UV radiation exposing time of the particles (aging)[1]. Microplastics with 2 mm in diameter, derived of available commercial materials, were degraded using UV lamp (400W) in different radiation times (2, 8 and 30 hours), meaning 3 months, 1 and 4 years of environmental UV radiation exposition, respectively, following D6954-04 (ASTM) standard. Expanded polystyrene (EPS), polypropylene (PP) and ethylene–vinyl acetate (EVA) were the polymers used. Thereupon, in erlenmeyers having 100 mL of sterile water with salinity 0 and 35 were added photodegraded/aged and new/virgin particles, in different concentrations (5, 25 and 125 particles) according to literature. These solutions were shaken (30 rpm) for 24 and 168 hours in dark. Acute toxicity tests were performed in planktonic crustaceans Artemia salina and Daphnia magna, representing marine and freshwater systems, respectively. A. salina nauplii were exposed to leachates for 24 hours, being evaluated the lethality and alteration in swimming capacity. About the D. magna newborn with 2and 26-hours age, were exposed for 48 hours to further immobility evaluation. The bioassays showed no effects in both organisms tested. Although the acute bioassays have not shown significant results, the use of more sensitive endpoints such as behavioural response and biomarkers are suggest.

Keywords : Artemia salina , Daphnia magna , degradation , Toxicity

# Monthly survey of microplastic beaching in the Arcachon Bay (France) and relationship with environmental factors

Lefebvre Charlotte, Clerandeau Christelle, Le Bihanic Florane, Jalón Rojas Isabel, Morin Bénédicte, Lecomte Sophie, Cachot Jérome.

Since the last two decades, a great number of studies have described microplastics (MP) contamination in different marine ecosystems. These plastic particles have a size inferior to 5 mm and their ubiquity raise environmental, economic and societal issues. In the Arplastic project, we described the visible fraction of MP (from 0.5 to 5 mm) that beached with the high tide line. Three beaches located outside, at the mouth and at the back of the Arcachon Bay (SW France) were studied. During 2019, samplings were done each month at an intermediate tidal coefficient (around 80) and along a 100m longitudinal transect at the high-water mark. Every 25m, a quadrat of 50 cm length sides was laid on the high-water mark and all trapped MP were visually sorted from organic and mineral matter. Then, morphometric data were recorded for each particles (size, shape, color, roughness,..) on stereomicroscope and polymer identification was made by ATR-FTIR. Moreover, data on wind, waves, river flow and currents were analyzed to investigate the main environmental factors driving MP beaching. We found that MP were more abundant on the beach located at the mouth of the bay ( $36.0 \pm 39.2$  MP.m-2) than on back and outside stations (respectively  $2.7 \pm 4.4$  and  $1.75 \pm 2.4$  MP.m-2). This may be due to strong currents occurring at the entry of the embayment. Primary and secondary MP were both mainly reported as pellets and fragments represented respectively 43 % and 34 % of all recorded shapes. Polymers with low density were particularly abundant. Polyethylene represented 63 % of all the particles while polystyrene accounts for 19 % and polypropylene for 13 %. We also observed that MP were mostly dropped off on the beach during autumn and winter period when wind, waves and river flow were more important.

Keywords : beaching , distribution , environmental factors , microplastic , polymer

# Physiological effects of microfiber ingestion by oyster larvae (*Crassostrea virginica*)

Knauss Christine, Mcfarland Katherine.

A growing body of evidence shows that microplastic ingestion causes harmful effects in many marine organisms including bivalves, but little is understood on the effects of early life stage exposure, when most vulnerable to environmental stress. The Eastern Ovster (Crassostrea virginica) is a valuable ecological and economic resource along the east coast of the United States, making it important to understand the dynamics between microplastics and oysters. This study investigates the physiological response of C. virginica larvae during exposure to microfibers. Two polymers, commonly found in the coastal environment, were chosen for the microplastic exposure solutions: polyethylene terephthalate (PET, polyester) (50/50 mix of 14x14 and  $14x28 \mu$ m) and nylon 6,6 (50/50 mix 10x10 and 10x30 µm). Larvae (6-7 days-post-fertilization) were exposed to one type of polymer solution at 100 or 1000 microfibers mL-1 for 6 days. Physiological rates (respiration, algal clearance, carbon assimilation, and growth) were measured. Preliminary results showed that C. virginica larvae responded differently to PET and nylon even though the fibers were similar in size. PET caused a significant growth penalty at 1000 microfiber mL-1 after 6 days but nylon fibers caused a significant increase in growth for the same concentration. Both polymer fibers altered algal clearance rates and respiration rates. Further results of the other parameters measured will be presented and discussed. This work details the ingestion of microplastics during the larval stage and highlights the critical need to investigate impacts of microplastics at all life stages.

Keywords : Crassostrea virginica , microfiber , nylon , oyster , polyethylene terephthalate

### Session 23.4\_Me. Chaired by Maria Cristina Fossi, Siena

**Balancing the budget: how animals spend energy to combat the effects of microplastics** *Trestrail Charlene, Shimeta Jeff, Nugegoda Dayanthi.* Paper number 333532

**Microplastics and ship paint in the Weddell Sea, Antarctica** Leistenschneider Clara, Burkhardt-Holm Patricia, Gerdts Gunnar, Primpke Sebastian. Paper number 334248

## Microplastics in seafood: preliminary results on the occurrence and anatomical distribution in wild populations of *Nephrops norvegicus* from the Adriatic Sea

Martinelli Michela, Gomiero Alessio, Guicciardi Stefano, Emanuela Frapiccini, Strafella Pierluigi, Angelini Silvia, Domenichetti Filippo, Belardinelli Andrea, Colella Sabrina. Paper number 334509

### **Balancing the budget: how animals spend energy to combat the effects of microplastics**

Trestrail Charlene, Shimeta Jeff, Nugegoda Dayanthi.

Animals must carefully budget their energy expenditure to fuel various biological processes: biomolecule synthesis, cell division and gamete production must all be fuelled from an animal's limited energy reserve. Organisms that eat microplastics display reductions in these parameters. which suggests that microplastics alter the amount of energy animals allocate to these important processes. The Dynamic Energy Budget model can help researchers identify how animals that eat microplastics alter the amounts of energy they allocate to biological processes. The model outlines how energy is allocated amongst competing biological processes, and so it can be used to identify which energy pathways are affected by microplastics. In turn, this can assist researchers to identify the mechanisms behind organism-level responses to plastics ingestion – a vital step in progressing our understanding of how micro- and nanoplastics affect animals. Despite the utility offered by the Dynamic Energy Budget model – and that fact that it has been successfully used in other biolgical fields – the model remains largely unknown amongst microplastics researchers. Here, we highlight how the Dynamic Energy Budget model can contribute to a mechanistic understanding of micro- and nanoplastics effects in animals. This presentation will: Use the Dynamic Energy Budget model to explain how organisms allocate energy to biological processes, like growth and reproduction. Demonstrate how to use the Dynamic Energy Budget model to identify biological pathways that are affected by micro- and nanoplastics. Outline which endpoints researchers should measure to adopt the Dynamic Energy Budget model in their studies.

Keywords : energy model , mechanistic understanding , sublethal responses , unifying theory

#### Microplastics and ship paint in the Weddell Sea, Antarctica

Leistenschneider Clara, Burkhardt-Holm Patricia, Gerdts Gunnar, Primpke Sebastian.

Microplastics (MP) have been found in nearly all oceans and seas around the world with highest densities mostly recorded in areas directly impacted by human activities or the ocean gyres. Yet, MP pollution has also reached remote regions including the Southern Ocean surrounding Antarctica, where studies are relatively scarce. Further research is needed to help determine possible sources and areas vulnerable to MP accumulations. Ship paints (SP) containing synthetic polymers as e.g. polyurethane, alkyd and epoxy resins have recently been recognized as a MP source and can be released, inter alia, during normal ship operation. However, there is insufficient knowledge about the qualitative and quantitative contributions of MP from this source. In this study, 34 surface water samples were taken in the Weddell Sea one of the most inaccessible places in the Southern Ocean. ATR-FTIR spectroscopy of visually sorted particles (>300µm, n=771) revealed MP concentrations ranging between 0 and 0.04 MP m-3 (mean 0.01 MP m-3). Polyester accounted for 48% of all assigned particles followed by PE (16%), PP (10%), Acrylates/PUR/varnish (8%) and synthetic resins (7%). Half of the sorted fragments had similar visual characteristics as SP taken from the research vessel as reference. Most of these fragments (90%), however, could not be clearly assigned by ATR-FTIR (HQ <60%). To further examine the composition and source of potential SP fragments Raman spectroscopy was applied as a complementary technique. Multivariate analyses will be performed to compare the obtained FTIR and Raman spectra of environmental SP with reference samples from the vessel and the same SP freshly applied in the laboratory. Currently, measurements with micro X-ray fluorescence spectroscopy are performed to investigate the elemental composition of the SP particles. Besides providing evidence of MP in one of the most remote areas in the Southern Ocean, this study will evaluate the potential vessel-induced contamination of water samples and enable an evaluation of three different forensic methods to analyze environmental SP fragments.

Keywords : Antarctica , Microplastics , Ship paint , Southern Ocean , Surface water

## Microplastics in seafood: preliminary results on the occurrence and anatomical distribution in wild populations of *Nephrops norvegicus* from the Adriatic Sea

Martinelli Michela, Gomiero Alessio, Guicciardi Stefano, Emanuela Frapiccini, Strafella Pierluigi, Angelini Silvia, Domenichetti Filippo, Belardinelli Andrea, Colella Sabrina.

The omnipresence of plastic litter in the marine aquatic environments is a major environmental problem, and micron sized plastic particles have been detected in many species of fish and shellfish consumed by humans. The present study aimed at characterizing the occurrence, the shapes, dimensional classes, counts and polymer type of plastic ingestion by N. norvegicus individuals (n = 20) collected from two wild populations of the Adriatic Sea (Fig. 1), focusing on three different anatomical compartments (gut, hepatopancreas and tail), separately analysed. The outcomes of the present study point out that MPs were found in all the investigated individuals with an average of about 17 MPs/individual. Particles were predominant over fibers with a ratio of about 3:1. The majority of both the particles and fibers were in the dimensional range of 50 - 100 µm while the majority of the fibers were in the dimensional range of 50 µm- 300 µm (Table 1). The predominant polymers were Polyester, Polyamide 6, Polyvinyl Chloride and Polyethylene which were found in all the investigated individuals and that, together, constitute about 61% of all the MPs found. Among the three investigated compartments, particles concentrated more in the hepatopancreas than gut and tail, with no significant difference between the latter. While, fibres concentrated more in gut and hepatopancreas than in tail, with no significant difference between the first two. The dimensional class of the MPs significantly affected their anatomical distribution, finding only the smaller ones in the tail (p; 0.05). Overall, no statistical differences between individuals from the two sampling sites (off Ancona and Pomo Pits) were observed. Neither sex, length nor weight of the individual influenced the level of retained MPs.

Keywords : MPs uptake , N. norvegicus , Seafood safety , Tissue Partition , µFTIR imaging

## Session 23.4\_Ma. Chaired by Ana Liria, Gran Canaria

#### Quantifying microplastics across trophic levels in marine food webs of coastal British Columbia, Canada

Covernton Garth, Cox Kieran, Fleming Wendy, Buirs Brittany, Davies Hailey, Juanes Frances, Dudas Sarah, Dower John. Paper number 334452

#### Microplastic contamination on coral reef ecosystems around Lizard Island, Great Barrier Reef, Australia

Santana Marina, Motti Cherie, Van Herwerden Lynne, Kroon Frederieke. Paper number 334459

#### Cephalopods and litter at sea: interactions and potential impacts

*Freitas Tainah, Leite Tatiana, Proietti Maira.* Paper number 334522

#### Investigating the risks of plastic pollution in the Galapagos Marine Reserve, Ecuador

Jones Jen, Muñoz-Pérez Juan Pablo, Porter Adam, Galloway Tamara, Godley Brendan, Lewis Ceri. Paper number 334581

# Quantifying microplastics across trophic levels in marine food webs of coastal British Columbia, Canada

Covernton Garth, Cox Kieran, Fleming Wendy, Buirs Brittany, Davies Hailey, Juanes Frances, Dudas Sarah, Dower John.

Microplastic contamination in marine environments is of increasing concern as plastic particles have been found to be globally ubiquitous across ecosystems. Concentrations ranging from 8–9,200 particles/m3 have been found in seawater collected throughout coastal British Columbia (BC). A large variety of aquatic taxa have been shown to ingest microplastic particles (MPs), raising the potential for detrimental health effects following ingestion. However, the extent to which MPs can be accumulated and transferred through food webs remains unknown. We quantified MP uptake in bivalves, crabs, echinoderms, and fish that feed at different trophic levels at three sites on southern Vancouver Island. We paired stable isotope food web analysis with MP concentrations in digestive tracts across all trophic levels, and in the livers of the fish. We determined that MPs (100-5000 µm along their longest dimension) are not accumulating to the degree that would constitute biomagnification in marine coastal food webs. This was also true for other anthropogenic particles made of natural material, primarily cellulosic fibres. Moreover, there was no consistent correlation between digestive tract MP concentration and trophic position of the various species, or for the livers of fish. This suggests that if exposure is considered in terms of body weight, animals at lower trophic levels are at the greatest risk for any potential health effects of MP exposure.

Keywords : Bioaccumulation , Biomagnification , Food webs

### Microplastic contamination on coral reef ecosystems around Lizard Island, Great Barrier Reef, Australia

Santana Marina, Motti Cherie, Van Herwerden Lynne, Kroon Frederieke.

The coral reefs of the Great Barrier Reef World Heritage Area (GBRWHA) are experiencing significant anthropogenic pressures, including climate change and poor water quality associated with land-based runoff. Marine debris, and in particular microplastic pollution, has recently gained increasing attention as an additional anthropogenic threat to the marine ecosystems of the GBRWHA. Here, we provide a baseline of microplastic contamination on coral reef ecosystems at the remote Lizard Island Marine National Park in the far northern GBRWHA, approximately 250 km north from the nearest urban centre (Cairns). Microplastic contamination was characterized and quantified in seawater and sediment samples, and in coral reef organisms such as hard corals, sponges, sea cucumbers, sea squirts and fish. Putative microplastics were detected in all samples, and their polymer type confirmed using Fourier transform infrared spectroscopy. Preliminary analyses confirm the presence of secondary microplastic fibres and particles comprising a diverse range of polymer types and sizes, particularly in the water column. Using these results, we have established the first baseline of microplastic contamination on coral reef ecosystems around Lizard Island, which will inform future studies to examine the ecological risks of microplastic contamination on GBRWHA coral reefs and indeed globally.

Keywords : abiotic , marine environment , organisms , plastic , tropical

#### **Cephalopods and litter at sea: interactions and potential impacts**

Freitas Tainah, Leite Tatiana, Proietti Maira.

Litter in the ocean, which is mostly composed by plastics, has numerous negative effects on several organisms; however, the extent of such impacts is not well known for all groups. Few studies have explored the interaction of invertebrates with litter, but negative impacts have already been recorded for corals, polychaetes, crustaceans and cephalopods. Considering the ecological and economic importance of cephalopods, it is necessary to better understand the impacts of litter on this group. In this work, we review the types of documented interactions between cephalopods and litter, in order to evaluate impacts and knowledge gaps. We performed a scientometric analysis of data collected from scientific literature and citizen science, and found 21 scientific articles and 243 underwater photos, in which we recorded 23 species of benthic octopuses and 7 cuttlefish interacting with litter worldwide. Microplastic ingestion has been observed for the squid Dosidicus gigas, and the transfer of synthetic microfibers from the pelagic octopus Argonauta nourvi to its predators has been investigated. The largest number of records involved use of litter as dens, and transparent plastic and glass items were the materials most used (respectively 21.5% and 28.4%). Asia presented the largest number of records, especially Indonesia, and the octopus Amphioctopus marginatus was the most common species observed. With citizen science images we also observed the deposition of squid eggs on fishing gear, but the main recorded interaction was litter use as dens - which could represent a potential positive impact. However, it is necessary to clarify the ecological implications of this choice and its long-term consequences to organisms. Regarding ingestion, further studies are needed to elucidate its occurrence and impacts on cephalopods and their predators, including humans. Citizen science provided important information, highlighting its value and the need for more scientific investigations on the subject.

Keywords : Citzen Science , Debris , Dens , Ingeston , Octopuses

## **Investigating the risks of plastic pollution in the Galapagos Marine Reserve, Ecuador**

Jones Jen, Muñoz-Pérez Juan Pablo, Porter Adam, Galloway Tamara, Godley Brendan, Lewis Ceri.

Ecuador's Galapagos Islands and their unique biodiversity are a global conservation priority. Here we quantify widespread plastic contamination of the marine ecosystem and design a systematic risk scoring analysis to identify the most vulnerable species. We report contamination of seafloor sediments, surface seawater, beaches, marine invertebrates and algae. Beach aspect was identified as the most significant predictor of pollution with highest accumulation on the eastern coast indicating major input of plastic waste to Galapagos from the Humboldt Current, signalling to continental and maritime sources. Local littering and waste management leakages accounted for just 2% of plastic items, highlighting the need for a regional approach to mitigation. We identify priority species for further investigation including pinnipeds, seabirds, turtles, sharks and corals due to risk of entanglement and ingestion. Finally, we recommend a monitoring and management framework for marine plastic pollution in remote oceanic islands.

Keywords : islands , marine , microplastics , mitigation , risk assessment , sources

### Session 23.5\_O. Chaired by Mateo Cordier, Guyancourt

**Collaboration of scientists, NGOs and businesses for a plastic-free Mediterranean Sea** *Courtial Lucile, Le Texier Marie, Mauvais Manon.* Paper number 334338

## A freeware tool for the manufacturer independent analysis of microplastics: Systematic Identification of MicroPLastics in the Environment (siMPle)

*Vollertsen Jes, Primpke Sebastian, Vianello Alvise, Gerdts Gunnar.* Paper number 334354

#### The necessity for monitoring of microplastics in Serbia

*Teofilovic Vesna*, Živković Milica, Stojić Nataša, Pucarević Mira, Miletić Srdan, Vrvić Miroslav. Paper number 334526

#### **Riverbank macrolitter in the Dutch Rhine-Meuse Delta**

Van Emmerik Tim, Roebroek Caspar, De Winter Winnie, Vriend Paul, Boonstra Marijke, Hougee Merijn. Paper number 224555

Paper number 334555

## Collaboration of scientists, NGOs and businesses for a plastic-free Mediterranean Sea

Courtial Lucile, Le Texier Marie, Mauvais Manon.

Numerous initiatives aim to prevent plastic pollution in the Mediterranean Sea. Yet, this semienclosed sea has the highest plastic density in the world. The precise impacts of this pollution, especially from microplastics, are still unknown. Compagnies are still partly unaware of the various existing solutions. To be able to deal with these issues, the BeMed Business Club allows companies to improve their practices through enhanced collaboration with scientists, NGOs and across the value chain. As such, the Club paves the way to social change with the help of sound scientific support (INRA, IRD, CNRS, ENSAM). Through two working groups, the Club combines project implementation and scientific work. In the pilot project working group, companies around the Mediterranean basin implement concrete solutions in the field supported by BeMed's network of local stakeholders and experts. Within the science – industry dialogue working group, scientists share their knowledge with compagnies from the entire chain – producers, brand owners, retailers and waste management compagnies – for them to develop relevant and optimized alternatives to plastic to decrease their plastic footprint. Since January and despite the coronavirus crisis, five major companies from various sectors – food and beverage, cosmetics, tourism, waste management – have already joined the Club. The first pilot project on plastic reduction in the tourism sector will be implemented in 2021 in Marseille in collaboration with local stakeholders. Simultaneously, compagnies are working hand in hand with scientists on including the end-of-life of a product in the evaluation of the environmental impacts of alternatives. This work defines common ground base knowledge for their activities. Through this unique science – industry - NGO collaboration, solutions will emerge and be implemented for a plastic-free Mediterranean. The success of the Club's objectives relies on the scientists and compagnies' wish to take part in this innovative collaboration.

Keywords : Adaptable , Adjust , behaviour , Collaboration , Collaborative , environmental exigencies , Good practices , inclusive , Innovation , marine ecosystems , Optimized , Pave the way , Recovery , Revolutionary , social change , working groups

## A freeware tool for the manufacturer independent analysis of microplastics: Systematic Identification of MicroPLastics in the Environment (siMPle)

Vollertsen Jes, Primpke Sebastian, Vianello Alvise, Gerdts Gunnar.

The harmonization of microplastics analysis is a challenging field due to the various steps involved ranging from sampling, sample extraction to analysis. Each of these steps has its challenges and, in addition, different analytical methods based on spectroscopy or thermo-analysis are available, each one providing different data quality and comparability. State of the art FTIR imaging allows the analysis of complete filter areas independently from human bias in a relatively short amount of time. For the development of standardized operational protocols (SOPs), comparable data determination is hampered by the different manufacturer software and commercial software available. To overcome this challenge and allow the harmonization of data analysis, we developed the tool siMPle. It allows the analysis of datasets measured on different instruments from the manufacturers Agilent, Bruker, Perkin Elmer and Thermo Fisher Scientific (further imports in development). Here, every spectrum can either be selected individually or analyzed via two pipelines for the automated analysis, the original MPhunter- and the widely applied automated analysis pipeline. Large datasets with more than 3.2 million can be handled by it with relative ease and moreover, siMPle significantly reduces the calculation time to perform Pearson's correlation. In fact, a dataset containing 1 million spectra can be analyzed in around 2h instead of ¿ 24h. The generated data was benchmarked and validated in accordance with the original automated analysis approach using Bruker OPUS to allow a harmonized comparison of results. This new tool is available as freeware and allows the harmonization of MP data analysis for spectroscopic data for future research.

Keywords : Automated interpretation of FTIR imaging data , Microplastics analysis , software ,  $\mu\text{FTIR}$  imaging

#### The necessity for monitoring of microplastics in Serbia

Teofilovic Vesna, Živković Milica, Stojić Nataša, Pucarević Mira, Miletić Srdan, Vrvić Miroslav.

Microplastics are silent pollutants present in the almost every corner of the planet, even the most isolated. European legislative has recognized the necessity for the monitoring of microparticles, which involves the establishment of monitoring programmes, enabling the assessment of the status of marine waters on a regular basis. Serbia continuously adopts and harmonizes laws and regulations with the regulations of the European Union (EU). Unfortunately, microplastics is not yet the subject of our Regulations. Main source of pollution of oceans with microplastics are rivers, therefore it is necessary to estimate river microplastic emissions to the world's oceans. Microplastics find the way to rivers, mostly from the waste waters from washing machines. Serbia is country with long network of freshwaters, including the second largest river in Europe – Danube. River that flows through many industrial cities, and carries great industrial and communal burden. By detail analysis of Serbian legislative, it was found that no law or regulative considers microplastics as pollutant. Therefore, the monitoring of microplastics is not obligatory. We consider that it is time that Serbia finally recognizes this environmental problem, and includes it in the existing legal framework. We propose that the microplastics in sewage waters are monitored on the regular basis, including tap and bottled waters, to see if there is contamination with microplastics. This information will contribute to establishment of monitoring programmes in Serbia and thus prevent further microplastic pollution. ACKNOWLEDGMENT: This paper is funded by the Agreement on the implementation and financing of scientific research NIO in 2020 of Ministry of Education, Science and Technological Development of the Republic of Serbia (Grants No: 451-03-68/2020-14/ 200032; 451-03-68/2020-14/200134; 451-03-68/2020-14/200026)

Keywords : Legislative Framework , Microplastics , Rivers pollution

#### **Riverbank macrolitter in the Dutch Rhine-Meuse Delta**

Van Emmerik Tim, Roebroek Caspar, De Winter Winnie, Vriend Paul, Boonstra Marijke, Hougee Merijn.

(Plastic) litter in aquatic ecosystems negatively impacts ecosystems, species and economic activities. Rivers play a key role in transporting land-based waste towards the ocean [1]. A large portion is retained within river basins, e.g. in the estuary, in sediments and on the riverbanks. To effectively identify litter sources, sinks and transport mechanisms, reliable data are crucial. Such data can support optimizing litter prevention mitigation and clean-up efforts. We present the results of a twoyear monitoring campaign focused on riverbank macrolitter (¿0.5 cm) in the Dutch Rhine-Meuse delta. Between 2017 and 2019, volunteers sampled 152,415 litter items at 212 unique locations [2]. All items were categorized based on the River-OSPAR method (based on the OSPAR beach litter guidelines), which includes 110 specific item categories across ten parent categories. The median litter density was 2,060 items/km, and the most observed items were foam, hard, and soft plastic fragments (55.8 %). Plastic bottles, food wrappings and packaging, caps, lids and cotton swabs were the most abundant specific items. The litter density and most abundant items vary considerably between rivers, along the river, and over time. For both rivers, the highest litter density values were found at the Belgian (Meuse) and German (Rhine) borders, and at the Biesbosch National Park, the most downstream location. We provide a first scientific overview of the abundance, top item categories, and spatiotemporal variation of (plastic) litter on riverbanks in the Dutch Rhine-Meuse delta. In addition, we evaluate the used River-OSPAR method and provide suggestions for future long-term monitoring strategies. The results can be used by scientists and policy-makers for future litter monitoring, prevention and cleanup strategies. References [1] van Emmerik, T, and A Schwarz. Plastic debris in rivers. WIRE: Water (2020) [2] van Emmerik, T, et al. Riverbank macrolitter in the Dutch Rhine-Meuse delta. Environmental Research Letters (2020), https://doi.org/10.1088/1748-9326/abb2c6

Keywords : citizen science , Macroplastic , monitoring , observations , Rhine , river , riverbank

### Session 23.5\_Me. Chaired by Isabelle Schulz, Brussels

#### View online submitted version

**ANDROMEDA:** Analysis techniques for quantifying nano- and microplastic particles and their degradation in the marine environment Richard Sempéré

**HOTMIC: Horizontal and vertical oceanic distribution, transport, and impact of microplastics** Aaron Beck

**FACTS: Fluxes and Fate of Microplastics in Northern European Waters** Jes Vollertsen

microplastiX: Integrated approach on the fate of MicroPlastics (MPs) towards healthy marine ecosystems Luca Brandt

**i-plastic: Dispersion and impacts of micro- and nano-plastics in the tropical and temperate oceans: from regional land-ocean interface to the open ocean** Patrizia Ziveri

**RESPONSE: Toward a risk-based assessment of microplastic pollution in marine ecosystems** Francesco Regoli

### Session 23.5\_Ma. Chaired by Bart Koelmans, Wageningen

Aquatic Microplastic Studies: A critique and suggestions for the future *Weis Judith*. Paper number 331684

#### **MP2: Toxicity effect of micro-plastic and micro-pollutants in human cells** *Rubin Andrey Ethan, Sarkar Amit Kumar, Zucker Ines.* Paper number 332054

## Microplastics as vectors of triclosan in aquatic environment: biological effects on freshwater microalgae

Verdú Irene, Gonzalez-Pleiter Miguel, Rosal Roberto, Leganés Francisco, Fernandez-Piñas Francisca. Paper number 333749

## Application of thermal extraction/desorption-pyrolyse-GC/MS to investigate sorption kinetics of trace organic chemicals on (sub)microplastic

*Reichel Julia, Drewes Jörg E., Graßmann Johanna, Knoop Oliver.* Paper number 333830

#### **Aquatic Microplastic Studies: A critique and suggestions for the future** Weis Judith.

While there are numerous papers on microplastics (mps) being published every week, there is a need for improvement for the field to mature. Papers proliferate that report the numbers of mps found in water bodies, but they cannot be compared because there are no standard methods for collection and analysis. It is clear that using nets for sampling misses most of the microfibers, which are the most abundant form when whole water samples are analyzed, but which frequently go through the nets. Microfibers are released from synthetic fabrics in washing machines. Microscopic identification is common, but has a high error rate compared to chemical analytical equipment which can also identify the polymers. Microscopic analysis also can mistake cotton microfibers for plastic. Most animals studied eat mps; but it is more interesting to discover what attracts them – is it olfactory or visual? Once mps are swallowed, what proportion pass through the gut and are defecated vs get stuck and clog up the gut vs go through the gut wall and move into the tissues? Mps are considered a vector for transfer of toxic chemicals in the food chain, including chemicals that are additives in the plastic itself plus environmental chemicals that adsorb on the particle's surface. Future research should investigate what proportion of adsorbed contaminants are removed in the digestive system vs. staying bound tightly to the mps; this will vary with the chemical, and the anatomy and chemistry of the animal's digestive system. Most experimental studies to date tend to expose animals to microspheres, which can be purchased commercially but are very rare in the environment, and use concentrations that are far above environmental levels. In the future, exposure studies should use environmentally relevant concentrations and the shapes and sizes of mps that are most abundant in the environment.

Keywords : analysis , counting , digestive system , ingestion , microfibers , shapes , spheres , synthetic textiles , toxicity

## MP2: Toxicity effect of micro-plastic and micro-pollutants in human cells

Rubin Andrey Ethan, Sarkar Amit Kumar, Zucker Ines.

Presence of microplastic in the environment is increasingly being reported in natural and artificial water sources, soil, and even air. Plastic particles are detected everywhere, from North Pole snowflakes and deep sea near Japan to indoor environments of our offices and homes. Estimations show that by 2050, the total volume of plastic waste in the oceans will be greater than volume of the aquatic species themselves. Plastic particles can be divided to primary microplastics synthesized in a microscale range to various applications and secondary microplastics which are bulk plastic products degraded into small microparticles. The presence of primary and secondary microplastic in the aquatic environment poses risks to the environment and human health. Particularly, oral consumption of microplastic-containing in food and water by humans is a major potential throughput of microplastic into human body, suggesting gut epithelial cells as a primary target for their successful integration. Recent research focus is also given to the potential of microplastics to adsorb hydrophobic organic and inorganic micropollutants (e.g., pesticides, drugs, heavy metals). Thus, microplastics may act as vectors to aquatic pollutants into the human body. Our research focuses on the potential synergetic toxicity of microplastics and micropollutants. We conducted a set of adsorption tests using primary polystyrene microplastics with varying physicochemical properties and a commonly use pesticide (Triclosan) under environmentally - relevant conditions. Our results show that the microplastic surface functionality is a major factor which drives the adsorption. Further, we used human epithelial cells to evaluate the toxic effect of microplastics, micropollutants, and their combination. Our results suggest that microplastics act as a potential vector of micropollutants toward human cells, resulting in an increased toxicity due to elevated local concentration effects.

 $Keywords: adsorption\ ,\ Caco2\ ,\ Microplastic\ ,\ micropollutants\ ,\ PS\ beads\ ,\ toxicity\ ,\ Triclosan$ 

# Microplastics as vectors of triclosan in aquatic environment: biological effects on freshwater microalgae

Verdú Irene, Gonzalez-Pleiter Miguel, Rosal Roberto, Leganés Francisco, Fernandez-Piñas Francisca.

In recent years the presence of microplastics (MPs) in every ecosystem has evidenced that these particles are in contact and causing effects on the biota (1). MPs are frequently found in nature because of their incomplete retention in WWTPs (2). There, MPs can get in contact with other pollutants. Triclosan (TCS) is an antimicrobial compound used in cosmetics, and household products, reason why it is frequently found in wastewater. Some research points out that MPs can adsorb and desorb pollutants (3), but the role that these plastic particles could have as carriers of pollutants and the influence of this phenomenon on biota is even less-known. In order to know the potential of MPs to transport contaminants and their influence on the biota, in this study we have worked with 8 types of MPs beads (LDPE, PA 6'6, PET, PP, POM, PS, PLA, PCL), the antimicrobial compound TCS as pollutant and with the cyanobacterium Anabaena sp. PCC7120, primary producer in freshwater ecosystems, as model for the toxicity experiments. For 4 h MPs were exposed to TCS in water. Then, the pre-exposed beads were transferred to the culture medium in order to study desorption. To investigate the biological effects of this pre-exposed beads, the cyanobacterium cells were inoculated. After 72 h, growth and chlorophyll a content were evaluated, showing that LDPE, PA and POM were able to adsorb and desorb TCS, acting as carriers and making it bioavailable for the cyanobacterium which was affected by the desorbed TCS. These results reveal a relevant adsorption and desorption of TCS on MPs with eco-toxicological implications.

Keywords : desorption , freshwater , microalgae , sorption , vectors

# Application of thermal extraction/desorption-pyrolyse-GC/MS to investigate sorption kinetics of trace organic chemicals on (sub)microplastic

Reichel Julia, Drewes Jörg E., Graßmann Johanna, Knoop Oliver.

Micro- and nanoplastic particles are increasingly regarded as vectors for trace organic chemicals. The analysis of the sorbed trace organic chemicals (TOrCs) is most often depending on elaborate extraction steps. Within the thermodesorption-pyrolysis-gas chromatography- mass spectrometry (TD-Pyr-GC/MS) it is possible to identify sorbed trace organic chemicals on micro-, submicro- and nanoparticles as well as the type of polymer in one analytical setup. In the first step, the pollutants are desorbed from the particles by thermodesorption and analysed by GC/MS. Subsequently, the polymers are decomposed by pyrolysis and the decomposition products can be identified by GC/MS analysis. The aim of this study is to investigate sorption kinetics of the TOrCs phenanthrene,  $\alpha$ cypermethrin and triclosan on reference polymers in the micro-, sub-micro- and nanoscale, including: PMMA (48 µm), PE (48 µm) and PS (40 µm, 78 nm). For a comprehensive analysis of the samples, both aqueous phase and particles are analyzed, as indicated in Figure 1. The particles and TOrCs are incubated over a selected time period. The particles are separated from the aqueous phase by filtration and the TOrCs remaining in in the aqueous phase are quantified using a stir bar sorptive extraction (SBSE) and TD-GC/MS. Additionally, the trace organic chemicals on the particles and the polymer are analyzed with TD-Pyr-GC/MS. This comprehensive analysis allows a mass balance of the trace substances to be established. First results of the sorption kinetics and their dependence on polymer type and size can be shown.

Keywords : Adsorption , GC/MS , Microplastic , Nanoplastic , Pyr , Sorption kinetics , TD

### Session 23.6\_O. Chaired by Mateo Cordier, Guyancourt

Are we underestimating anthropogenic microfiber pollution? A critical review of occurrence, methods and reporting

Athey Samantha, Erdle Lisa. Paper number 333690

## Textile fibers in oceanic surface waters: a global perspective on their abundance and composition

Suaria Giuseppe, Achtypi Aikaterini, Perold Vonica, Lee Jasmine R., Pierucci Andrea, G. Bornman Thomas, Aliani Stefano, Ryan Peter. Paper number 334129

**Microplastics, tire wear particles and heavy metals in road dust from pervious pavements** *Rasmussen Lasse A., Stephansen Diana A., Vollertsen Jes.* Paper number 334270

**Plastic pollution and economic growth: the influence of corruption and lack of education** *Cordier Mateo, Uehara Takuro, Baztan Juan, Jorgensen Bethany.* Paper number 334722

# Are we underestimating anthropogenic microfiber pollution? A critical review of occurrence, methods and reporting

Athey Samantha, Erdle Lisa.

Anthropogenic microfibers, a ubiquitous environmental contaminant, can be categorized as synthetic, semi-synthetic or natural according to material of origin and production process. Although natural textiles fibers, such as cotton, are harvested from natural sources, chemical additives, including colorants and finishes, are applied to textiles to enhance performance and prevent degradation of the material. While most research and communication on microfibers has focused on the sources, pathways and effects of synthetic fibers in the environment, semi-synthetic and natural fibers are sufficiently persistent to undergo long-range transport and accumulate in the environment, where they may be ingested by biota. The challenges of enumeration and identification of natural and semi-synthetic fibers in environmental samples lead us to question whether current methods comprehensively capture and estimate inputs and occurrence of anthropogenic fibers in the environment. The goals of this study are to highlight gaps in research and to recommend best practices for enumeration and identification of anthropogenic fibers. We conducted a systematic literature review of 327 peer-reviewed, original research studies documenting microfibers in the environment. Our preliminary results show most microfiber studies focus on aquatic environments, with few focusing on the indoor or terrestrial environment. Roughly half of studies surveyed report the presence of natural or semi-synthetic fibers. Of the 129 studies surveyed thus far that report natural and/or semi-synthetic fibers, 71% employ digestion methods that have been demonstrated to cause destruction of naturally-derived anthropogenic fibers, indicating these studies may be underestimating anthropogenic microfiber abundance. Here we uncover major gaps in anthropogenic microfiber research, as well as identify best practice recommendations for successfully enumerating and identifying semi-synthetic and natural fibers from environmental samples. Further, we show that by focusing exclusively on the sources, pathways and effects of synthetic fibers in the environment, we are neglecting a major component of anthropogenic microfiber pollution.

Keywords : Anthropogenic fibers , Cotton , Microfibers , Microplastics , Textiles

# Textile fibers in oceanic surface waters: a global perspective on their abundance and composition

Suaria Giuseppe, Achtypi Aikaterini, Perold Vonica, Lee Jasmine R., Pierucci Andrea, G. Bornman Thomas, Aliani Stefano, Ryan Peter.

Textile fibres are ubiquitous contaminants of emerging concern. Traditionally ascribed to the 'microplastics' family, their widespread occurrence in the natural environment is commonly reported in plastic pollution studies, with the misleading belief that they largely derive from wear and tear of synthetic fabrics. Their synthetic nature has been largely used to motivate their persistence in the environment, thus explaining their presence in virtually all compartments of the planet, including sea-ice, deep-seas, soils, atmospheric fall-out, foods and drinks. As of today however, an extensive characterization of their polymeric composition has never been performed, even though the evidence that most of these fibres are not synthetic, is slowly emerging in the scientific literature. By compiling a global dataset of more than 916 seawater samples collected in six different ocean basins, we confirm that, although with some regional differences, textile fibres are ubiquitous in the world seas, but mainly composed of natural polymers. The chemical characterization of almost 2000 fibres through µFTIR techniques revealed that only 8.2% of oceanic fibres are actually synthetic, with the rest being predominantly of animal (12.3%) or vegetal origin (79.5%). These results demonstrate the widespread occurrence of cellulosic fibres in the marine environment, emphasizing the need for full chemical identification of these particles, before classifying them as microplastics. On the basis of our findings it appears critical to assess origins, impacts and degradation times of cellulosic fibers in the marine environment, as well as to assess the wider implications of a global overestimation of microplastic loads in natural ecosystems. Lastly, our results highlight a considerable mismatch between the global production of synthetic fibers and the current composition of marine fibers, a finding that clearly deserves further attention.

Keywords : fibres , FTIR , microfibers , microplastics , plastic pollution , surface waters

# Microplastics, tire wear particles and heavy metals in road dust from pervious pavements

Rasmussen Lasse A., Stephansen Diana A., Vollertsen Jes.

Pervious pavements are a relatively new technology for managing the increasing amount of stormwater runoff from urban areas. Besides managing stormwater, studies suggests that these systems can also improve water quality. Microplastics have gained significant attention in recent years and studies on large scale emissions highlight tire wear as one of the important sources for the spreading of microplastics to the environment. In this study, the deposits accumulating in pervious pavements from seven Danish roads were analysed. At each location road deposit were collected from upwards of 800 m2 of road by a designated street cleaning truck using high pressure rotors and suction. The collected material was transferred to a custom-made settling tank of 1 m3 and left for settling for three days. Initially, most of the water was removed by filtration on a 10 µm filter and the retained particle transferred back the to the settling tank. Afterwards a subsample were extracted by three glass sediment corers and combined. After extensive sample preparation involving density separation with sodium polytungstate at 2.0 g cm-3, Fentons oxidation and enzymatic digestion, samples were analysed by FPA-µFTIR imaging for the identification of microplastics and Py-GC-MS for tire wear. Furthermore, heavy metals were analysed with ICP-OES and organic carbon by a TOC analyser. Analysis of tire wear, heavy metals, and TOC are ongoing, while the FPA-µFTIR imaging is completed. The results showed a high presence of microplastic in road dust (excluding tire wear) with concentrations ranging from 1.45x105 to 1.19x106 particles/kg, corresponding to 7.6 to 100.4 mg/kg in estimated mass. Across all samples (excluding tire wear) the most common polymers identified were polypropylene (46.5 %), polyester (18.0 %) and polyamide (17.4 %). Concentrations of microplastic and identified polymers were similar to studies on microplastics in stormwater retention pond sediments.

Keywords : microplastics , pervious pavement , road dust , tire wear , urban runoff

# Plastic pollution and economic growth: the influence of corruption and lack of education

Cordier Mateo, Uehara Takuro, Baztan Juan, Jorgensen Bethany.

Green economic growth led by technological solutions is often mentioned as a solution for mitigating plastic pollution. However, economic growth appears to be in contradiction to planetary boundaries. By developing two worldwide socio-economic models, for forecasting inadequately managed plastic waste up to the year 2050 across 217 countries and territories, we demonstrate the adverse ecological impacts of the lack of regulatory processes and educational environmental programs. We used country-by-country data from the World Bank for the model estimates. The global cumulative stock of plastic waste that is inadequately managed is predicted to increase from 61–72 million metric tons (MT) in 1990 to 5109-5678 MT by 2050. Four scenario analyses told different stories: the business-as-usual (BAU) scenario, Mitigation scenario 1: capping GDP, Mitigation scenario 2: extending education, and Mitigation scenario 3: fighting corruption. In "Capping GDP," the annual amount of inadequately managed plastic waste slightly increases and reaches 64–119 million MT/year in 2050 instead of 61–110 million MT/year in the BAU scenario. In the "extending education" scenario, the amount decreases by 34% compared to the BAU scenario in 2050. In the "fighting corruption" scenario, the amount decreases by 60%. We provide further details in the country-by-country predictions.

Keywords : corruption , economic scenario , education , environmental Kuznets curve , plastic pollution , regression analysis

### Session 23.6\_Me. Chaired by Sabine Pahl, Plymouth

**Mussel power: A nature-based solution to marine microplastics?** *Cole Matthew, Coppock Rachel, Artioli Yuri, Torres Ricardo, Galli Giovanni, Yunnie Anna, Tom Vance, Lindeque Pennie.* 

**Decontamination of 6 sites in hard to access areas as a tool to change behavior** *Néollier Marie-Amélie.* Paper number 334386

**Plasticdemia: the "nurdles" case.** *Pietroluongo Guido, Azzolin Marta.* Paper number 334470

Paper number 333676
#### Mussel power: A nature-based solution to marine microplastics?

Cole Matthew, Coppock Rachel, Artioli Yuri, Torres Ricardo, Galli Giovanni, Yunnie Anna, Tom Vance, Lindeque Pennie.

Stemming the inputs of microplastic debris into the marine environment is a key societal challenge. Mussels are voracious filter-feeders, which provide an environmentally friendly means of removing waterborne pollutants (e.g. nitrates) and improving water quality. Here we explore whether mussels could be implemented as part of a nature-based solution to microplastics, stemming the flow of plastic particulates and microfibres from source to sea. We employ an interdisciplinary approach, comprising controlled laboratory exposures, modelling and stakeholder engagement, to evaluate the rate at which mussels can remove microplastics under different flow conditions, predict the fate of these microplastics and consider the practicalities of deployment. A custom-designed flume tank was used to demonstrate mussels (Mytilus spp.) can act as biological filters, efficiently removing waterborne microplastics from flowing water and repackaging these plastics into their faeces and pseudofaeces. Further laboratory experiments and computational modelling show microplasticsladen mussel faeces will rapidly sink out of the water column. We propose mussels could be used as an efficient, low-cost and environmentally friendly means of removing waterborne microplastics near source.

Keywords : bioremediation , ecology , mytilus

#### **Decontamination of 6 sites in hard to access areas as a tool to change behavior** Néollier Marie-Amélie.

The marine park of the Iroise sea (PNMI) covers a surface of 3500 square km. This area, enables us to take into consideration all the socio-economic activities where 'man' and sea interact in a coherent way. Established in 2007, the PNMI guickly focused towards tackling the macro-waste problem. In 2010, we started work on our first project based on the OSPAR protocol. Our findings ran contrary to the national data; it appears that 70% of the macro-waste in Iroise Sea was of maritime origin. 13% of that pollution was contributed by 'Industrial fishing', making it the most polluting activity. In 2019 the park became a partner of the cross-Channel project Preventing Plastic Pollution. The team's experience acquired allowed us to come up with a wide range of solutions to provide actionable plans against the rise of macro-waste. As a result of these plans macro waste from industrial fishing in harbours and ports became a target. The plan presented, involves the decontamination of 6 sites in hard to access areas. We have concentrated our work on the Molene archipelago islands, in ports and harbour areas as well as an old open garbage dump at the bottom of a cliff on the island of Ouessant. These actions rely on several parameters. First, governance; All those involved need to be able to take responsibility for their actions (trainings, meetings). Second, expertise; Macro waste removed needs to be quantified and qualified (report post decontamination). Third, environmental education; Each action will be planned and targeted and educationnal tools will be created (durable panels, trainings about the impact of macro-waste pollution, beach cleanups). We are working in areas where macro-waste has been identified. Through those processes and by enabling and working alonsgside different local actors, we wish to maintain the benefit of these actions over time.

Keywords : decontamination of sites in hard access areas , education , Governance , harbour , islands

#### Plasticdemia: the "nurdles" case

Pietroluongo Guido, Azzolin Marta.

While globally public attention and policy decisions are mainly focused on single-use plastic and the microplastic degradation process, the impact and spread of other plastic sources, such as fishing nets and raw materials, are underestimated. This is the case of nurdles: raw material in the form of pellets from which any plastic product is made of. Nurdles manufactory industries are located in specific areas where their spill is more concentrated. Still, the yearly loss of billions of nurdles is a consequence of all the plastic item production process and transport. For their size, nurdles are considered microplastic and, depending on the final product, they can be made of different colour. shape and by a single or a mix of different polymers. In the environment, and especially in the marine ecosystem, like any other plastic, they are persistent, not biodegradable and carrier of toxins. Using a free access database (www.nurdlehunt.org.uk) and a beach patrol survey method from January to August 2020, the Adriatic Sea was used as a special area due to its particular closed geography and the absence of nurdles industries in the Mediterranean Sea. The result of this survey showed that nurdles of any kind are widely present along the whole Italian coastline. Nurdles environment domination is totally out of control, despite some industries have adopted its own guidance to reduce the loss and a few voluntary groups promote best practices along the entire plastics supply chain. Plastic invention helped humanity in many scopes but now it's time to select which kind of plastic is essential for human life. If public perception, activism, scientists and policy will continue to focus on littering and clean-up aspect and not on the source of the production and the education of public habits and choices the issue will never come to a solution.

Keywords : change perception , human health , marine litter , microplastic , nurdles , plastic policy , plastic pollution , plastic spill

#### Session 23.6\_Ma. Chaired by Salud Deudero, Mallorca

#### Trapping and removal of marine plastics by seagrasses

Sanchez-Vidal Anna, Canals Miquel, De Haan William P., Romero Javier, Veny Marta. Paper number 334106

**Bioavailability of microplastics to marine zooplankton: effect of shape and infochemicals** Botterell Zara, Beaumont Nicola, Cole Matthew, Hopkins Frances, Steinke Michael, Thompson Richard, Lindeque Penelope. Paper number 334107

#### **Occurrence of microplastic in commercial fish from Northern Java Sea, Indonesia** Yona Defri, Harlyan Ledhyane Ika, Fuad M. Arif Zainul, Prananto Yuniar Ponco. Paper number 334148

#### First global map of risks of microplastic in the ocean surface

Everaert Gert, De Rijcke Maarten, Lonneville Britt, Janssen Colin, Backhaus Thomas, Mees Jan, Van Sebille Erik, Koelmans Albert, Catarino Ana I, V, egehuchte Michiel. Paper number 334357

#### Trapping and removal of marine plastics by seagrasses

Sanchez-Vidal Anna, Canals Miquel, De Haan William P., Romero Javier, Veny Marta.

One of the main challenges of present research on plastic pollution is to estimate the amount of plastic debris in the ocean, and understand their origin, fluxes and pathways, and where they ultimately accumulate. The seafloor is a sink for plastic debris. However, little is known on the fate of plastic debris once there. Here we investigate microplastics and larger plastic debris within the so-called "Neptune balls", formed by debris of the seagrass Posidonia oceanica washed up on beaches of a Mediterranean Sea island. We found up to 1470 plastic items per kg of seagrass, which were essentially filaments and fibers of polyamide and fragments of polyethylene terephthalate, polyethylene, and polypropylene. Our findings show that seagrass meadows ease the accumulation of high-density plastic debris and their entanglement with natural lignocellulosic fibers detached from Posidonia under relatively calm conditions. Plastic trapping Neptune balls are washed ashore during stormy conditions. Our results evidence how a Mediterranean key habitat supporting fisheries, sequestering carbon and providing coastal protection, also counteracts plastic pollution.

Keywords : microplastics , seaballs , seafloor , seagrass

#### Bioavailability of microplastics to marine zooplankton: effect of shape and infochemicals

Botterell Zara, Beaumont Nicola, Cole Matthew, Hopkins Frances, Steinke Michael, Thompson Richard, Lindeque Penelope.

The underlying mechanisms that influence microplastic ingestion in marine zooplankton remain poorly understood. We investigate how microplastics of a variety of shapes (bead, fibre and fragment), in combination with the algal-derived infochemicals dimethyl sulfide (DMS) and dimethylsulfoniopropionate (DMSP), affect the ingestion rate of microplastics in three species of zooplankton, the copepods *Calanus helgolandicus* and *Acartia tonsa*, and larvae of the European lobster *Homarus gammarus*. We show that shape affects microplastic bioavailability to different species of zooplankton, with each species ingesting significantly more of a certain shape: C. helgolandicus – fragments (P<0.05); A. tonsa – fibres (P<0.01); H. gammarus larvae – beads (P<0.05). Thus, different feeding strategies between species may affect shape selectivity. Our results also showed significantly increased ingestion rates by C. helgolandicus on all microplastics that were infused with DMS (P<0.01), and by *H. gammarus* larvae and *A. tonsa* on DMS-infused fibres and fragments (P<0.05). By using a range of more environmentally relevant microplastics, our findings highlight how the feeding strategies of different zooplankton species may influence their susceptibility to microplastic ingestion. Furthermore, our novel study suggests that species reliant on chemosensory cues to locate their prey may be at an increased risk of ingesting aged microplastics in the marine environment.

Keywords : Copepod , selectivity , Zooplankton

#### **Occurrence of microplastic in commercial fish from Northern Java Sea, Indonesia** Yona Defri, Harlyan Ledhyane Ika, Fuad M. Arif Zainul, Prananto Yuniar Ponco.

The occurrence of microplastic in the marine environment could lead to the accumulation of microplastic in the marine biota. The ingestion of microplastic in marine fishes has been studied intensively worldwide. This study aims to assess microplastic concentration in two commercial fish species vellowstripe cad (Selaroides leptolepis) and common ponyfish (Leiognathus equulus) from the northern Java Sea, Indonesia. Ten fish samples from each species were collected directly from the fishermen in the Brondong Port, Lamongan. Microplastics were visually identified using microscope from three different organs (gill, gastrointestinal tract and muscle) after organic matter digestion. In total, 117 items and 97 items of microplastics were extracted from S. leptolepis and L. equulus, respectively. The abundance of microplastics in the S. leptolepis was in the range of 0.2–5.56 item g-1 and in the L. equulus was 0.2-8.83 item g-1. The results show that both species contained microplastics in rather similar number. The abundance of microplastics were counted higher in gills followed by gastrointestinal tracts and muscle for both fish species. Gill and gastrointestinal tracts were considered as direct pathways for microplastics to enter fish tissues. Three types of microplastic were recorded with fiber was the dominant type of microplastic collected from most of the fish organs. Fragments appeared in higher number in the gills for both species, while film was observed in a very small number in all the fish organs. Fiber has been known to accumulate in fish from many different studies and it is related to the abundance of fiber in the aquatic environment.

Keywords : Common ponyfish , fish organs , Java Island , plastic pollution , yellowstripe cad

#### First global map of risks of microplastic in the ocean surface

Everaert Gert, De Rijcke Maarten, Lonneville Britt, Janssen Colin, Backhaus Thomas, Mees Jan, Van Sebille Erik, Koelmans Albert, Catarino Ana I, V, egehuchte Michiel.

Current studies on the risk of microplastic (MP) in the aquatic environment suggest that the in situ concentrations are on average several orders of magnitude lower than the concentrations where effects are expected to occur. There is a need to identify hotspots of risk to prioritise mitigation measures, as MP concentrations are expected to increase in the future. Here, we examine the risk of floating MP in the ocean surface by integrating environmental MP concentrations with ecotoxicity data. We first quantified unacceptable levels of MP concentrations based on ecotoxicity data available in scientific literature. In parallel, we quantified past (1970), current (2010) and future (2050 and 2100) environmental concentrations of MP based on microplastic distribution. To draw conclusions about the past, current and future risk of MP at the ocean surface [; 5 m depth], we compared in situ MP concentrations with the corresponding unacceptable levels using a probabilistic approach. Effect data for 23 different species from eight phyla were included in our assessment: Arthropoda, Chordata, Cnidaria, Echinodermata, Haptophyta, Mollusca, Ochrophyta, and Rotifera. The resulting median unacceptable level was 1.21 \* 105 MP m-<sup>3</sup> (95% CI: 7.99 \* 103 MP m-<sup>3</sup> – 1.49 \* 106 MP m<sup>-3</sup>). We found strong indications that organisms in parts of the Mediterranean Sea and the Yellow Sea are currently at risk. By 2100, we expect that 68.7% and 53.9% of the Mediterranean Sea and the Yellow Sea will have unfavourable conditions for marine life due to MP pollution, under a worst case and in a business as usual scenarios, respectively. Our results showed substantial spatial differences in the risk of MP in the ocean surface layer (0 - 5 m depth). Global mapping of the MP risks is instrumental to identify marine regions that need increased attention for mitigation measures.

Keywords : Environmental risk assessment , Marine ecosystems , Microplastic , Multidecadal , Spatial distribution

#### Gaia's room LIVE at 18h30

#### Highlight from Plymouth. Everywhere: Life in a littered world.

Curated by Joanne Lee and Rosemary Shirley

Litter is everywhere. It is on country lanes and city streets, washed up in polar regions and deposited on mountains as particles present in 'plastic rain'. It can be so small it needs to be viewed through a microscope and so large that its true scale can only be understood through satellite imagery. It's the coffee cup in the hedge and the Pacific garbage patch.

This online exhibition presents works by international artists who explore everyday practices of littering and consider its effects. It asks: What might the future look like in our littered world? Are there ways that humans, animals and plants can co-exist with or even prosper amongst the rubbish? And what will it take to clean up our act?

Poster Session 23.7, 23.8 and 23.9

#### Session 23.7\_O. Chaired by Aquilino Miguelez, Lanzarote

**Microplastics in sinking particulate matter of the Inner Sea in the Maldives** *Fischer Elke, Pfeiffer Felix, Wiesner Martin.* Paper number 328234

**Plasticrusts and pyroplastic: two novel plastic debris types detected in Giglio Island, Italy** *Ellrich Julius A., Ehlers Sonja M..* Paper number 329513

Surface water microplastic pollution in marine waters of Latvia Barone Marta, Vecmane Elīna, Burdukovska Valentīna, Suhareva Natālija, Putna-Nīmane Ieva, Aigars Juris. Paper number 333281

#### **Microplastics in sinking particulate matter of the Inner Sea in the Maldives** Fischer Elke, Pfeiffer Felix, Wiesner Martin.

The Maldives with 1,190 small islands grouped in two chains of 26 atolls that surround the so-called Ïnner Seä. In the context of this stunning beauty, the question of waste management arises. According to UNICEF (2019), 280,000 plastic bottles are used and discarded daily in the capital Malé alone. Statistical data from the Maldives Customs Service documents that 325 million plastic bags were imported into the Maldives in the years 2006 - 2010 (Zuhair, 2011); imports increased dramatically to 104 million plastic bags in 2018 (UNICEF 2019). Due to this extensive consumption and the mismanagement of waste, even uninhabited islands of the archipelago are littered with plastic. Efforts to reduce this problem are ongoing. While large plastic objects on beaches and in the water are easy to spot, there is a growing need for knowledge about the microplastic contamination of the Inner Sea. Within the framework of the BMBF-funded project Maldivian Monsoon and Carbonate Platform Processes" (Institute of Geology of the University of Hamburg), two mooring arrays were deployed in the northern and southern part of the Inner Sea in 2014. Sediment traps were attached to the moorings at depths of 80 and 200 m and programmed to collect sinking particulate matter at 18-day intervals for one year. The time series covers the SW- and NE-monsoon seasons with strong fluctuations in precipitation, wind strength, aeolian input, surface and deep-water currents, primary productivity and inflow of water from the Arabian Sea and the Bay of Bengal. Accompanying, sub-samples of the trapped material will be analysed qualitatively and quantitatively for plastic particles in the laboratories of the MRC. Preliminary results show that microplastics are ubiquitously present in relevant concentrations. Besides the development of an adopted digestion method, type- and size-specific analyses of the particles using RAMAN spectroscopy are implemented.

#### View online submitted version

Keywords : carbon , Maldives , microplastic , sediment traps





#### Microplastics in marine suspended solids from sediment traps in the **Inner Sea of the Maldives**

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lamour

#### Introduction

- Studies on microplastic (MP) occurrences in the Maldives as well as studies on MP occurrences with the deployment of sediment traps is still very limited
- Previous studies in the Maldives focus on beach sediments, reef sediments and surface waters. For this reason, the present study represents the first study on MP occurrences in the waters of the Inner Sea from sediment traps
- By using moored sediment traps, a time component is integrated into the investigations and in general a better understanding of the vertical distribution of the MP occurrences is gained
- Samples from two mooring configurations with three sediment traps (N = northern deep trap 200 m below sea surface, Ns = northern shallow trap 80 m below sea surface, S = southern trap 200 m below sea surface) were investigated (see figure 1)
- By assuming ellipsoidal particle shapes and calculating the mass share of carbon per polymer type, both the polymer masses and the polymer-based carbon masses were determined
- Additionally, enabled by the use of sediment traps, flow rates or sedimentation rates of MPs and polymer-based carbon were obtained and related to the total flux of marine suspended solids (mss)

#### Results & Discussion

MP particle numbers, sizes and concentrations In 14 samples a total of one microplastic fiber (see figure 4a) and 2,365 microplastic

(left) and a schematic configuration (right).

- particles in quantities between 22 and 1015 particles per sample were found The particle shapes are dominated by irregularly shaped fragments and spherical
- structures (see figure 4a) The major dimension of all particles ranged from 20  $\mu m$  to 1,900  $\mu m.$  Of 2,365 particles only six have a major dimension >1 mm
- The relative frequency of particles increases as the major dimension decreases (see figure 4b). This tendency is also evident for each individual sediment trap. 98% of all counted particles have a major dimension between 20  $\mu m$  and 300  $\mu m$
- The particle concentrations range from 33 particles  $g^{-1}\,dry$  weight (dw) mss to 870 particles  $g^{-1}\,dw$  mss with a mean value of 394  $\pm$  255 (mean  $\pm$  standard deviation (sd)) particles  $g^{-1}\,dw$  mss (see figure 5 and figure 6a)
- With southwesterly current conditions at a depth of 200 m the particle concentrations in samples with the same sampling times (N3, S3, N5, S5, N12, S12, N16, S16) or short consecutive sampling times (N1, S2) are always higher in N than in S (see figure 5). There are significant differences (p < 0.05) between N and S regarding the particle concentrations with prevailing southwesterly currents



#### Distribution of polymer types

- Ten different polymer types were identified, with PS (72%) and PC (23%) being the most frequently found polymer types (see figure 5). Considering the density of thes polymers and the demand and/or the use of these polymers on the Maldives these findings are comprehensible
- The discrepancy of the plastic occurrences e.g. of the polyolefins PE and PP between the samples from the sediment traps of this work in comparison to the occurrence of the polyolefins in beach sediments, reef sediments and surface water near the investigation area of this work is very strong. This suggests that the effects of density change (through biofouling or ingestion and subsequent excretion in faecal pellets) and biological transport mechanisms through marine organisms are only partially effective or even counteractive. Additional factors such as vertical currents or local effects that have not yet been considered may also influence the buoyancy and might even lead to reemergence of plastic particles

#### Material and Methods

- 14 samples were examined, which together collected 252 days of ms
- The development of a laboratory analytical protocol for the digestion of biogenic matter resulted in the application of hydrochloric acid (HCl), hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>), sodium hypochlorite (NaClO) and phosphoric acid (H<sub>3</sub>PO<sub>4</sub>) in different concentrations and at different application times (see figure 3)
- chitinous material (see figure 2). The methods for digestion of chitin-containing structures used in MP studies so far (mainly chitinase) did not achieve the intended results
- Preliminary tests showed that 85% H<sub>3</sub>PO<sub>4</sub> is a suitable chemical for chitin destruction (see figure 2). LDPE, HDPE, PP, PS and PET are hardly affected by the application of 85% H<sub>3</sub>PO<sub>4</sub>. However, PA is dissolved
- For particle identification, the samples were stained with Nile red (dissolved in chloroform). After filtration of the stained samples, filters were photographed and particles were counted and measured (major and minor dimension) under the fluorescence microscope. A subset of the MPs was identified by  $\mu \text{Raman-spectroscopy}$  (see figure 3)
- The ratio of the minor and major dimension of all particles (n = 2,365) was calculated and yielded a median value of 0.56. The unknown third dimension (thickness) was calculated assuming the ratio of the thickness and the minor dimension of the particle to be the same as the ratio of the minor dimension and the major dimension of the particle. Thus, the thickness was estimated as 56% of the minor dimension





nosaic images of a prelimina h 85% H-PO,. Chitin-sickle st



me of the applied laboratory protocol for the processing of marine samples fro

the final sedimentation levels of this work) mass per square meter and sample for MP- total polymer-based carbon mass per samp polymer-based carbon content based on th total carbon flux per sample for MP particle polymer-based carbon mass per square me	, the da particle ole per i le initia es <1 m eter for	ily MP   es <1 mr nitial sa I carbor m and t MP part	oarticle so n (only fo mple we weight he daily s cicles <1 n	edimen or N and ight, th or the in edimen nm (onl	tati IS), e nitia nteo ly fo
	Min	Max	Median	Mean	- 8
Total polymer mass per sample per initial sample weight (all sizes) (ng mg <sup>a</sup> )	6.1	268.6	39.5	75.5	
Average MP particle mass per sample (all sizes) (ng)	40.7	479.8	126.2	182.3	33
Average MP particle mass per sample (particles <1 mm) (ag)	40.7	479.8	94.7	137.4	
MP particle sedimentation rate per sample (particles <1 mm) (m $^{\circ}$ d $^{\circ}$ ) (only N and S)	19.6	156.0	64.0	66.9	4
MP Particle sedimentation weight per sample (particles <1 mm) ( $\mu gm^2d^3$ (only N and S)	1.1	30.8	6.8	9.4	1
Total polymer-based carbon mass per sample per initial sample weight (all sizes) (ng mg*)	5.1	245.0	34.9	68.6	ż
Polymer-based carbon content of initial carbon weight per sample or initial total carbon flux per sample (particles <1 mm) [%]	0.01	0.22	0.05	0.07	*
Polymer-based carbon sedimentation weight per tample (particles <1 mm) (µg m² d²) (only N and S)	13	27.5	5.8	8.4	

#### Conclusion

- The application of 85% H<sub>3</sub>PO<sub>4</sub> for the destruction of chitin represents a possible alternative to enzymatic digestion with chitinase or lysozyme for future MP studies with chitincontaining sample material
- A combination of current conditions, location of the sediment traps and the island morphology may influence the particle concentrations per sample
- Most likely, the MP occurrence is made up of plastic from the Maldives itself and plastic that is introduced into the Inner Sea from further away by currents
- For a better understanding of the spatial and temporal distribution of MP, the dynamic behaviour of MP, the polymer type distribution or the influence of e.g. currents or effects like biofouling, sediment traps are best suited and therefore highly recommended for increased application. Permanent installations with an annual evaluation rhythm as well as the establishment of the MP particle mass as an additional, reasonable and target-oriented measure for MP studies could provide interesting insights

Contact: Felix Pfeiffer, CEN-MRC, University of Hamburg, felix.pfeiffer@studium.uni-hamburg.de

Mooring Configuration Configuration ADV-S nfiguratio ig. 1: Positions of the mooring configurations Jorth and South in the Inner Sea of the Maldives

c layout of a mooring

- The samples from the Maldives contained a lot of



- Fig. 5: Microplastic particles gr<sup>1</sup> dry weight mss per sample with the extrapolated polymer type shares and the current conditions prevailing at the sampling period in the depths of the corresponding sediment trap. N = North, NE = Northest, TW = Northwest, S = South, SE
- The assumption of ellipsoidal MP particle shapes proved to be valid
- In mean 67  $\pm$  44 MP particles m<sup>-2</sup> sediment daily (see table 1)
- In a simplified extrapolation, the MP sedimentation rates for Maldivian waters result in a daily sedimentation of about 860 kg MP <1 mm (see figure 6b)
- differences between the traps regarding MP particle concentration were found, but no significant differences between the traps regarding the total polymer mass per sample per initial sample weight could be found. This shows that interpretations may differ depending on the selected measure. Future studies should consider both particle number and particle mass
- polymer-based, i.e. originates from the MP (see table 1)



v mss per sedimen (m²) around the Ma e statistic. b: Theoretical rectangular sea a xtrapolated and simplified results of abou

- Flux rates and mass-specific results
- When comparing N and S at prevailing southwest currents, significant
- In median 0.05% (mean 0.07  $\pm$  0.07%) of the total carbon flux of mss <1 mm is

#### Plasticrusts and pyroplastic: two novel plastic debris types detected in Giglio Island, Italy

Ellrich Julius A., Ehlers Sonja M..

Plasticrusts and pyroplastic are novel plastic debris types that have only recently been reported for the first time from Madeira island (NE Atlantic Ocean) and the southern United Kingdom, respectively. While plasticrusts seemingly result from plastic debris being wave-swept across rugose rocks, pyroplastics derive from burnt plastic waste. During field surveys, we detected plasticrusts on a wave-exposed rocky shore and pyroplastic on a wave-sheltered sandy beach in Giglio island, Tyrrhenian Sea, Italy. At the lab, we identified the plasticrust material as polyethylene (PE) and the pyroplastic material as polyethylene terephthalate (PET) using state-of-the-art Fourier-transform infrared (FTIR) spectroscopy. These polymers are widely used in everyday products and, therefore, heavily contribute to plastic pollution in the Tyrrhenian Sea and other aquatic and terrestrial ecosystems worldwide. While plasticrusts might be ingested by co-occurring grazers (such as intertidal snails and crabs), pyroplastic can release toxic substances (e.g., lead) that are potentially harmful to plants, animals and humans, equally. Therefore, the recent accumulation of plasticrust and pyroplastic reports along European shorelines is concerning. Thus, we propose that plasticrust and pyroplastic occurrence should be monitored and that the effects of these novel debris types on marine and terrestrial organisms should be examined.

Keywords : Marine waste , Mediterranean , rocky intertidal , sandy beach , Tyrrhenian Sea

#### Plasticrusts and pyroplastic: two novel plastic debris types detected in Giglio island, Italy

#### Julius A. Ellrich<sup>1</sup> & Sonja M. Ehlers<sup>2,3</sup>

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#### **INTRODUCTION:**

Plasticrusts and pyroplastic are novel plastic debris types that have only recently been reported for the first time from Madeira island (Atlantic Ocean, Gestoso et al. 2019) and the southern United Kingdom, respectively (Turner et al. 2019). While plasticrusts result from plastic debris being wave-swept across rugose rocks, pyroplastics derive from burnt plastic waste. During field surveys in Giglio island (Tyrrhenian Sea, Italy), we detected plasticrusts on a wave-exposed rocky shore and pyroplastic on a wave-sheltered sandy beach (Ehlers & Ellrich 2020, Fig. 1).





Fig. 1 A) Location of Giglio island in the Tyrrhenian Sea. B) Locations of the five surveyed habitats (H1-H5) in Giglio. H2 and H3 were adjacent habitats and, thus, depicted by a single dot. C) The wave-exposed rocky shore (H1). D) The wave-sheltered beach (H2).

#### **METHODS:**

In the field, we measured plasticrust density, area and percent cover using quadrats (10 cm x 10 cm). At the lab, we determined plasticrust thickness using a digital microscope and pyroplastic size using digital calipers. Furthermore, we identified the plasticrust and pyroplastic materials (i.e., polymer types) with Fourier-transform infrared spectroscopy (FTIR, Fig. 2). We conducted our FTIR measurements in attenuated total reflectance (ATR) mode in a wavenumber range between 4000 and 370 cm<sup>-1</sup> with 8 co-added scans and a spectral resolution of 4 cm<sup>-1</sup>. Then, we compared the obtained spectra with the Bruker spectral library in Opus 7.5 software.



**Fig. 2** Sonja M. Ehlers identifying polymer types using FTIR (Vertex 70, Bruker, Ettlingen, Germany) at the Federal Institute of Hydrology in Koblenz, Germany.

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#### MICRO2020 INTERNATIONAL CONFERENCE



#### RESULTS:

**Plasticrust** (Fig. 3A) density was  $3.25 \pm 1.65$  plasticrusts/dm<sup>2</sup> (mean  $\pm$  SE; n= 4 quadrats). Plasticrust area was  $0.46 \pm 0.08$  mm<sup>2</sup> (n= 13 plasticrusts). Plasticrust cover was  $0.02 \pm 0.01$  % (n= 4 quadrats). Plasticrust thickness ranged between 0.5 and 0.7 mm. FTIR analyses revealed the plasticrust material as polyethylene (PE, Fig. 3B). We did not detect any plasticrusts on the wave-sheltered rocky shores (H3-H5, Fig. 1B).



Fig. 3 A) The detected blue plasticrusts consisted of (B) PE material.

The **pyroplastic** had a stone-like appearance (Fig. 4A) with blue inclusions (Fig. 4B). Pyroplastic size was approximately 2 cm  $\times$  1.4 cm  $\times$  0.5 cm (length x width x height). FTIR analyses showed that the pyroplastic material was polyethylene terephthalate (PET, Fig. 4C). On the beach, we detected burnt charcoal near the pyroplastic. Finally, we found several PET beverage bottles (Fig. 4D, E) in each habitat (H1-H5, Fig. 1B).



**Fig. 4 A)** The detected pyroplastic with blue inclusions. **B)** Pyroplastic close-up. **C)** The FTIR spectrum that revealed PET as the pyroplastic material. **D)** PET bottle found on the beach (H2, Fig. 1B). **E)** The FTIR spectrum that confirmed PET as the bottle material.

#### **DISCUSSION:**

Our results from Giglio resemble findings from Madeira and the United Kingdom indicating that plasticrusts and pyroplastics are not local phenomena. Plasticrusts are generated by sea waves washing plastic debris across rugose rock (Gestoso et al. 2019) which suggests that wave exposure may influence plasticrust generation. This notion is supported by the fact that we detected the plasticrusts on a waveexposed rocky shore (H1) but did not detect any plasticrusts on wave-sheltered rocky shores (H3-H5, Fig. 1). Since the plasticrusts were roughly within the microplastic (MP) size range (i.e., particles < 5 mm) and as invertebrates, such as grazing snails, readily consume MPs (Ehlers et al. 2020), it would be interesting to investigate wether invertebrates ingest MPs off the plasticrusts to examine this potential MP pathway into the foodweb. Finally, our findings of PET pyroplastic, PET beverage bottles and burnt charcoal on the beach (H2, Fig. 1) suggest that the pyroplastic resulted from a campfire or waste incineration fire. Since plasticrusts and pyroplastic might harm organisms through MP ingestion and toxic substance release, respectively (Gestoso et al. 2019, Turner er al. 2019), we recommend that future studies should monitor the distribution and abundance of these novel plastic debris types and examine the ecotoxicity of these potentially hazardous materials.

#### 

We thank Professor Dr Jochen H. E. Koop for letting us use the FTIR spectrometer at the Federal Institute of Hydrology in Koblenz, Germany.

#### Surface water microplastic pollution in marine waters of Latvia

Barone Marta, Vecmane Elīna, Burdukovska Valentīna, Suhareva Natālija, Putna-Nīmane Ieva, Aigars Juris.

Microplastic pollution in the marine environment is a globally growing concern. Monitoring spatial distribution of microplastic concentrations, type, size and chemical composition may help to identify sources and entry pathways. Such information has crucial role in initiating focused mitigation. This study investigates microplastic pollution in marine surface waters of Latvia in order to understand the dynamics involved in microplastic spatial distribution. Surface water microplastic samples were collected in 45 transects during time period from May 2018 to September 2018 using Manta net (300 µm) that was attached to the side of the vessel in approximately 7 meter distance and trawled for 1 hour at speed of approximately 2 knots. Biological matter was digested using 10% sodium hydroxide, 15% hydrogen peroxide and enzymatic treatment. Next, samples were filtered on GF filter and visual examination was done under the microscope Leica DM400 B LED with camera DFC 295. Particles were analysed for their colour and size using image analysis, and each detected particle was affiliated to one of the six categories (fragment, filament, sphere, film, granule, foam). Identification of particle's chemical structure was done by Attenuated Total Reflection Fouriertransform infrared spectroscopy method using Thermo Fisher Scientific Nicolet iSO20 spectrometer. Microplastics were detected in all samples with concentrations ranging between 0.08 particles/m3 to 2.54 particles/m3. The greatest concentrations of particles were observed near costal recreational sites and river estuaries. Most of the particles belong to filament (53%) and fragment (41%) categories. From 3553 particles chosen for analysis of chemical structure 93% were plastic polymers. The most prevalent polymer types were polyethylene (68%) and polypropylene (13%). Research has been conducted with the support of Ministry of Environmental Protection and Regional Development Project No. IL/106/2017 "Improvement of knowledge of the state of the marine environment in the marine waters under the jurisdiction of Latvia"

Keywords : Baltic Sea , marine water , microplastic , polymers , spatial distribution , surface water



#### Composition and spatial distribution of microplastic in marine waters of Latvia

Barone, M.<sup>1,2</sup>, Vecmane, E.<sup>1,2</sup>, Burdukovska, V.<sup>1</sup>, Suhareva, N.<sup>1</sup>, Putna-Nīmane, I.<sup>1</sup>, Aigars, J.<sup>1</sup>

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METHODO

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#### **BACKGROUND & AIM**

Microplastic pollution in the marine environment is a globally growing concern. Monitoring spatial distribution of microplastic concentrations, type, size and chemical composition may help to identify sources and entry pathways. Such information has crucial role in initiating focused mitigation. This study investigates microplastic pollution in marine surface waters of Latvia as a first step to understand the dynamics involved in microplastic spatial distribution and chemical composition as well as to improve and optimise sample treatment process.

#### **STUDY AREA**

Samples were collected in the marine waters of Latvia – open sea and semi-closed Gulf of Riga (Figure 1).

The sampling sites were selected to represent coastal as well as open waters, with particular attention to ports.



#### **RESULTS: spatial distribution**

- Results show the presence of microplastics in all samples (from 0.08 to 2.54 particles/m³)
- Open sea part has lower microplastic concentration (0.08-1.11, average 0.42 particles/m<sup>3</sup>) than semi-closed Gulf of Riga (0.11-2.54, average 0.67 particles/m<sup>3</sup>)
- Highest particle concentration was recorded at the Southern part of Gulf of Riga and might be caused by inflowing Daugava river water and costal currents
- Spatial distribution of microplastic abundances were highly variable among investigated sites
   Graphs for snated detrif

#### **RESULTS: type**

Detected particles were affiliated to one of the six categories (Figure 2)



From total amount of particles

#### Most common types were:

• Filaments (55.86%)

Least common types were:Beads (1.22%)

- aments (55.86%)
- Plastic fragments (37.66%)Film particles (4.91%)
  - Foam (0.30%)Pellets (0.04%)
  - 1 ellets (0.0-

#### **RESULTS: size**

A tendency was observed for the abundance of particles to increase as the size of particles decreases (Figure 3)



Sampling specifics: net attached to the side of vessel and trawled for <b>1 hour at speed of 2 knots</b>			
nes			
Equipment: Leica DM400 B LED and camera DFC 295			
lm,			
orm			
020			

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#### SAMPLE TREATMENT METHOD DEVELOPMENT

To reduce the time necessary for sample analysis, **sample size was reduced using Folsom Plankton splitter** to determine the best aliquot size (Figure 4), and treatment process was improved (Figure 5) **experimentally assessing the best treatment steps** and performing quality control.



#### **RESULTS: plastic polymers**

- Dominant microplastic polymers were polyethylene compounds and degradation products (Figure 6)
- Relatively smaller group was polypropylene and polystyrene particles



#### CONCLUSION

- Abundance of microplastics and composition by type and chemical structure varies between sampling sites and the time of sampling.
- · Open sea part has lower microplastic concentration than Gulf of Riga.
- Improved sample treatment method significantly reduced time for visual analysis.
- Further research should be done for monitoring purposes by performing repeated sampling at the same sampling sites for several periods to assess the seasonal and spatial dynamics of microplastic.
- Abiotic factors such as water physical properties, currents, weather and others should be taken into account when analysing aquired data.

Research has been conducted with the support of Ministry of Environmental Protection and Regional Development project No. IL/106/2017 "Improvement of knowledge of the state of the marine environment in the marine waters under the jurisdiction of Latvia".

#### Session 23.7\_Me. Chaired by Yannick Lerat, La Trinité sur Mer

#### Integrated approach on the fate of microplastics towards healthy marine ecosystems (MicroplastiX)

Brandt Luca, Sardina Gaetano, Capuano Tonia, Monteiro Raqueline, Araujo Moacyr, Schwamborn Silvia, Neumann Leitão Sigrid, Schwamborn Ralf, Muller Carolin, Dudeck Tim, Ekau Werner, Frias Joao, Nash Roisin, O'connor Ian, Pedrotti Maria Luiza, Lombard Fabien, Montone Rosalinda, Fredou Flavia, Fredou Thierry, Justino Anne, Muniategui Soledad, Andrade Jose Manuel, Fernandez Veronica, Fischer Franziska, Fischer Dieter, Mincarone Michael, Lenoble Veronique, Mounier Stephane, Casotti Raffaella, Donnarumma Vicenzo, Mazzocchi Maria Grazia, Iudicone Daniele.

Paper number 333161

#### The characterization of biofilm formed on microplastics

*Rozman Ula, Kalčíková Gabriela.* Paper number 333429

**Keeping it sIMPLER: SensIng Marine Plastic Litter using Earth observation in River outflows** *Atwood Elizabeth C., Martinez-Vicente Victor, Cole Matthew, Lindeque Penelope K., Pham Thi Chin, Pham Van Hieu, Biermann Lauren, Mata Aser.* Paper number 334402

#### Integrated approach on the fate of microplastics towards healthy marine ecosystems (MicroplastiX)

Brandt Luca, Sardina Gaetano, Capuano Tonia, Monteiro Raqueline, Araujo Moacyr, Schwamborn Silvia, Neumann Leitão Sigrid, Schwamborn Ralf, Muller Carolin, Dudeck Tim, Ekau Werner, Frias Joao, Nash Roisin, O'connor Ian, Pedrotti Maria Luiza, Lombard Fabien, Montone Rosalinda, Fredou Flavia, Fredou Thierry, Justino Anne, Muniategui Soledad, Andrade Jose Manuel, Fernandez Veronica, Fischer Franziska, Fischer Dieter, Mincarone Michael, Lenoble Veronique, Mounier Stephane, Casotti Raffaella, Donnarumma Vicenzo, Mazzocchi Maria Grazia, Iudicone Daniele.

Among all anthropogenic materials ever manufactured, plastic is by far the most versatile, with a wide range of applications, from product packaging to medical equipment. Its worldwide exponential use alongside inefficient waste management have led to the accumulation of large and small plastic items in the environment. Once released in the environment, plastics undergoes weathering and biofouling processes that might contribute to the fragmentation of larger items into microplastics. MicroplastiX is a JPI Oceans international interdisciplinary research project aiming to improve the understanding of the degradation mechanisms that affect microplastics in the ocean. To overcome knowledge gaps on weathering, degradation, and fragmentation, MicroplastiX will implement a holistic approach that combines field data with laboratory experiments, while evaluating the interactions of plastic items with biota in the water column, both horizontally and vertically. Several case studies in different environmental matrices will be explored to assess abundance and distribution pathways in relation to depth and distance to the shore. Concerning fate and impacts of MPs in marine organisms, the intake, identification, quantification of MPs and plastic-associated chemicals in marine biota (e.g. fish and mollusks) will be assessed in several ecosystems, ecological habits, and temporal scales. The project will also explore biofilm growth and colonization rates through ad-hoc in-situ and laboratory experiments, to assess how MP buoyancy, environmental dynamics, and transported contaminants affect microorganisms growing on the plastic surface. The planned surveys, laboratory activities and analytical development will estimate MP ingestion and egestion in zooplankton and other model organisms. Based on the data collected, a set of multiscale numerical models will be developed to predict the fate and pathways of microplastics in the environment. This will contribute to better decision-making and help to identify potential critical matrices of plastic accumulation.

Keywords : biofouling , microplastics , modelling , weathering

# **OCENNS** MicroplastiX

# Integrated approach on the fate of microplastics towards healthy marine ecosystems











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# **Background and Rationale**

metals from the surrounding environment, potentially microplastics can sorb persistent organic pollutants and trace environment. Winds, oceanic currents and animals contribute to potentially increasing the number of fragments available in the decade has identified that solar radiation, abrasion and salinity mountain tops to benthic sediments. Research over the last environment, being found in all environmental matrices from Microplastics have been commonly reported in the marine increasing their toxicity and harmfulness. Therefore: them a ubiquitous problem. Also, it has been identified that contribute to the breakdown of plastic items into smaller pieces, the widely distribution of microplastics across the globe, making

# Microplastic fragments are ubiquitous and potentially harmful

Brazil. countries – Sweden, Germany, France, Italy, Spain, Ireland and of experience and expertise on the topic was established, from 7 international and interdisciplinary consortium with a wide range assess the problem magnitude, efficiently manage sources and environment that scientists need to bridge in order to effectively degradation and fragmentation of plastics in the marine reduce potential pathways into the ocean. As such, an Yet, there are knowledge gaps associated to the weathering,



degradation on plastic materials along with the consequent project which will investigate the effects of weathering and MicroplastiX is an international, multidisciplinary research fragmentation into smaller pieces known as microplastics (MPs)

distribution and dispersal on the marine environment, as well This project will also explore how these processes affect MP as effects on marine biota.

# Aims and Objectives

MPs dynamics collection and will feed into an advanced multi-scale modelling of Field and laboratory experiments will complement the in-situ data benthic) while deploying a comprehensive quality assurance scheme sediments (intertidal and benthic) and biota (pelagic, demersal and Mediterranean Sea by sampling water (surface and column) MicroplastiX aims to holistically assess MPs in the Atlantic Ocean and

 How microplastics are transported from the beaches and coastal How turbidity and stirring affect microplastic buoyancy; The main scientific questions related to this project are: collaborative monitor research distribution understanding hot-spots focusing relevant intertidal microplastics. Irish Sa environment abundances demersal MicroplastiX benthic coast biota multidisciplinary Brazil Batterns dot investigation System identify water column estimate habitats MPS pelagic modelling ecologic sampling surface waters Mediterranean Sea experiments degradation Canary Current Upwelling System pathways Atlantic Ocean weathering sediments

• What are the effects of Stokes drift, Langmuir circulation, and areas to the open ocean;

other, non-linear wind effects on the microplastic dispersion and distribution;

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horizontally and vertically; accumulation in the water column and/or on the seafloor, both How microplastics are affected by movement, entrainment and

bio aggregation and biofouling on the buoyancy of microplastic What are the effects and impacts of degradation, fragmentation, particles;

How do microplastics impact marine organisms;

software to simulate the microplastics physical and biochemical How can we develop, optimise and validate mathematical tools and processes in seawater

# Project overview

specifically designed to assess MPs abundance, distribution MicroplastiX is divided into five interrelated work packages specific set of analytical and technical protocols. Data collected in pathways, biofouling, ingestion and bioaccumulation, under a the project will inform multi-scale models, as described in the figure below:



# Reach out to us

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https://tinyurl.com/ResearchGateMicroplastiX







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Marine Institute

#### The characterization of biofilm formed on microplastics

Rozman Ula, Kalčíková Gabriela.

Microplastics (MPs) are a large group of plastic particles (1–1000 µm) and have been found literary everywhere around the world. When MPs enter the aquatic environment, different inorganic and organic substances start to adsorbed on the MPs surface, followed by the colonization of microorganisms. For biofilm characterization, the most popular methods are surface characterization by microscope and microbial community structure determination. However, many more methods can help characterize biofilm, but they have not been often used. In our study, polyethylene MPs (confirmed by FTIR), extracted from a facial scrub, were incubated in freshwater (Glinscica River) for 15 weeks ( $23 \pm 2$  °C, 125 rpm, light/dark 16h/8h). Every week, MPs were filtered and added to a new freshwater in order to ensure enough nutrients and microorganisms. The most visible and stable biofilm occurred after several weeks of incubation. Therefore, we evaluated changes in biofilm composition from week 13 to week 15. Within two weeks, the amount of biofilm on microplastics increased from  $34 \pm 8\%$  to  $43 \pm 6\%$ . An integral part of each biofilm are also extracellular polymer substances (EPS), which increased from  $0.29 \pm 0.05$  mg/gbiofilm to  $0.59 \pm 0.10$  mg/gbiofilm. Chlorophyll a content was  $0.18 \pm 0.03$  mg/gbiofilm and  $0.37 \pm 0.11$  mg/gbiofilm at week 13 and 15, respectively, which indicated that microalgae are likely to be present in the biofilm. Ureases are indispensable enzymes in numerous organisms, therefore the determination of urease activity indicate the activity of microorganisms in biofilm. Urease activity increased from  $0.28 \pm 0.01$ mghydrolysed N/gbiofilm to  $0.95 \pm 0.27$  mghydrolysed N/gbiofilm within two weeks. We also observed changes of MPs behaviour, the density of particles significantly increased with time, and they started to sink.

Keywords : biofilm , freshwaters , microbeads





#### The characterization of Biofilm formed on microplastics



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#### Introduction

MPs (1-1000  $\mu$ m) enter the aquatic environment; adsorption of different inorganic and organic substances, followed by colonization of various microorganisms – **biofouling** 

#### Materials & Methods

- PE MPs, extracted from a facial scrub
- Incubation of MPs in freshwater (Glinscica River) for 15 weeks (23 ± 2 °C, 125 rpm, light/dark cycle 16h/8h)
- Every week a new freshwater was added
- Week 13 and 15: biofilm characterization



#### Results

Table 1: Biofilm characteristics at week 13 and 15.

Method	Week 13	Week 15
Mass of biofilm per mass of MPsB (%)	34 ± 8	43 ± 6
EPS (mg/g <sub>biofilm</sub> )	0.29 ± 0.05	$0.59 \pm 0.10$
Chlorophyll a (mg/g <sub>biofilm</sub> )	0.18 ± 0.03	0.37 ± 0.11
Urease activity (mg $_{hydrolysed N}/g_{biofilm}$ )	$0.28 \pm 0.01$	0.95 ± 0.27



Figure 1: PE MPs (left) and PE MPs with biofilm (right).

#### Conclusions

- The amout of biofilm on microplastics increased within the two weeks, consequently the amount of EPS, chlorophyll *a* content and urease activity also increased.
- We observed **changes of MPs behaviour**; the density of particles significantly increased with time and they started to slowely sink and the end of the experiment, the majority of the particles settle to the bottom.

More about our research:



https://planterastics.fkkt.uni-lj.si/



#### Keeping it sIMPLER: SensIng Marine Plastic Litter using Earth observation in River outflows

Atwood Elizabeth C., Martinez-Vicente Victor, Cole Matthew, Lindeque Penelope K., Pham Thi Chin, Pham Van Hieu, Biermann Lauren, Mata Aser.

The SIMPLER project aims to develop and validate new Earth Observation (EO) approaches to quantify input of marine plastic debris from rivers into coastal waters, using the medium-sized river system in Danang, Vietnam, as a pilot case study. Our key aim is development of EO-based algorithms to quantify microplastic flux rates from the river. Rivers flowing into oceans have been identified as one of the largest land-based plastic debris inputs to marine plastic pollution budgets, with Southeast Asian rivers contributing an estimated two thirds input to the annual global marine plastic debris budget. To date, EO detection algorithms have focused primarily on floating macroplastic detection in optically simple water bodies, such as the open ocean and relatively clear coastal waters. River mouths, with their often highly turbid and optically-complex waters, have long proven difficult for remote sensing detection of water constituent concentrations. EO detection of microplastics poses additional challenges, including: 1) microplastic concentrations often being not sufficiently high to change the water surface optical reflectance signal, and 2) strong absorption of infrared light, required for positive identification of hydrocarbons, within the water's surface. These limitations suggest that the only potential successful path to an EO algorithm for river mouth microplastic quantification will be dependent on using a proxy water surface signal for detection. Theoretical basis for intended flux quantification algorithms will be presented, and current state-ofthe-art detection approaches discussed, including required assumptions and expected methodological limitations. As part of project activities, local stakeholders will be equipped with simplified techniques for plastic pollution measurement and reporting. Data generated by local monitoring agencies can be used for satellite validation into the future, thus contributing to study sustainability beyond the scope of the current project. Algorithms will be designed with an eye towards scalability to other Southeast Asian river systems.

Keywords : remote sensing , river flux , satellite quantification , Vietnam

#### Keeping it SiMPLER: Sensing Marine Plastic Litter using Earth Observation in River Outflows

Elizabeth C. Atwood, Victor Martinez-Vicente, Matthew Cole, Penelope K. Lindeque, Pham Thị Chin, Pham Van Hieu, Lauren Biermann, Aser Mata liat@pml.ac.uk

#### Aim

Develop and validate new Earth Observation (EO) approaches quantifying river input of marine plastic debris into coastal waters, specifically macro- and microplastic flux rates over time. Rivers flowing into oceans have been identified as one of the largest land-based plastic debris inputs to marine plastic pollution budgets, with Southeast Asian rivers contributing an estimated two thirds input to the annual global marine plastic debris budget. Project study area is the medium-sized river system in Da Nang, Vietnam, but developed algorithms will be designed with an eye towards scalability to other Southeast Asian river systems.



#### Theoretical basis for intended flux quantification algorithm

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MP = (-0.034 ± 0.15608) + (0.044 ± 0.01716)\*SPN

ehl *et al.* 2020

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#### Challenges

EO detection algorithms focus primarily on floating macroplastic detection in optically simple water bodies, such as the open ocean and relatively clear coastal waters. River mouths are characterized by often highly turbid and optically-complex waters, thus posing additional difficulties for remote sensing detection of water constituent concentrations. EO detection of microplastics poses additional challenges, including:

- microplastic concentrations often not sufficiently high to change water surface optical reflectance signal
- strong absorption of infrared light, required for positive identification of hydrocarbons, within the water's surface

This suggests that most promising path to an EO algorithm for river mouth microplastic quantification will be dependent on using a proxy water surface signal, such as Suspended Particulate Matter (SPM) for detection.

In situ sampling has been delayed due to COVID-19 travel restriction, all results are thus purely theoretical thus far.

#### Collaborations working in parallel on the Da Nang river system





Sentinel-2

#### Next steps

In situ sampling plans have been significantly changed due to the COVID-19 pandemic, with onsite field campaign activities currently rescheduled for 2021. Simplified data collection activities with local partners, sampling for both macro- and microplastics as well as water quality parameters, will allow for greater temporal coverage such as seasonal variability. Model parameterization will be further expanded once data become available, thus allowing for improved uncertainty estimates in modelled river flux rates. Macroplastic flux rate calculation was not covered in this poster but will in part utilize methods laid out in Biermann *et al.* (2020).

#### Stakeholder engagement

Local stakeholders will be equipped with simplified techniques for plastic pollution measurement and reporting. Data generated by local monitoring agencies can be used for satellite validation into the future, thus contributing to study sustainability beyond the scope of the current project.

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#### Session 23.7\_Ma. Chaired by Irene Brandts, Barcelona

**Oxidative stress and inflammatory response in gut of** *Sparus aurata* **exposed to microplastics** *Solomando Martí Antònia, Capó Xavier, Alomar Carme, Alvarez Elvira, Ferrer Montserrat, Pinya Samuel, Deudero Salud, Sureda Antoni.* Paper number 334186

Effects of microplastics on head-kidney gene expression and biochemical biomarkers in adult zebrafish

Limonta Giacomo, Mancia Annalaura, Abelli Luigi, Fossi Maria Cristina, Caliani Ilaria, Panti Cristina. Paper number 334285

**Is aquaculture production of bivalves affected by microplastic contamination?** *Silva D. C. C., Marques J. C., Gonçalves A. M. M.* Paper number 334458

#### Oxidative stress and inflammatory response in gut of *Sparus aurata* exposed to microplastics

Solomando Martí Antònia, Capó Xavier, Alomar Carme, Alvarez Elvira, Ferrer Montserrat, Pinya Samuel, Deudero Salud, Sureda Antoni.

The presence of plastic in oceans is extremely worrying because they possess potential threats for marine organisms including plastic entanglement and ingestion, amongst others. Moreover microplastics (MPs) have become an emerging contaminant causing widespread concern due to its potential toxic effects associated to Persistent Organic Pollutants and other contaminants added during their manufacturing processes or sorbed to their surface once in the marine environment. However, while the number of studies documenting the ingestion of MPs by fish has increased, fewer studies have addressed the toxicological effects derived from the ingestion of MPS in longterm laboratory conditions. The aim of this study was to evaluate the physiological response of gilthead seabream (Sparus aurata) exposed to low-density polyethylene (LDPE) plastic during 90 days followed by an additional 30 days of depuration through the application of oxidative stress biomarkers in the gut. Two different treatments were applied by means of diet and three replicate tanks were randomly assigned to each treatment. Fish were exposed to a control diet without MPs and a treatment diet enriched with 10% LDPE MPs. To analyse MPs ingestion effect on the oxidative stress, inflammatory response and antioxidant and detoxification system, catalase, superoxide dismutase, glutathione S-transferase, myeloperoxidase activities, and malondialdehyde and protein carbonyl levels were determined in 81 gut samples. The results showed that compared with controls, the activities of all enzymatic biomarkers significantly increased after 90 days of exposure to enriched diet with LDPE. After the detoxification period, all biomarkers levels recovered to the initial activities, as in the time 0. As a conclusion, MPs exposure during 90 days on gilthead seabream affected the physiological response through antioxidant and detoxification enzymes activation and oxidative damage in gut, yielding novel insights into the consequences of MPs exposure of this cultivated and commercial species.

Keywords : Biomarkers , Gilthead seabream , Gut , Inflammatory response , Microplastics , Oxidative stress

#### Oxidative stress and inflammatory response in gut of *Sparus aurata* exposed to microplastics

Antònia Solomando, Xavier Capó, Carme Alomar, Elvira Álvarez, Montserrat Compa, Samuel Pinya, Salud Deudero, Antoni Sureda Research Group in Community Nutrition and Oxidative Stress (UIB), Interdisciplinary Ecology Group (UIB) and Centro Oceanográfico de Baleares (IEO), Palma de Mallorca, Spain

#### **INTRODUCTION**

**Microplastics (MPs)** pollution is widespread in marine ecosystems and a major threat to biodiversity. Given the ubiquity and small size of MPs, they can be ingested by a variety of aquatic organisms such as fish and accumulate in the gut. The toxicity of MPs in cells mainly arises from oxidative stress via the generation of an excess of reactive oxygen species (ROS), which can alter the redox balance, damage cellular components and induce an inflammatory response.





The 30-day detoxification period induced a decrease in antioxidant activities in the MPs treatment.

MDA



GSH activity increase over time with significant differences at T60 and T90 with respect to T0 in the MPs treatment and The values of the MPs treatment at T90 were significantly higher than those of the control group.

A clear increase of the lipid peroxidation and oxidized proteins – MDA and protein carbonyl derivates levels -, were observed after 90 days of MPs exposure. Carbonyl groups remained more stable than MDA. For all biomarkers the values of the MPs treatment at T90 were significantly

**Oxidative damage biomarkers** 

CONTROL

Carbonyl

Results are expressed as the mean ± s.e.m. Statistics: \$ indicates significant differences (p<0.05) between values of control group vs. MPs group; # indicates significant differences respect to T0; \* indicates significant differences respect to T90 MPs. (Two-way ANOVA analysis).

higher than those of the control group.

#### CONCLUSION

MPs exposure during 90 days on *S. aurata* affected the physiological response through induction of antioxidant and detoxification enzymes, pro-inflammatory response activation and oxidative damage in gut, which was recovered after a washout period of 30 days.



**Detoxification enzyme activity** 



GST activity increased significantly at T60 and T90 in the MPs treatment, and decrease in the depuration period recovering the initial values



MPO showed a clear increase indicating the activation of a pro-inflammatory response in the MPs treatment.

#### ACKNOWLEDGEMENTS

#### Effects of microplastics on head-kidney gene expression and biochemical biomarkers in adult zebrafish

Limonta Giacomo, Mancia Annalaura, Abelli Luigi, Fossi Maria Cristina, Caliani Ilaria, Panti Cristina.

Due to massive production, improper use and disposal of plastics, microplastics have become global environmental pollutants affecting both freshwater and marine ecosystems. Several studies have documented the uptake of microplastics in wild species and their biological effects, ranging from epithelial damage, inflammation, biochemical biomarkers response, and gene expression alteration. However, the biological effects of microplastics are not fully understood yet, especially the potential impact on immune functions. In this study, adult zebrafish have been exposed for twenty days to two concentrations of a mix of polystyrene and high-density polyethylene microplastics. Gene expression was evaluated in head-kidney, while biochemical biomarkers were evaluated in head and body homogenates. Acetylcholinesterase activity was slightly inhibited by microplastics exposure, while no significant effects on lactate dehydrogenase activity were observed. Microplastics exposure upregulated genes involved in sterol biosynthesis, xenobiotics metabolism, and adaptive immunity. The results presented in this study support the hypothesis that microplastics exposure could somehow affect acetylcholinesterase functioning and induce immune and metabolic responses at transcriptional level.

Keywords : acetylcholinesterase , biomarkers , Danio rerio , gene expression , immune system , microplastics

#### Effects of microplastics on head-kidney gene expression and biochemical biomarkers in adult zebrafish

Giacomo Limonta<sup>1</sup>, Annalaura Mancia<sup>2</sup>, Luigi Abelli<sup>2</sup>, Maria Cristina Fossi<sup>1</sup>, Cristina Panti<sup>1</sup> <sup>1</sup>Department of Physical Sciences, Earth and Environment, University of Siena, Siena, 53100, Italy. <sup>2</sup>Department of Life Sciences and Biotechnology, University of Ferrara, Ferrara, 44121, Italy. Email: limonta@student.unisi.it

#### Introduction & objective

Environmental contamination by microplastics (MPs) has been recorded worldwide, with highest reported concentration above 100 mg/m<sup>3</sup> within oceanic gyres (Goldstein et al., 2012). MPs have been isolated from various fish tissues, such as gastro-intestinal tract, liver, and gills, the reported toxicological effects suggest that MPs could lead cause tissue damage, inflammation, oxidative stress, metabolic alterations and affect acetylcholinesterase (AChE) activity (Fackelmann and Sommer, 2019).

The objective of this study was to test a set of molecular and enzymatic biomarkers. Eight genes (gene set 1) were selected from the results of a previous study conducted in our lab, which found differently expressed genes in zebrafsh liver after a similar MPs exposure. Four genes (Gene set 2) were selected for their known role in the immune system, for which head kidney is the primary organ in teleost. Moreover, acetylchlinesterase (AChE) and lactate dehydrogenase (LDH) activity were measured in zebrafish tissues.

#### **Materials & methods**

Experimental design: MP particles were administered daily for 21 days in association with food. The control group was given regular dry fish food, the two other groups were fed with control food spiked with a MPs mix (50% HD-PE and 50% PS). The water was changed daily before the feeding, and the tanks accurately rinsed to avoid MP accumulation over time.

Microplastics: PS and HD-PE irregularly shaped fragments were purchased from Toxemerge Pty Ltd as virgin microplastic powders (dimensional distribution:  $90\% < 90 \ \mu\text{m}$ ;  $50\% < 50 \ \mu\text{m}$ ;  $10\% < 25 \ \mu\text{m}$ ; 10μm).

Tissues sampling: All fish were euthanized through anesthetic overdose (MS-222 - 200 mg/L). The head-kidney was dissected and immediately submerged in RNAlater (10 µl/mg of tissue).

RNA extraction. cDNA synthesis and RT-gPCR: Head-kidneys from three fish from the same biological replicate were pooled together. Total RNA was extracted from 30 mg of tissue using the kit RNeasy plus mini (Qiagen). Reverse transcription and RT-oPCR were performed according to the method described in Limonta et al. (2019). The results were normalized based on the housekeeping genes: beta actin, ribosomal protein L8, and eukaryotic translation elongation factor 1.

Acetylcholinesterase and lactate dehydrogenae activity: AChE activity was quantified in the whole zebrafish head, according to the spectrofluorimetric method described in Casini et al. (2006). The LDH activity was measured in the whole body except the head, according to the method described by Menezes et al. (2006)

#### **Results & discussions**

#### Molecular biomarkers (Gene set 1)

- $\bullet$  The mRNA level for this gene was significantly higher in fish treated with 100  $\mu g/L$  of MPs in respect to the control (p=0.01, t-test).
- Cyp2p8 is a gene involved in the detoxification metabolism which activates in response to xenobiotics such as PAHs (Curtis et al. 2017). It's expression in relation to MPs contamination should be further investigated, to confirm whether it may be responsive to MPs exposure





#### Acetylcholinesterase

- •A significant 18 % inhibition was found after exposure to 1000  $\mu$ g/L of MPs, compared to the control (p<0.05 t-test).
- ·Low concentration of MPs may not be able to affect AChE activity, as reported in a study by Barboza et al. (2019) as well.



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Experimental setup. Adult zebrafish (n=36) were kept in filtered and dechlorinated water. Photoperiod:12h light:12h dark. Water temperature: 28 °C.

#### Molecular biomarkers (Gene set 2)

- Among the set of immune genes, treatment with 100 µg/L of MPs was able to up-regulate the t-cell receptor (tcra), compared to the control (p=0.02, t-test).
- T-cells receptors are responsible for recognizing antigens bound to either MHCI and MHCII molecules. It has been reported that MPs could cause gut dysbiosis (Qiao et al., 2019), and if pathogens are involved they may cause an activation of the adaptive immune response.





#### Lactate dehydrogenase

- The LDH activity results show no significant differences across the three groups (CTRL, LMPs, HMPs).
- Neverthless, some studies reported increase in LDH activity following MPs exposure, (Banaee et al., 2019).

#### Conclusions

Overall, these findings contribute to the available data regarding the biological effects of MPs on fish, and in particular on the head-kidney, an organ in which the effects of MPs are still rarely studied. The results indicate that MPs could affect genes involved in detoxification metabolism and immune system. In light of this we believe that the nature of the relationship between MPs and the immune system should be further investigated

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#### **Is aquaculture production of bivalves affected by microplastic contamination?** Silva D. C. C., Marques J. C., Gonçalves A. M. M..

Marine bivalves have a wide distribution and play vital roles regarding the ecosystem structure and functioning. Some of these species are highly appreciated by humans and have an important commercial value. To meet the ever-growing requirement for seafood resources, including marine bivalves, aquaculture production has expanded worldwide over the past few decades, and is now responsible for producing 92% of the total marine bivalves available for human consumption. In Portugal, marine bivalves are produced in aquacultures at several transitional waters systems, including the Ria de Aveiro coastal lagoon. Despite being ideal sites for bivalve production, these systems are highly impacted by a vast number of pollutants, including microplastics, that may be ingested or absorbed and cause negative impacts in the marine biota, influencing their growth, survival and biochemical composition. Regarding this major concern, it is crucial to understand if microplastics are in the tissues of marine bivalves that are being produced in aquacultures for human consumption, as well as in its surroundings, i.e. in the water and sediment surfaces. Therefore, this on-going study aims to address the occurrence and seasonal variation of microplastics in different tissues (muscle tissue, digestive gland, gills, and remaining visceral mass) of the marine commercial bivalve species Crassostrea gigas and Ruditapes philippinarum produced in a Portuguese aquaculture located in the Ria de Aveiro coastal lagoon and in the surrounding environmental matrices (water and sediment surfaces). The preliminary results reveal that the microplastics most abundant in the water surface of the aquaculture tanks are fibers, films and beads. It is expected, to find similar microplastics in the sediment surface of the aquaculture tanks, as well as in the tissues from the bivalves destined for human consumption.

Keywords : aquaculture , bivalves , Crassostrea gigas , human consumption , microplastics , Ruditapes philippinarum , sediment , water

#### Is aquaculture production of bivalves affected by microplastic contamination?

#### D. C. C. Silva<sup>1,1</sup>, J. C. Marques<sup>1</sup>, A. M. M. Gonçalves<sup>1,2</sup>

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**=** 100 -500 μm = 500 - 1000

1 - 2.5 mm

2.5 - 5 mm

Filamen

Film

Fragm

- Microbead

#### Introduction

Marine bivalves are an important group of invertebrates with a wide distribution that play important roles in Trophic webs and in several ecosystem processes [1]

Bivalves have a good nutritional value, and some species are highly consumed and appreciated by humans. The global production of marine bivalves has been increasing worldwide over the past few decades, mainly because of the expansion of the aquaculture production that represents 92% of the total production of these organisms [2].

Aquaculture production of marine bivalves occur mainly in extensive open areas placed in transitional waters systems, that can be highly impacted by a vast number of pollutants, including microplastics (MPs). These plastic particles can be found in different environmental matrices, such as water, sediment and biota. These particles can be ingested by marine bivalves, posing a threat to them, to the entire marine food chain, and ultimately to humans [3].

Regarding the major concern of microplastic pollution and its implications to seafood quality and human health, this study aims to understand the occurrence and seasonal variation of microplastics in different lissues (visceral mass, digestive system, gills, muscle) of the marine bivalves Pacific cupped oyster, *Crossostrea gigas* (Thunberg, 1793), and Japanese carpet shell, *Ruditopes philippinorum* (Adams & Reevel850), produced in a Portuguese aquaculture for human consumption, and in the surrounding environmental matrices (water and sediment surfaces)

#### Materials and Methods

Water, sediment and bivalve samples were collected in December of 2019 and July of 2020 in a bivalve aquaculture located in the Ria de Aveiro coastal lagoon (Portugal).

Two sampling stations (white dots in the figure the right) on: were defined according to where each bivalve species was produced



Water samples were pre-filtered through a set of sieves with different mesh sizes (S mm and 38 µm) and the microplastics were extracted by wet peroxide oxidation (30%  $H_2O_2$ ), density separation with ZnCl<sub>2</sub> and filtration [4].

Sediment samples were submitted to an oxidation treatment (10% H<sub>2</sub>O<sub>2</sub>) prior to density separation with  $ZnCl_2$  and filtration of microplastics [S]

Bivalve organisms were divided in the following tissues: visceral mass, digestive system, gills and muscle. Microplastics were extracted from bivalve tissues through digestion with 15%  $H_2O_2,$  followed by direct filtration [6,7].

The filters (0,45 µm) containing the recovered samples from water, sediment and bivalve lissues were visually inspected for microplastics by stereomicroscopy. The number of microplastics found in each sample and Their physical properties (type, size, color) were registered and analyzed.



#### Conclusions

Microplastic pollution is affecting the aquaculture production of C gigos and R philippinorum intended for human consumption, since MP particles, mostly fibers, fragments and films, were found in both environmental matrices (water and sediments) and in the different tissues of the two bivalve species analyzed. No significant seasonal variation of MP abundance was observed in water, sediment or bivalve samples. MP abundances recorded for both environmental matrices were not significantly different from one another. In C. gigos the main lissues where MPs were found are the digestive system, gills and muscle, while in R. philippinorum are the digestive system, gills, muscle and foot, R. philopingrum showed a significantly higher MP abundance than C. gigos, which could translate to a higher risk for human health in case of consumption of contaminated bivalves

#### **Future Work**

The next steps of this onoping research will be the identification of the plastic polymers of the MPs found by Micro-Fourier Transformed Infrared spectroscopy (u-FTIR), and the study of potential nutritional effects of MP acute exposure to the bivalve species by determining their biochemical composition (fatty acid, carbohydrate and protein contents)

#### Acknowledgments

This study was supported by Fundação para a Ciência e a Fecnologia (FC1) through the strategic projects UIUB/04292/2020, granted to MARE. Mar ne and Environmental Sciences Centre and UIUP/5001//2020 UIUB/5001//2020 granted to CESAM. Centre for Environmental and Marine Studies U.C.C. Sitva thanks to FC1 for the doctoral grant SFRH/BD/143686/2019 funded by National Funds and Community Funds through FSE. A. M. M. Gonçalves adknowledges University of Comitor for the contract (TDS-718-725).















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#### Session 23.8\_O. Chaired by Bethany Jorgensen, Ithaca

**The presence and significance of microplastics in surface water in the lower Hudson River Estuary 2016-2019** *Polanco Helen.* Paper number 333523

#### Arenas Blancas Beach (El Hierro, Canary Islands, Spain): A new hot spot of microplastic debris?

Hernández-Sánchez Cintia, González-Sálamo Javier, Villanova-Solano Cristina, Ben-Charki El-Mousati Naoual, Sevillano González Marta, Hernández-Borges Javier. Paper number 333791

#### A full circle: microplastics on the sea surface, in biota and on the seafloor in a marine protected area

Compa Montserrat, Marató Mercè, Ríos-Fuster Beatriz, Alomar Carme, Deudero Salud. Paper number 334399

#### The presence and significance of microplastics in surface water in the lower Hudson River Estuary 2016-2019

Polanco Helen.

Microplastics are a major environmental issue of concern that affects all waterways. However, limited research has been conducted to determine the severity of microplastics specifically in the Lower Hudson River Estuary (LHRE). Since 2016, Hudson River Park has collaborated with Brooklyn College to survey microplastics within Park waters, between Chambers Street and 59th Street in Manhattan, New York. It was hypothesized that microplastic concentration is influenced by proximity to combined sewer outfall (CSO). The Park's microplastics study additionally aimed to understand how microplastic concentration is influenced by precipitation, and the semidiurnal tidal cycle. Samples were collected at channel and near-shore locations at both downtown and midtown sites. Microplastics were counted and categorized based on plastic type using a stereo microscope. Statistical analyses revealed that concentrations from 2018 varied significantly from all other years (p<0.009\*),that near-shore sites tend to exhibit higher concentrations than channel sites (p<0.03\*), and that fragments were highly prevalent in all samples (70%). Additional data will enhance the understanding of the presence of microplastics in the Lower Hudson and elucidate the effects of wet weather on plastic concentrations.

Keywords : Combined Sewer Outfall , Hudson River , Keywords: Microplastic , New York City

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# HUDSON RIVER PK The Presence and Significance of Microplastics in Surface Water in the Lower Hudson River Estuary: 2016-2019

Helen Polanco<sup>1</sup>, Siddhartha Hayes<sup>1</sup>, Carrie Roble<sup>1</sup>, Marika Krupitsky<sup>1</sup>, Brett Branco, PhD<sup>2,3</sup>; Hudson River Park Trust, New York, NY<sup>1</sup>; Aquatic Research and Environmental Assessment Center, Brooklyn College, Brooklyn, NY<sup>2</sup>; CUNY Graduate Center<sup>3</sup>







Park's downtown (1a) and midtown waters (1b). Red dots indicate Fig. 1a & 1b | Satellite images of trawling sites conducted monthly (June Combined Sewer Outfall (CSO) points. Oct.) at both channel and near shore locations within Hudson River

# INTRODUCTION

proximity to combined sewer outfall (CSO) (Fig. 1a & 1b). This ongoing Since 2016, Hudson River Park's Estuary Lab has collaborated with presence of microplastics in the Hudson Estuary and aimed to understand monitoring project has helped develop a baseline understanding of the microplastics, plastics <5mm in size, in the Park's Estuarine Sanctuary. It Brooklyn College to survey the concentration and distribution of semidiurnal tidal cycle. how microplastic concentration is influenced by precipitation, and the was hypothesized that microplastic concentration is influenced by

### METHODS

- samples within an attached 1L container. During each trawl, a 1m wide, 0.3mm mesh neuston net collected
- and 5.6 mm) and dried at 90°C overnight. Samples were filtered through a stacked series of sieves (0.3mm, 1mm
- Organic matter was degraded using wet peroxide oxidation; samples filtered out using a 0.3mm nitrex sieve. were divided using salt gradient density separation, and plastics were
- Plastics were counted and categorized, based on size and type, using 10x-40x magnification stereo microscopes





# RESULTS

- and near-shore concentrations in 2016 and 2017 (p<0.03\*) higher than both channel concentrations in 2016, 2017, and 2019 (p<0.002\* Significant difference was observed between near-shore (NS) and channel Tukey tests showing that 2018 near-shore concentrations were significantly (C) locations over all four years (ANOVA F(1,70) = 5.1, p<0.03\*)(Fig. 2), with
- Mean concentration in 2018 was three times greater than both 2016 (243,772) and 2019 (244,142), and six times greater than 2017 (143,204) (Fig. 3)
- 2017, and 2019. test p<0.009\*)(Fig. 3). No significant difference was found between 2016 2018 and every other year (ANOVA F (1,70) = 5.2, p<0.03\*; post hoc Tukey Significant differences in microplastic concentrations were observed between
- Fragments averaged 70% of all microplastic pieces observed across all years (Fig. 4).
- tound No significant difference in samples collected during ebb and flood tides was



Fig. 2 | Distribution of microplastic concentrations in samples collected at channel and nearshore sites 2016 - 2019



defined as June-October. versus mean seasonal rainfall in inches, 2016-2019. Sampling season is Fig. 3 | Percent change of mean seasonal microplastic concentration



Fig. 4 | Combined proportion of plastic types found from 2016 to 2019. Where total pieces of plastics = 46, 158.

## DISCUSSION

- is related to the water's proximity to shore and CSO locations The results of this study suggest that the presence of microplastics in the river
- plastics are not being disposed of properly and are ending up in local The prevalence of fragments in our microplastic samples indicates that larger
- Although no significant correlation between microplastic concentration and into smaller pieces due to UV and salt exposure (Fig. 4). waterways after CSO events, triggered by rain fall, and continue to degrade
- plastics could be related to the slight increase in rainfall in 2018. rain was found using spearman's r, a possible explanation for this increase in
- Chemical analysis using pyrolysis, gas chromatography- mass spectrometry (GC–MS) was conducted on archived samples from August and October of 2018 in a collaboration with NOAA, where pieces exhibited several of the
- The continued collection of samples and factoring in of tidal forces and and CSOs. Additionally, future microplastic pollution surveys, conducted by hydrodynamics could elucidate the relationship between microplastics, rain common chemical compositions of various plastics. other discharge reduction measures can assess the effectiveness of these Estuary Lab staff, following the 2019 styrofoam ban in New York City and

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- and sediments Laboratory methods for the analysis of microplastics in the marine environment: recommendations for quantifying synthetic particles in waters

# ACKNOWLEDGEMENTS

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Fragment Foam Pellet Line Film Nurdle

#### Arenas Blancas Beach (El Hierro, Canary Islands, Spain): A new hot spot of microplastic debris?

Hernández-Sánchez Cintia, González-Sálamo Javier, Villanova-Solano Cristina, Ben-Charki El-Mousati Naoual, Sevillano González Marta, Hernández-Borges Javier.

El Hierro is the southern and westernmost of the Canary Islands (Spain). With a population under 11,000 inhabitants during 2019 [1] and with the absence of industries, it is the first of the islands to cover more than half of its energetical needs through renewable energy. However, all these relevant issues do not make the island immune to the microplastic problem. In the Canary Islands, the presence of microplastics has already been studied in Tenerife, Gran Canaria, Fuerteventura, Lanzarote and La Graciosa since 2014 [2–4]; however, until now there are no studies of the arrival of plastic litter to the most occidental islands of La Palma, La Gomera or El Hierro. The present work has studied the incidence and type of microplastics (1-5 mm) and mesoplastics (5-25 mm) that have reached Arenas Blancas beach, located in the northwest coast of El Hierro island (with northeast orientation), from October 2019 to March 2020 (10 sampling dates with 3 sampling points each). The amount of microplastic debris that arrive to the beach is comparable to those of the most contaminated of the archipelago. Moreover, the abundance of pellets found repeatedly during the samplings shows that this contamination does not come from the island or those nearby as a result of the lack of this type of industry. References 1 Instituto Canario de Estadística, Estadísticas de la Comunidad Autónoma de Canarias, http://www.gobiernodecanarias.org/istac/jaxi-istac/tabla.do, (accessed 10 Sep 2020). 2 A. Herrera, M. Asensio, I. Martínez, A. Santana, T. Packard and M. Gómez, Mar. Pollut. Bull., 2018, 129, 494–502. 3 C. Álvarez-Hernández, C. Cairós, J. López-Darias, E. Mazzetti, C. Hernández-Sánchez, J. González-Sálamo and J. Hernández-Borges, Mar. Pollut. Bull., 2019, 146, 26–32.

Keywords : beach , microplastic , pellets



ARENAS BLANCAS BEACH (EL HIERRO, CANARY ISLANDS, SPAIN): A **NEW HOT SPOT OF MICROPLASTIC DEBRIS?** 

Cintia Hernández-Sánchez<sup>1,2</sup>, Javier González-Sálamo<sup>2,3</sup>, Cristina Villanova-Solano<sup>3</sup>, Naoual Ben-Charki<sup>3</sup>,

Marta Sevillano-González<sup>3</sup>, and Javier Hernández-Borges<sup>2,3</sup>



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#### INTRODUCTION

El Hierro is the southern and westernmost of the Canary Islands (Spain). With a population under 11,000 inhabitants during 2019<sup>1</sup> and with the absence of industries, it is the first of the islands to cover more than half of its energetical needs through renewable energy. However, all these relevant issues do not make the island immune to the microplastic problem. In the Canary Islands, the presence of microplastics has already been studied in Tenerife, Gran Canaria, Fuerteventura, Lanzarote and La Graciosa since 2014<sup>2.3</sup>; however, until now there are no studies of the arrival of plastic litter to the most occidental islands of La Palma, La Gomera or El Hierro.

The present work has studied the incidence and type of microplastics (1-5 mm) and mesoplastics (5-25 mm) that have reached Arenas Blancas beach, located in the northwest coast of El Hierro island (with northeast orientation), from October 2019 to March 2020 (10 sampling dates with 3 sampling points each). The amount of microplastic debris that arrive to the beach is comparable to those of the most contaminated of the archipelago. Moreover, the abundance of pellets found repeatedly during the samplings shows that this contamination does not come from the island or those nearby as a result of the lack of this type of industry.

#### **EXPERIMENTAL**



> The sampling of Arenas Blancas beach revealed a mean concentration of 2573 items/m<sup>2</sup> of meso and microplastics during October 2019-May 2020, which is comparable to those of the most contaminated beaches in the Canary Islands, warning about a possible new hotspot of massive arrival of plastic particles

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👼 TFinnova

- > Most plastic particles found showed light and transparent colours (52 % of mesoplastics and 64 % of microplastics), followed by yellow microplastics (11 %) and blue mesoplastics (10 %).
- > Most mesoplastics were fragments, while the most abundant microplastics were pellets followed by fragments, which suggests that this contamination does not come from El Hierro island
- > IR analyses are being currently developed in order to determine the composition of the plastic particles found in Arenas Blancas beach.

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#### ACKNOWLEDGEMENTS

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MAC 2014-2020 Cooperación Territorial 🔐 MEDI Interreg 🛄
# A full circle: microplastics on the sea surface, in biota and on the seafloor in a marine protected area

Compa Montserrat, Marató Mercè, Ríos-Fuster Beatriz, Alomar Carme, Deudero Salud.

Marine diversity is currently being threatened by plastic pollution in coastal areas, and despite the restrictions of human activities in Marine Protected Areas, these areas are becoming sinks for plastic debris. Here we present preliminary results from the overlap of micro-plastic items on the sea surface through surface trawls, ingested in pelagic and benthic fish and benthic marine invertebrates and micro-plastics in nearshore sediments via sediment grabs in Cabrera Marine-Terrestrial National Park located in the Balearic Islands in Spain. Samples were collected at three sites during the 2019 summer campaign, integrating various degrees of restrictions from day/night anchoring to no navigation permitted. It is essential to the integration of samples within the same areas to identify potential overlap in the items in the different compartments. Preliminary results have identified the presence of microplastic items in all compartments of the study area: sea surface-biota-seafloor highlighting the depth of the current microplastic threat in marine ecosystems. These preliminary results have identified an overlap of items within their diet and on the seafloor and sea surface, an indication of intake through the passive ingestion of while feeding on seafloor sediments or those suspended in the first layer of the sea surface. Initial results show that despite the protection status and the restrictions enforced within MPA, plastic debris is a persistent impact.

Keywords : fish , invertebrates , Marine Protected areas , Plastic intake , plastic pollution

### A FULL CIRCLE: MICROPLASTICS ON THE SEA SURFACE, IN BIOTA AND ON THE SEAFLOOR IN A MARINE PROTECTED AREA IN THE BALEARIC ISLANDS, SPAIN



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Plastic pollution in coastal areas in a growing threat and despite the increased restrictions of human activities to protect marine diversity in Marine Protected Areas, these areas are becoming hotspots for plastic debris. Here we present preliminary results from the overlap of micro-plastic (MPs) items on the sea surface (through surface trawls) ingestion (through gut content analyses in pelagic and benthic fish and marine invertebrates) and in shallow coastal sediments (via sediment grabs) in Cabrera Marine-Terrestrial National Park located in the Balearic Islands in Spain (Figure 1). Samples were collected at four sites during the 2019 summer campaign, integrating various degrees of restrictions from day/night anchoring to no navigation permitted. It is essential to the integration of samples within the same areas to identify potential overlap in the items in the different compartments.



#### Methods

Biota

Three Compartments:

Sea

surface

Seafloor

sediments

#### Field Work:

At each of the four locations, three sea surface trawls were performed for an 15 minutes for a total of 12 net tows (Figure 3).

Species representative of pelagic and benthic species were selected. Two species of fish *Oblada melanura* (n=15) and *Serranus scriba* (n=15). Three species of benthic invertebrates were analyzed for the ingestion of microplastics, two species of sea urchins *Paracentrotus lividus* and *Arbacia lixula* (n=20) and sea cucumbers *Holothuria sp* (n=15).

Shallow coastal sediment samples were collected during scuba surveys at each location (n=4).



- No significant differences were found between fish the three locations for fish ingestión (Figure 4).
- The study location for Olla had the highest average ingesiton at 0.14  $\pm$  0.21 MPs/individual while the least abundant was in Santa Maria 0.02  $\pm$  0.02 MPs/individual.
- No significant differences were found between locations for invertebrates (KW, p > 0.05).
- For invertebrates, the study location for Es Port had the highest average ingesiton at 0.34+- 0.34 MPs/individual while the least abundant was in Santa Maria 0.10 ± 0.16 MPs/individual.
- Overall, ingestion in invertebrates (54%) was higher than in fish species (27%).
- Overall, MPs were present in all compartments in the MPA of Cabrera National Park indicating urgent monitoring and mitigation measures are required to further protect these areas from marine litter.

# Results & Discussion



#### Seafloor sediment:





Ifremer



Figure 3 Example of a net tow performed during the 2019 summer field work at the Cabrera Marine-Terrestrial National Park located in the Balearic Islands in Spain.

- MPs were present in all of the net tows (Figure 2).
- Estells was a hot spot area for MPs on the sea surface with an average of 2.8 ± 2.02 MPs/m<sup>2</sup> and while Santa Maria had the least amount with an average of 0.03 ± 0.007 MPs/m<sup>2</sup>
- Considering Estells is a hotspots for floating MPs, without considering this hotspot area, a KW analysis indicated the concentrations in Olla sere significantly higher than those found in Es Port and in Santa Maria.



- The sediment samples from each location were heterogenous (Figure 5).
- In Olla, MPs items were present in all size fractions. Olla and Estells had the highest fraction of MPs in the sediment samples.



### Session 23.8\_Me. Chaired by Eva Cardona, Menorca

#### **3PAC: A platform for plastic particles analyses & characterization**

Dehaut Alexandre, Doyen Périne, Grard Thierry, Souissi Sami, Monchy Sébastien, Amara Rachid, Duflos Guillaume. Paper number 333182

# Marine Microplastics: an educational module for the identification of microplastics in environment samples with Nile red

Wichels Antje, Kieserg Christina, Hart Lisa Denise. Paper number 333924

#### Paddle surfing for science on microplastic pollution

Uviedo Oriol, Higueras Sara, Ballesteros Maria, Curto Xavier, De Haan William P., Sanchez-Vidal Anna. Paper number 334119

#### **3PAC:** A platform for plastic particles analyses & characterization

Dehaut Alexandre, Doyen Périne, Grard Thierry, Souissi Sami, Monchy Sébastien, Amara Rachid, Duflos Guillaume.

In the recent years, research on microplastics (MP) and nanoplastics (NP) has largely developed with better insights on the way analyses have to be conducted. It is now clear that research in this topic necessitates huge investment in terms of expertise, infrastructure and materials in order to insure the best quality for results. In the framework of a French regional project, CPER Marco, different laboratories from Universities, CNRS and Anses decided to create a common platform (3PAC) in order to share knowledge, expertise, working methodologies and analysis devices. This platform presently gather different equipment: monitored laboratory aquaria for controlled exposition studies, a laminar flow cabinet to avoid contaminations, stereomicroscopes to characterize particles and a µRaman, a Py-GC/MS and a Py-GC/HRMS for the identification of polymers. In the future, acquisitions of new analytical equipment are planned including light scattering technology for NP characterization and µFT-IR for polymer identification. By now, expertise of the different partners allows to cover a large field of applications from both laboratory and field studies. Recently investigations have been carried out on sediment, organisms from the environment and their microbiote, but also caging of marine organisms, exposition of bivalves in controlled conditions. The ambition of 3PAC is to answer national and transnational call for projects, making available an ecosystem gathering expertise, technologies and infrastructures.

Keywords : analysis , analytical platform , field and laboratory studies , identification , microplastics



# **3PAC: A Platform for Plastic Particles** Analyses & Characterization

Alexandre DEHAUT ª, Périne DOYEN <sup>b</sup>, Thierry GRARD <sup>b</sup>, Sami SOUISSI <sup>c</sup>, Sébastien MONCHY <sup>d</sup>, Rachid AMARA<sup>d</sup> & Guillaume DUFLOS <sup>a</sup> \*Anses, Laboratoire de Sécurité des Aliments, Boulevard du Bassin Napoléon, 62200 Boulogne-sur-Mer, France \*Unix. Littoral Côte d'Opale, USC Anses, EA 7394 – ICV – Institut Charles Vioilleite, 62200 Boulogne-sur-Mer, France «Univ. Litte, CNRS, Univ. Littoral Côte d'Opale, UMR 8187, LOG, Laboratoire d'Océanologie et de Géosciences, 32 Avenue Foch, 62930 Wimereux, France

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# Laboratory facilities















Work with two models of marine organisms: copepods (Acartia tonsa, Pseudodiatomus sp., Eurytemora sp., etc.) and mussel (Mytilus edulis).

Work in aguaria in a monitored environment with controlled water and air supplies.

# Field studies facilities





#### Caging experiences:

Caging of marine organisms (bivalves, fish) Used for biomonitoring of specific places Control over the sampled area, possibility of having in situ metadata Compromise between field data and laboratory experiments





Field sampling possibilities close to laboratories:

Natural deposit of bivalves, large beaches, estuaries Access to sampling campaigns Establishment in one of the first French fishing port

# Ad hoc working place



Manipulations performed in a specific laboratory Use of laminar flow cabinet (HEPA 14 filters)

# Particles characterization



### Stereomicroscopy:

Observation and measure of particles Automatic counting of particles Estimation of the size/volume

# Identification of polymers





Non destructive approach Identification of small polymers (down to 5 µm)

### Py-GC/MS:

Analyses of single polymers or mixture Possible quantification of polymers No interference of colouring agents

### Py-GC/HRMS:

Possibility to detect nanopolymers Detection of additives and Hydrophobic organic compounds (HOCs)



3PAC platform is supported by the European Union (ERDF), the French State, the French Region Hauts-de-France and Ifremer, in the framework of the project CPER MARCO 2015-2020.



Investigate, evaluate, protect











# Marine Microplastics - an educational module for the identification of microplastics in environment samples with Nile red

Wichels Antje, Kieserg Christina, Hart Lisa Denise.

Since the 50s of the last Millennium the world production of plastics has increased massively and led to a continuous increase in the amount of plastic waste in the world's oceans. The plastic material is biologically inert, hardly subject to mineralization and fragmented in the environment. Plastic particles are continuously becoming smaller and more frequent (microplastics; MP). How the waste gets into the sea, how it is detected and discovered by researchers and what consequences it can have for living organisms are topics of an education program on marine litter implemented in the AWI school lab OPENSEA. We focus on independent hands-on experiments which are framed by several modules. The detection of marine litter as components of the current modules at the school lab. Our aim is to impart scientific knowledge and awareness through experimental work. We have implemented experiments to sample plastic waste of different size classes in the environment and characterize the plastic material (microscope, infrared spectrometry). Our new module is based on the fluorescent dye Nile red aiming to visualize microplastic particles in environmental samples. It offers a research-oriented approach and an independent research access applying a new, cost effective, and harmless method for testing and staining with Nile red dye for school labs. Environmental samples and defined plastic polymers are stained with Nile red to examine the polymer in more detail. When excited with blue light, the dye bound to the polymer emits in the vellow-red range. The fluorescent colour of the plastic piece depends on the chemical structure of the respective polymer. Depending on this method, learning modules and experiments for students were developed, and evaluated in the OPENSEA school lab. In addition to working materials for students and teachers, we provide background material, on the long run this module will be available online.

Keywords : awareness , experiment , high school students , nile , red , scientific knowledge

## Marine Microplastics - an educational module for the identification of microplastics in environmental samples with Nile red

Lisa Denise Hart<sup>1,2</sup>, Christina Kieserg<sup>1</sup> and Antje Wichels<sup>1\*</sup>

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Methods

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micro2020:333924

OPENSEA

Since the middle of the last century the world production of plastics has increased massively and led to a continuous increase in the amount of plastic waste. The environmental degradation of plastic waste through physical and chemical alterations creates continuous fragmentation of large plastic items into small ones known as microplastics (MP). How the waste gets into the sea, how it is detected and discovered by researchers and what consequences it can have for living organisms are topics of an education program on marine litter implemented in the AWI school lab OPENSEA. We focus on hands-on experiments which are framed by several modules. Our aim is to impart up-to-date scientific knowledge and awareness through scientific experimental work for school classes (age 16-19). We offer experiments to sample plastic waste of different sizes in the environment and characterize the plastic material (microscope, infrared spectrometry). A new module is based on the fluorescent dye Nile red aiming to visualize microplastic particles in environmental samples.

#### Visualization of fluorescent samples by using a "dark reader"

Environmental samples stained with the fluorescent dye Nile red are examined. When excited with blue light using the "dark reader", specific fluorescence can be visualized. To introduce the phenomenon and learn about fluorescence in general, we extracted Protoporphyrin IX from brown eggs and visualized auto-fluorescence (emission of red fluorescence) exemplarily. We also can make use of fluorescence for environmental science related questions. Using Nile Red, the dye bound to polymers emits yellow-red fluorescence. The Nile red fluorescence of stained plastic polymers depend on their chemical structure, some of them can be distinguished due to their emission colors. Learning modules and experiments for students were developed in the OPENSEA school lab, to convey how fluorescent staining can be used to analyze environmental samples (see detection of plastic polymers) contaminated with plastics.

### "Dark reader" Blue light transilluminator for fluorescent samples (excitation 440-510 nm)

- Amber screen emission filter (>570 nm)

#### Nile red

- Fluorescent dve
- Emission ca. 520-600 nm (in EtOH)
- Staining of hydrophobic components

#### Fluorescing eggshells



This experiment serves to convey the principle of auto-fluorescence and shows how this phenomenon is made visible. We developed an experiment with auto-fluorescent Protoporphyrin IX from brown egg shells. The students' task is to make this substance visible with the help of the following materials:

- Brown eggshells
- Ethyl acetate (Nail polish remover)
- Acetic acid (Vinegar 5 %)

alcohol and excited by a specific wavelength. For an introduction to the topic of fluorescence

Dark reader with an amber screen



Eggshell debris containing rest of Protoporphyrin IX

eggshells









First inspection of plastic particles





Fluorescent plastic particles stain

### and to illustrate the phenomenon, the students will receive additional material: Helpsheets with structural formulas of all important substances used in the experiment Atomic model to illustrate the principle of fluorescence

#### Detection of plastic polymers by using fluorescence

The properties of fluorescence can be used for environmental research. In order to transfer this to laboratory work, the students receive an environmental sample to work with. Various materials are used:

Nile red on a basis of Ethanol (20 % w/w)

Video about the principle of fluorescence

- Different plastic polymers •
- Artificial sample (mixed)
- Dark reader with an amber screen
- Compartment dryer (60 °C)



By staining with Nile red, plastic polymers can be partially differentiated due to their specific fluorescence. Different plastic polymers in a mixed sample can be distinguished after staining with Nile red, and excited with blue light.

#### Conclusion

This hands- on experimental module is divided into several small experiments aiming to identify plastic particles based on their specific fluorescence. Students work independent, individually or in a team on different scientific questions:

- 1. How to identify different plastic polymers? 2. Is the fluorescence of stained plastic polymers affected by color or shape of the MP?
- 3. Can Nile red staining be used to detect and identify different MP's in an artificial sample?

Finally, students will be able to evaluate the applicability of the method of Nile red staining in comparison to other application methods (microscope, infrared spectrometry).

Acknowledgement We are grateful to the Microplastic Working group of G. Gerdts at the AWI for scientific support and Marie Fischer for her highly appreciated contribution to the OPENSEA school lab Photographs: AWI school lab OPENSEA







"Fluorescing

Structural formula of Nile red

Artificial plastic sample before (A) and after (B) staining with Nile rea

#### Paddle surfing for science on microplastic pollution

Uviedo Oriol, Higueras Sara, Ballesteros Maria, Curto Xavier, De Haan William P., Sanchez-Vidal Anna.

Research on microplastics has rapidly expanded in recent years and has led to the discovery of vast amounts of microplastics floating offshore in all main oceanic gyres and including the Mediterranean Sea. However, there is a lack of information from a few meters from the coastline where the largest plastic mass flux is suspected to occur. The reason behind is the general use of manta trawls towed by boats or research vessels to obtain samples, which hinders nearshore sampling. We have designed a manta trawl to collect samples in the near-shore from any type of recreational sports floating gear like kayaks, sailboats, rowing boats, windsurf boards and others. Data generated is comparable to that obtained with traditional scientific equipment towed from boats. During one year, starting from October 2020, 10 social, environmental or sports associations along the NW Mediterranean coast will be acquiring scientific samples in the nearshore within the frame of two citizen science monitoring projects lead by the Spanish delegation of the non-governmental organization Surfrider Foundation Europe and the University of Barcelona. The projects "Paddle Surfing for Science" and "PlastiPlancton BCN" represent a paradigm shift in microplastic research, allowing to fill the gap in knowledge of this transition coastal area, and actively involving citizens in the generation of new monitoring data. This will allow to promote public understanding of science, overcome costly research sampling of the nearshore, and raise awareness on plastic pollution in the marine environment.

Keywords : citizen science , microplastics , paddle surf

# Paddle surfing for science on microplastic pollution

Oriol Uviedo<sup>1,2</sup>, Sara Higueras<sup>1,2</sup>, Maria Ballesteros<sup>2</sup>, Xavier Curto<sup>2</sup>, William P. de Haan<sup>1</sup>, Anna Sanchez-Vidal<sup>1</sup> <sup>1</sup> GRC Geociències Marines, Departament de Dinàmica de la Terra i de l'Oceà, Universitat de Barcelona, 08028 Barcelona, Spain. <sup>2</sup> Surfrider Foundation Europe, Spanish Delegation, 20012 Donostia, Spain.







#### Our science: laboratory work

Samples will be sieved and plastics carefully extracted under a stereo-microscope coupled with a DS-Fi2 camera. All extracted plastic particles will be weighted and photographed. ImageJ will be used to count and characterise particles (see poster by Liam de Haan at the MICRO2020 conference: «A method for accelerated processing of microplastic samples using a flatbed scanner and MPScanTool plugin for ImageJ». Furthermore, particles will be chemically identified using a Perkin Elmer Frontier Infrared Spectrometer (FT-IR) at the Scientific and Technological Centres of the University of Barcelona (CCITUB).

#### Our goal: monitor floating microplastics in the nearshore

Research on microplastics has rapidly expanded in recent years and has led to the discovery of vast amounts of microplastics floating offshore in all main oceanic gyres and including the Mediterranean Sea. However, there is a **lack of information from a few meters from the coastline** where the largest plastic mass flux is suspected to occur. The reason behind is the general use of manta trawls towed by boats or research vessels to obtain samples, which hinders nearshore sampling.

#### Our tools: the paddle trawl

We have designed a **manta trawl to collect samples in the near-shore** from any type of recreational sports floating gear like kayaks, sailboats, rowing boats, windsurf boards and others. Data generated is comparable to that obtained with traditional scientific equipment towed from boats.



Design of the lightweight low-cost paddle trawl to be towed from a paddle surf board (environment and climate-friendly and CO2 neutrol). The paddle trawl comprises a net with a mesh size of 330 µm and a mouth in one end and a collector bag at the other end, two floating wings sideward of the mouth made of low-density wood, a couple of clams just ahead of the collector bag, and a trawling rope and fixing shackles. Eventually a flowmeter can be attached. Details in Camins et al. (2020) (https://doi.org/10.1016/j.scitotenv.2019.136178).

#### Our citizen science project: Paddle Surfing for Science

During one year, starting from October 2020, **11 social, environmental or sports associations along the NW Mediterranean coast will be acquiring scientific samples in the nearshore** within the frame of two citizen science monitoring projects lead by the Spanish delegation of the non-governmental organization Surfrider Foundation Europe and the University of Barcelona. The projects "Paddle Surfing for Science" and "PlastiPlancton BCN" represent a paradigm shift in microplastic research, allowing to fill the gap in knowledge of this transition coastal area, and **actively involving citizens in the generation of new monitoring data**. This will allow to promote public understanding of science, overcome costly research sampling of the nearshore, and raise awareness on plastic pollution in the marine environment.



Map of the North Western Mediterranean with the locations monitored during 1 year by different associations. Each of them got the compromise to paddle for approximately 1 nautical mile, record the transect using the App Wikiloc (search #surfingforscience to see our tracksl) and send us the collector bag, every 15 days.

The COVID19 pandemic is seriously challenging the citizen science field work as the work is based on collaborative field work, not individual initiatives. Mobility in Catalonia is severely constrained as we are in weekend lockdown, but some of the associations are making strong efforts to go outside, paddle and contribute to the project, despite the challenging conditions. **The project moves forward!** 

#### **Preliminary findings**

Our results reveal that densities of floating plastics in the nearshore off Barcelona agglomeration are similar to those found offshore in the Catalan coast. However, we observe high variability due to eastern storm occurrence. We also observe that whereas floating microplastics dominate offshore, greater proportions of mesoplastics and macroplastics dominate at the nearshore waters, specially in between the breakwaters. Indeed, the breakwaters, that protect against wave action and coastal erosion, may behave as plastic traps. This is an indication of the importance of the nearshore as a source of plastic fragments to the open sea and calls for increased research in this area.









### Session 23.8\_Ma. Chaired by Juan Baztan, Crozon

**Cytotoxicity of Polystyrene microplastic particles in murine cell lines** *Völkl Matthias, Rudoph Julia, Jérôme Valérie, Scheibel Thomas, Freitag Ruth.* Paper number 334154

**LimnoPlast ITN: Chronic toxicity of microplastics in fish species** *Koenig Azora, Carney Almroth Bethanie, Sturve Joachim.* Paper number 334172

# Ingestion and effects of polyethylene terephthalate (PET) microplastics on two marine benthic invertebrates with different feeding strategies

De Felice Beatrice, Ferrario Cinzia, Gazzotti Stefano, Bacchetta Renato, Ortenzi Marco, Parolini Marco.

Paper number 334305

#### **Cytotoxicity of Polystyrene microplastic particles in murine cell lines**

Völkl Matthias, Rudoph Julia, Jérôme Valérie, Scheibel Thomas, Freitag Ruth.

The worldwide spread of microplastic and its potential hazard for the environment have been promoting scientific interests and public awareness. The increasing spreading of microplastic particles (MPPs) in the biosphere raises many questions on their (negative) effects on multicellular organisms. Currently, there is a common assumption that the entry of the nanostructures into vital biological systems could cause damage, which could subsequently cause harm to human health. However, the impact of MPPs (size  $\leq 5\mu m$ ) at the cellular level is mostly unknown. After enteral (i.e., through the gastro-intestinal) and/or pulmonary uptake. MPPs come in contact with the cells of organ lining tissues in particular the epithelial ones. From there, MPPs can be up taken by epithelial cells and / or penetrate more deeply into the tissues incidentally leading to a systemic distribution in the body. Macrophages, in their nature as scavenger cells, are very likely to get in contact with MPPs as well. Here, we studied potential effects of plain Polystyrene– MPP on cellular level. We used murine macrophages (ImKC, J774A.1) and epithelial (BNL CL.2; STC-1) cell lines. The particles size (0.2; 0.5; 1; 2; 3; 6 µm) as well as the particles concentration was varied systematically. Using confocal microscopy and scanning electron microscopy, we demonstrated a cell-dependent uptake of the MPPs. A putative cytotoxicity was analysed by standard MTT Assay. As further stress marker for cells, we quantified the intracellular quantity of reactive oxygen species after the MPPs uptake using a flow-cytometry-based detection assay.

Keywords : Cytotoxic effects , Epithelial cells , Macophages , murine cells , Uptake



# Cytotoxic effects of polystyrene particles on murine cells [334154]



<u>Matthias Völkl\*1</u>, Julia Rudolph\*2, Valérie Jérôme1, Thomas Scheibel2, Ruth Freitag1 \*Both authors contributed equally

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#### What kind of biological effects are induced by microplastic particles (MPP) in mammalian cells?

- Uptake and distribution
- Cellular effects (toxicity, inflammation) after MPP uptake
- Accumulation or excretion of particles after ingestion
- Long-term effects on cells



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 Image: Constraint of the same of th



The influence of **plain PS-MPP with varying size** on murine macrophages (J774A.1 / ImKC) and epithelial cells (STC-1 / BNL CL.2) was analysed. The **uptake** of MPP could be shown by confocal microscopy. Macrophages showed a high uptake frequency for all MPP-sizes (Figure 1). For epithelial cells (not shown) a lower and size dependent uptake frequency was found.

A standard cytotoxicity assay (MTT) showed, even for highest concentrations, no effects for epithelial cells. Very high concentrations (> 100  $\mu$ g/ml) showed a slight cytotoxicity for macrophages (Figure 2).

**Reactive oxygen species (ROS)** increased in a concentration dependent manner in ImKC cells. For the remaining cell lines no significant increase was found (Figure 3).



**Summary:** For untreated PS-MPP no significant short-time cytotoxicity on murine cell lines could be determined. In following project steps, single cell analysis and long-term investigations will be extensively analysed.



Deutsche



#### LimnoPlast ITN: Chronic toxicity of microplastics in fish species

Koenig Azora, Carney Almroth Bethanie, Sturve Joachim.

LimnoPlast ITN (Marie Skłodowska-Curie Actions, Horizon2020) addresses the issue of microplastic pollution in the environment from an interdisciplinary environmental, technical and social point of view to find new innovative solutions together. Whereas most research on microplastics is conducted on marine ecosystems, LimnoPlast focuses on freshwater systems. This contribution to the holistic project is the investigation of acute and chronic toxicity of microplastics in freshwater fish species. We will design studies that differentiate between polymer microparticles and the chemicals inherent to these materials. Freshwater fish species (perch, stickleback, and brown or rainbow trout) are selected by their ecological relevance and establishment as model organisms with available knowledge on their physiology and genetics. Different life stages, from larvae to adult, and fish cell cultures will be studied. In addition, we will address impacts in fish species exhibiting different feeding strategies. Both aspects are supposed to give a broader spectrum of real life conditions. The toxicity of microplastic polymers will be assessed with several endpoints on life cycle parameters (e.g. growth, reproduction), metabolism (including proteomics), general stress response and behavior. The toxicity of conventional and biodegradable microplastic polymers and their leachates will be tested in comparison.

Keywords : freshwater fish , LimnoPlast , microplastics , toxicity

# LimnoPlast ITN: Chronic toxicity of microplastics in fish species

#### Azora König, Joachim Sturve, Bethanie Carney Almroth

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The issue: Increasing plastic pollution and the degradation of these plastics to microplastics in the environment is an omnipresent problem. The endangering of marine ecosystems by polymer particles and fibers has caught the attention of both scientists and the public, whereas freshwater ecosystems require more research on this topic. This is the target of LimnoPlast ITN, an interdisciplinary approach on microplastics in freshwater systems.

Microplastics have been shown to harm organisms on several levels in a variety of studies<sup>2</sup>, yet the mechanisms and extent are still unknown. Further studies are required to determine the which combinations of factors are driving impacts (particles, polymers, chemicals).

The project: This research project is part of the ITN project LimnoPlast, funded by the European Union's Horizon 2020 research and innovation programme under the Marie Skłodowska Curie grant agreement no 860720.

Acute and chronic toxicity of several different polymers, which are biodegradable and/or biobased, will be studied. Particles ranging in size from 45 - 200 µm will be used in in vivo exposure experiments with several freshwater fish species. Focus will be placed on fish species endemic to Northern Europe, like rainbow trout, stickleback or perch. The fish will be exposed to microplastics via food pellets, trophic transfer and/or water/sediment exposures. We will place focus on end points with ecological relevance, and the fish will be exposed in adult and larval stages. Studies of toxicity will be based on behavioral endpoints, gene/protein expression, hormone levels, life cycle parameters and immune responses. In vitro assays may be used to elucidate toxicity of chemical extracts. Additionally, accumulation of microplastics in fish tissues and faeces will be determined, as there will be possible changes in the polymers as they pass through the gut.



The projected outcome: The fate and toxicity of chosen microplastics will be determined, and the impacts in life stages, exposure time and polymer/particle properties further elucidated.





This project has received funding from the European Union's Horizon 2020 research and innovation programme under the Marie Skłodowska-Curie grant agreement No 860720



### Ingestion and effects of polyethylene terephthalate (PET) microplastics on two marine benthic invertebrates with different feeding strategies

De Felice Beatrice, Ferrario Cinzia, Gazzotti Stefano, Bacchetta Renato, Ortenzi Marco, Parolini Marco.

The contamination of deep marine ecosystems is an issue of growing concern. Several studies showed how microplastics (MPs) with a density higher than seawater can sink and accumulate in marine bottom sediments. Polyethylene terephthalate MPs (PET-MPs) represent one of the most common plastic items sinking in deep marine ecosystems worldwide, but to date the information concerning the fate and toxicity towards benthic organisms is still scant. This study investigated the ingestion/egestion and the alteration of oxidative status (i.e., the amount of pro-oxidant molecules (ROS), the activity of antioxidant (SOD, CAT and GPx) and detoxifying (GST) enzymes, and lipid peroxidation) induced by the exposure to irregular shaped PET-MPs in two benthic organisms with different feeding strategies, a filter feeder, the Manila clam Ruditapes philippinarum and a grazer, the sea urchin Paracentrotus lividus. Seven day exposures where performed to administer to clams two concentrations of PET-MPs (0.125 and 12.5 µg/mL), while sea urchins were dietary exposed to three amount (8, 80 and 800 particles/g of food) of PET-MPs. Our results showed that both Manila clams and sea urchins were able to ingest and egest PET-MPs and that their transit throughout the digestive system did not cause tissue alterations to the digestive tract of both species. At biochemical level, PET-MPs induced an oxidative stress situation in gills, but not in the digestive gland, isolated from treated Manila clams. A slight overproduction of pro-oxidant molecules and a modulation of antioxidant enzymes were noted in the proximal part of sea urchin oesophagus. These results showed that the exposure to sinking, irregular shaped PET-MPs might induce the onset of an oxidative stress situation and represent a threat to marine benthic organisms regardless of their feeding strategies.

Keywords : benthic organisms , oxidative stress , polyethylene terephthalate



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AIM This study investigated the ingestion/egestion, the alteration of oxidative status and the possible tissue damages induced by the exposure to sinking irregular shaped polyethylene terephthalate microplastics (PET-MPs) in two benthic organisms with different feeding strategies: a filter feeder, the Manila clam *Ruditapes philippinarum* and a grazer, the sea urchin *Paracentrotus lividus*.



These results showed that the exposure to sinking irregular shaped PET-MPs might induce the onset of an oxidative stress situation and **represents a threat to marine benthic organisms** regardless of

RESULTS

**CONCLUSIONS** 

their feeding strategies.

MICR02020

INTERNATIONAL CONFERENCE

23-27 INVENIES 2020 LINCERTEIND BEYIND FATE AND IMPACTS OF MICROPLASTICS: KNOWLEDGE AND RESPONSEDULTIES

### Session 23.9\_O. Chaired by Adam Porter, Exeter

**Spatial distribution of plastic pollution in a protected coastal Mediterranean wetland** *Cesarini Giulia, Cera Alessandra, Battisti Corrado, Scalici Massimiliano.* Paper number 334397

**Microplastics in Maxwell Bay (Fildes Peninsula, Antarctica)** *Krojmal Evelyn*. Paper number 334439

A comparative assessment of microplastic abundance inside and outside a Marine Protected Area: The case of the National Marine Park of Zakynthos Isl., Ionian Sea, Greece Digka Nikoletta, Adamopoulou Argyro, Kokkali Athina, Danae Patsiou, Bray Laura, Kaberi Helen, Zeri Chrsitina, Tsangaris Catherine. Paper number 334476

# Spatial distribution of plastic pollution in a protected coastal Mediterranean wetland

Cesarini Giulia, Cera Alessandra, Battisti Corrado, Scalici Massimiliano.

Plastic litter was widespread worldwide, and in particular in the Mediterranean Sea, which is one of the most impacted areas. Coastal ecosystems, that provide several ecosystem services, are the most vulnerable marine habitats threated by plastic litter can derive from both sea and land. Although there are different management measures of marine litter, plastic pollution has impacted even the Marine Protected Areas. To deepen this topic, our research focused on plastic distribution in a protected costal area, 'Torre Flavia Wetland'. We also evaluated whether there was a correlation between the accumulation of plastic and natural debris. We collected plastic or natural items in 16 sampling sites of two transepts located in different areas of beach. Our results highlight plastic contamination in the study area, identifying the possible sources of different plastic deposition. A highest concentration of plastic is observed near the river mouth and breakwater, but overall microplastics is the most abundant size category of plastic litter found. The same factors seem influenced also the deposition of natural debris as there is a positive correlation between the weight of plastic and natural debris. Moreover, we observe a selection of plastic shapes, in the beach more fragments are found while the egagropiles (spheroids of Posidonia oceanica) entrapped more fibres. The plastic presence in the beach of 'Torre Flavia Wetland' generate concerns on local fauna and ecosystem conservation, reducing moreover the aesthetic and economic value. In addition, egagropiles are a source of food and shelter for different organisms. It is therefore necessary to carry out clean-up beach actions, considering however the greater effort required for the removal of microplastics compared to larger items. Voluntary actions as citizen science projects can provide a valid support to increase the management of plastic litter.

Keywords : beach litter , citizen science , coast , ecosystem conservation , Marine Protected Area , Mediterranean Basin , microplastic , plastic

# Spatial distribution of plastic pollution in a protected coastal Mediterranean wetland



#### Giulia Cesarini<sup>1</sup>, Alessandra Cera<sup>1</sup>, Corrado Battisti<sup>2</sup> and Massimiliano Scalici<sup>1</sup>



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#### BACKGROUND

- Plastic is persistent pollutant that widespread worldwide, impacting every type ecosystems. Beaches and dunes are considered hotspots of plastic pollution
- Mediterranean Sea is one of the most impacted areas. The main factors are: the semi-enclosed nature and special hydrodynamics, a coast with a high population density, intensive fishing, transport and tourism activities.
- Plastic litter can increase in permeability and grain size of sand modifying the biogeochemical cycles, interact with dune vegetation, adsorb environmental chemicals and impact several organisms, specially the meiofauna community.
- Aim: investigate (i) the occurrence and distribution of plastic litter in a protected area and (ii) the relationship between plastic litter and natural debris.

#### METHODS

- The study area is the beach of 'Torre Flavia Wetland' Natural Monument (Special Protection Area IT6030020), a small protected coastal Mediterranean wetland (15 ha) located on the Tyrrhenian seashore.
- Plastic or natural items are collected in 16 sampling sites of two transepts located in different areas of beach. The two transepts, A and B, are 500 m apart (Figure 1).
- Each sample is sorted according to the characteristics of the items: anthropogenic (*i.e.* plastic litter) or natural (*i.e.* vegetal or animal origin).
- Each item is weighted, after being dried at room temperature for 24 hours.
- Anthropogenic items are classified according to shape (*i.e.* fragments, pellets, fibres), colour and size (*i.e.* macro, meso e microplastics).
- The egagropiles of Posidonia oceanica collected, are opened and the eventual plastic items entrapped are also classified.

# RESULTS & DISCUSSIONS

- Transept B has more plastic items than transept A (Figure 2). The proximity of transept B to the river mouth and breakwater and being a more touristic area could explain the higher concentration of plastics.
  - Microplastic items are the most abundant size found in both transepts and white is the prevalent colour (Fig. 3), probably due to environmental exposure of plastics deposited on the beach.
  - The most frequent plastic shape found in the beach is fragment (63.3%), followed by pellet (36.3%) and fibre (0.4%).



Figure 3. Occurrence of plastic colours according to size (micro-, meso-, macroplastic) and shape (fragment, pellet, fibre) of the items.



Figure 4. Number, shape and colour of plastic items entrapped in the 4 egagropiles (EGA1-4) collected from transept B.

✓Our results show the presence of plastic litter on the coast of the protected area 'Torre Flavia Wetland', generating concerns on ecosystem conservation.

and natural weights of transept B samples (Pearson, r= 0.6, p<0.05).

between the size of plastic fiber and natural one present in the egagropiles.

✓There are several special protection species (147/2009/UE 'Bird' Directive) that could be threatened by the presence of plastic. For example, many birds nesting in the wetland can ingest microplastics litter.

- ✓ Detect spatial distribution of plastics along the coast and identify the sources, allows to intervene on the areas that most need removal and restoration interventions.
- ✓ Identified the size of plastic litter, it is possible proceed with targeted clean-up actions according to the presence of large items or microplastics.
- $\checkmark$  Citizen science projects can increase the success of beach cleaning actions.

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PLASTIC ITEMS BY SIZE

rectangles) in 'Torre Flavia Wetland' beach.

Figure 1. Study area (red line) and two sampling transepts (yellow

Figure 2. Number of plastic items and examples collected from transept A and B and divided by size (macro-, meso-, microplastic).



**CONCLUSIONS** 

Natural debris and plastic items are present in all sampling points in variable weight. Natural debris does not show a difference between the transepts but there is a positive significant correlation between plastic

#### **Microplastics in Maxwell Bay (Fildes Peninsula, Antarctica)**

Krojmal Evelyn.

Plastic pollution in aquatic ecosystems causes enormous concern due to its rapid dispersion and persistence in the environment. The marine Antarctic system does not escape this worldwide problem, as macro and microplastics in surface waters and marine-coastal zones have already been detected, which can result in a significant threat to this environment and its fragile fauna. However, the magnitude and sources of this threat in the Antarctic are far from being understood, and for this, the generation of basal information has been identified as a priority. This study aims at evaluating the presence of microplastics in surface marine water samples of Maxwell Bay (King George island, 62° 11'4''S and 58° 51'7''O), between 2016 and 2019. Water samples were taken with a Manta net (220 microns) in three different areas, one in front of the Collins glacier, another in front of the Uruguayan Antarctic Base, and a third at the adjacent bay Caleta Norma. Shape, colour, size and density of microplastics were analysed with polarized light microscopy, in the three replicates taken in each area. Microplastic densities varied between 4.4-30.5 items.m-3, which represents a difference of at least one order of magnitude higher with other Antarctic studies. We found no differences between sampling areas or years. Fibers were the most represented shape in each year and area sampled. In general, the average percentage of fibers and fragments in each area and year had a relation of approximately 90% of fibers and 10% of fragments, except for one site (1-2017) that had 68.2% and 30.7%, respectively. The sizes of the fibres oscillated between 0.001-4.92 mm and the fragments between 0.039-3.12 mm. Our results provide basal information that contributes directly to the objectives and challenges of the Antarctic Treaty System.

Keywords : Antarctica , Microplastics , Polarized light , Zooplankton

# MICROPLASTICS IN MAXWELL BAY (FILDES PENINSULA, ANTARCTICA)

EVELYN KROJMAL<sup>1</sup>, JUAN PABLO LOZOYA<sup>1</sup>, FRANCO TEIXEIRA DE MELLO<sup>1</sup>, JAVIER LENZI<sup>2</sup>, FLORENCIA ROSSI<sup>1</sup>, BÁRBARA DE FEO<sup>1</sup>, FIORELLA BRESESTI<sup>1</sup>, GISSELL LACEROT<sup>1</sup>

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The marine Antarctic system does not escape the worldwide problem of the plastics. Here, macro and microplastics in surface waters and marine-coastal zones have already been detected, posing a significant threat to this environment and its fragile fauna. However, the magnitude and origins of this threat in the Antarctic are far from being understood, and the generation of **basic information** has been identified as a priority.

#### **OBJECTIVES**

#### — General

 $\mathbb{E}$ 

Evaluate the presence of microplastics in surface water in Maxwell Bay (Antarctica, Fildes Peninsula,  $62^{\circ}$  11'4''S and 58° 51'7''O).

#### — Specific

(I) Elaborate and evaluate a suitable methodology protocol to analyze microplastics in ocean water samples.

(II) Analyze shape, size, colour, density (items/ $m^3$ ), as well as temporal and spatial distribution of microplastics.

#### <u>©</u> RESULTS



Fig. 4. Examples of MPs found in water samples

- The average total density found was 13,4 items/m<sup>3</sup> in 2016, 8,6 items/m<sup>3</sup> in 2017 and 3,2 items/m<sup>3</sup> in 2018 (items smaller than 220 >> micrometers were not considered).
- Fibers were the most common shape found.

und

- > The most represented sizes were smaller than the net mesh size.
- Densities were significantly higher in 2016 and 2017, compared to 2018 (df=2, p=0,046).
- No significant differences between sampling areas densities (f=1,56, p=0,27).



The study was conducted in the Scientific Antarctic Base Artigas (Maxwell Bay, Fildes Peninsula,  $62^{\circ}$  11'4'' S y 58° 51'7'' W).



Water samples were taken with a Manta net (220 microns) with flowmeter in three different zones, every summer between 2016 and 2018.





# **DISCUSSION**

- Densities were at least one order of magnitude higher than studies in nearby zones of the Antarctic Peninsula (e.g.:  $0.002/m^3$  Kuklinski et al., 2019;  $0.0035-0.00075/m^3$  Lacerda et al., 2019).
- Even though we found no evidence of laboratory contamination in the controls, we found a large amount of MP smaller than our sampling mesh size. Aggregation (Wang et al., 2020) with larger items during sampling could be responsible for this pattern, although more studies are necessary to understand it.

BIBLIOGRAPHY: \*Kuklinski, P., et al., (2019) Marine Pollution Bulletin, 149, 110573. \*Lacerda, A. L. D. F., et al., (2019) Scientific Reports 9.(1): 3977. \*Wang X., et al., (2020) Journal of Hazardous Materials, 402,









### A comparative assessment of microplastic abundance inside and outside a Marine Protected Area: The case of the National Marine Park of Zakynthos Isl., Ionian Sea, Greece

Digka Nikoletta, Adamopoulou Argyro, Kokkali Athina, Danae Patsiou, Bray Laura, Kaberi Helen, Zeri Chrsitina, Tsangaris Catherine.

Microplastics (plastic particles < 5 mm) are becoming a serious threat for the marineecosystems and one of the major concerns is related to their ubiquity: they are found from the sea surface and water column to the beach and seabed sediments or even ingested by marine organisms. This study provides a comparative assessment of microplastic pollutionin a non-protected and a protected marine area (MPA) in the Ionian Sea, by determining microplastic abundance, types, and polymer composition in three environmental compart-ments (sea surface, seabed sediment and marine biota). Specifically, surface water samples, seabed sediment samples and fish samples (Mullus surmuletus) were collected from two sitesinside and two sites outside the National Marine Park of Zakynthos, in June 2019. Microplastics were detected in all environmental compartments and were categorised by shape(i.e. fibers, fragments, films and pellets), colour (i.e. blue, black, red and transparent), sizeclass (i.e.<100µm, 100-500µm, 500-1000µm and>1000µm) and polymer type (i.e. PE,PP, PET, PVC, PS and PA). Microplastic abundances and categories among environmen-tal compartments were estimated and compared in all study sites. Ingested microplasticsby the benthic feederM. surmuletuswere tested for potential correlation to microplasticsdetected in the seabed sediment. This is one of the first works on microplastic abundancein MPAs in Greece and estimates the potential exposure of these ecosystems to this kind of pollution. Acknowledgments: This study was funded by the INTERREG MED project"Plastic Busters MPAs: preserving biodiversity from plastics in Mediterranean Marine Pro-tected Areas", co-financed by the European Regional Development Fund (grant agreementNo 4MED173.2M123027)

Keywords : ingestion , microplastics , MPA , sea surface , seabed sediment



International Conference. 23-27 November 2020. Lanzarote. Fate and impacts of Microplastics: Knowledge and Responsibilities



A COMPARATIVE ASSESSMENT OF MICROPLASTIC ABUNDANCE INSIDE AND OUTSIDE A MARINE PROTECTED AREA: The case of the National Marine Park of Zakynthos isl., Ionian Sea, Greece Nikoletta Digka, Aryiro Adamopoulou, Athina Kokkali, Danae Patsiou, Laura Bray, Helen Kaberi, Chrsitina Zeri, Catherine Tsangaris

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**INTRODUCTION** Microplastics (MPs-plastic particles <5 mm) are a serious threat for marine ecosystems and a major concern is their ubiquity: they are found from the sea surface and water column to the beach and seabed sediments or even ingested by marine organisms.



Observation under stereomicroscope
 FTIR for polymer identification
 Statistical analysis (t-test, R statistics 3.4.0)

#### DISCUSSION & CONLUSIONS

 $\checkmark$  Preliminary analysis show lower MP abundances (mean ± SE) inside the National Marine Park of Zakynthos in water, sediment and fish samples, although differences are not significant (p >0.05, t-test).

 $\checkmark$  Preliminary results (10% of all items) indicate PE as the most common polymer type in all environmental compartments tested.

 $\checkmark$  This is one of the first studies on microplastic abundance in MPAs in Greece that estimates the potential exposure of these ecosystems to this kind of pollution.



This study was funded by the INTERREG MED project "Plastic Busters MPAs: preserving biodiversity from plastics in Mediterranean Marine Protected Areas", co-financed by the European Regional Development Fund (grant agreement No 4MED17 3.2 M123 027).

**RESULTS** - MPs sized between 100-500 µm were the most abundant in sediment and fish samples.

- Fragments (> 50% of all items), fibers and films were detected in all environmental compartments. Foams were only found in surface water samples.

- Light colored MPs were the most common MPs in all environmental compartments.

-Polyethylene (PE) was the majority of the plastics tested.



In surface water average abundance ranged from 0.13- 0.25 MPs/m<sup>2</sup>



ranged from 4-57.6 MPs/kg dry weight

In fish average abundance ranged from 0.5-1.75 MPs/fish

### Session 23.9\_Me. Chaired by Patricia Ostiategui, Gran Canaria

# Spatial distribution and characterization of microplastics in subtidal sediments under stormwater discharge influence

Díaz-Jaramillo Mauricio, Rodriguez Florencia, Pegoraro César, Islas Maria Soledad, Suarez Ayelén, Gonzalez Mariana. Paper number 334446

# Mesoplastics on Joaquina beach, Santa Catarina Island, Brazil: sources, characterization and temporal trend

*Camila Andreussi, Zanetti Daniela, Leonel Juliana.* Paper number 334506

#### **Diving for assessing seafloor marine litter in shallow coastal MPAS** Morató Trobat, Mercè Compa, Ferrer Monserrat.

Paper number 334535

# Spatial distribution and characterization of microplastics in subtidal sediments under stormwater discharge influence

Díaz-Jaramillo Mauricio, Rodriguez Florencia, Pegoraro César, Islas Maria Soledad, Suarez Ayelén, Gonzalez Mariana.

Although microplastics are transported from land-based sources to aquatic environments there is currently little knowledge about its fate from land sources to coastal marine waters. In addition to riverine discharges, storm drainages from urban settlements might contribute an important source of microplastics. Mar del Plata city harbor (Buenos Aires, Argentina) holds the most important fishing fleet and shipyard facility in Argentina. Although that several contaminants have been reported in sediments from this area, the occurrence of microplastic has not be studied. The aim of this study was to evaluate the spatial distribution of microplastic in subtidal sediments (5-12 mt deep) from 4 sites in Mar del Plata's harbor, differing in their distance to "Del Barco" creek stormwater outlet. Microplastic content was determined (n:3 per site) by a density floating method using a zinc chloride (ZnCl2) solution ( $\delta$ 1.65). Mean total abundance of microplastics ranged from 1600 to 8700 items per kg d.w. Sediments from areas near to the discharge showed significantly higher abundances (p

Keywords : Argentina , Harbor , Microplastic , Stormwater discharge , subtidal sediments

# SPATIAL DISTRIBUTION AND CHARACTERIZATION OF MICROPLASTICS IN SUBTIDAL SEDIMENTS UNDER STORMWATER DISCHARGE INFLUENCE

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#### <u>Mauricio Díaz-Jaramillo</u><sup>\*1</sup>, Florencia Rodriguez<sup>1</sup>, Cesar Pegoraro<sup>1</sup>, Maria Soledad Islas<sup>1</sup>, Ayelén Suarez<sup>2</sup> and Mariana Gonzalez<sup>1</sup>

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Although microplastics are transported from land-based sources to aquatic environments there is currently little knowledge about its fate from land sources to coastal marine waters. In addition to riverine discharges, storm drainages from urban settlements might represent a punctual microplastic's source. The Mar del Plata's city harbor (Buenos Aires, Argentina) holds the most important fishing fleet and shipyard facility in Argentina. Although that several contaminants have been reported in sediments from this area, the occurrence of microplastic has not been yet studied. The aim of this work was to evaluate the spatial distribution of microplastic in subtidal sediments (5-12 mt deep) from 4 sites in Mar del Plata's harbor, differing in their distance to "Del Barco" creek stormwater outlet.



### **Discussion and Conclusions**

- Quality assurances showed good recovery rates. However, data obtained with handmade artificial sediments from translucent colored bottle/lids (PP,PET) pointed out that the visual examination and recovery of small particles might represent a source of underestimation for real samples.
- Sediments nearest to the stormwater discharge area showed higher levels of total microplastics than distant areas, our findings highlights the role of storm drain runoff in the microplastics transport to coastal areas. The outfall within a closed area as the harbour favours microplastic's deposition leading to high total abundances.
- We hypothesized that black particles observed presumably corresponded to tire wear particles transported by the stormwater outfall. On the other hand the red type particles would correspond to the polyester resin or other types of particles from ships/boats maintenance, recently observed in other port areas. Those features would be answered by further FT-IR confirmation analysis.
- These results represent the first analysis regarding the occurrence of microplastics in sediments from Mar del Plata's harbor and the influence of its most important stormwater outfall. This preliminary approach should also give insights about the different sources of microplastic that may be present in big port cities.

# Mesoplastics on Joaquina Beach, Santa Catarina Island, Brazil: sources, characterization and temporal trend

Camila Andreussi, Zanetti Daniela, Leonel Juliana.

The quality of sandy beaches is at risk because of the incidence of plastic, a contaminant of great concern due to their impacts to the ecosystem. The temporal variability of mesoplastics (5 to 25mm) at Joaquina beach. from July/2018 to January/2020, was analyzed aiming to evaluate which factors. natural and/or anthropic, are the main responsible for its occurrence. Monthly samplings were carried out in 2 transects (strandline x backshore) at 12 fixed points. Items sampled were counted, measured, photographed and characterized. In the total 284 items were sampled with density varying from 0 to 24 items m-2 (average=6.4); lower than those reported for other parts of the world (i.e. India and Korea). As expected, higher concentrations of mesoplastics were found in the backshore region highlighting its importance for trapping litter. Fragments were the predominant type of plastic found (76%), which sources are not traceable, followed by Styrofoam (17%), which is associated with fishing since its incidence was higher in the months of less use of the beach by tourists. Occurrence of mesoplastic showed no relation with seasonal use of the beach suggesting that their presence may be related to meteorological and oceanographic conditions. For example, the highest amount of plastic was found when strong winds blowing in the southwest direction were registered, whereas the lower amount was found collected concomitant to north winds. The lowest occurrence of items was observed in May, which also had the highest precipitation in the sampling period (196.60 mm), implying that rainwater runoff can remove particles from the beach. The influence of storm events, such as cold fronts and meteorological tides, and how they can generate an increase in the number of items were also reported.

Keywords : coastal ecosystem , contaminant , fragments , winds



# MICRO20:334506

Mesoplastics on Joaquina beach, Santa Catarina Island, Brazil: sources, characterization and temporal trend



Camila Andreussi, Daniela Zanetti, Juliana Leonel

The temporal variability of mesoplastics (5 to 25mm) at Joaquina beach, from July/2018 to January/2020, was analyzed aiming to evaluate which factors, natural and/or anthropic, are the main responsible for its occurrence.



Figure 1 - Map of the study area, Joaquina beach located in the southeastern sector of the island of SC.















- Fragments were the predominant type of plastic in all samples.
- There was a greater occurrence of items in the backshore, where dunes play a key role in the accumulation and retention of debris.
- Occurrence of mesoplastic showed no relation with seasonal use of the beach suggesting that their presence may be related to meteorological and oceanographic conditions.

Acknowledgement: OCAPES

#### Diving for assessing seafloor marine litter in shallow coastal MPAs

Morató Trobat Mercè Compa Ferrer Monserrat.

The Mediterranean Sea is one of the most affected seas by marine litter and Marine Protected Areas (MPAs) are not free from this type of pollution (Alomar et al., 2016). Litter in the marine environment is a risk for different species and ecosystems (Deudero and Alomar, 2015), and the Balearic island are susceptible to plastic pollution in coastal areas especially during the summer season (Compa et al., 2019). The abundance and composition of marine litter were surveyed on six beaches and twelve coastal seafloor areas were monitored through scuba diving surveys in Cabrera National Park (Balearic Islands) to characterize and study marine litter abundance and composition in this MPA. According to beaches, artificial polymer materials made up 94% of the items identified, in contrast, in the seafloor samples, the artificial polymer materials were the main fraction (96%) in a seafloor located within the port follow by glass and ceramics (up to 83%) in an integral reserve area. Preliminary results highlight that there is variation in the amount of macrolitter arriving to the beaches and the seafloor of Cabrera National Park. Overall, more than 4750 macro litter items were identified on the beaches and in the surrounding seafloor shallow areas indicating not only local sources from within the port area but also high transference to areas where human activities are either limited or with high restrictions.

Keywords : Anthropogenic impacts , Balearic Islands , Benthic litter , Coastal conservation , Marine Protected Areas , Western Mediterranean Sea

### DIVING FOR ASSESSING SEAFLOOR MARINE LITTER IN SHALLOW COASTAL MPAS; IS THERE A RELATION WITH THE BEACHED LITTER?

Mercè Morató, Montserrat Compa, Carme Alomar, Elvira Álvarez and Salud Deudero<sup>2</sup>

MICR02020 ATIONAL CONFERENCE FATE AND IMPACTS OF MECROPI ASTICS: KNOWLEDGE AND RESPONSIBILITIES

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The Mediterranean Sea is one of the most affected seas by marine litter, and Marine Protected Areas (MPAs) are not free from this type of pollution (Alomar et al., 2016). Considering its widespread presence in the marine environment, there is evidence and concern on its availablity for different species with different trophic guilds (Alomar and Deudero, 2015) and coastal species have been seen to be more susceptible to this type of contamination (Compa et al., 2019). The Plastic Busters MPAs: preserving biodiversity from plastics in Mediterranean Marine Protected Areas is an Interreg Med funded project aiming to maintain biodiversity and preserve natural ecosystems in pelagic and coastal marine protected areas. Within its objective, beaches were visually surveyed and the seafloor was monitored through scuba diving surveys to characterize and study the marine litter in an effort to identify potential sources.



National Park The study area (Figure 2) consists of the beaches

and shallow coastal areas of Cabrera National Park (Balearic Islands).



Fig 2. Study area in Cabrera national Park Table 1. Top 10 of total items on beaches and marine seafloor of Cabrera National Park. Red the most abundant

**M** ethods

Litter classification and

quantification

**Results & Discussion** 

According to beaches, artificial polymer materials made up 94% of the items identified, in contrast. in the seafloor samples, the artificial polymer materials were the main fraction (96%) in a seafloor located within the port follow by glass and ceramics (up to 83%) in an integral reserve area. Preliminary results highlight that there is variation in the amount of macrolitter arriving to the beaches and the seafloor of Cabrera National Park. Overall. more than 5000 (4395 items of macrolitter on beaches, and 668 of macrolitter on

marine macro litter items were

identified on the beaches and in the surrounding

seafloor shallow areas indicating not only local

sources from within the port area but also high

transference to areas where human activities are

A Spearman's correlation indicated there is a

significant relationship (R = 0.42, p < = 0.001)

between the total number of items on the seafloor and the number of items on the beaches

either limited or with high restrictions.



fixed section of a beach covering the whole area from the strandline to the back of the beach, and the survey area for macrolitter on the seafloor is defined by time scuba diving visual transects.

For litter classification and quantification (number of items) the "Joint List of Marine Litter Items Categories" was used for comparison amongst sites (table 1).

Top 10	0 Items Beaches			
TOP 10	Material type	Code	Items name	Number of items (2 total)
1	ARTIFICIAL POLYMER MATERIALS	G79	Plastic pieces 2.5 cm > < 50cm	1005
2	ARTIFICIAL POLYMER MATERIALS	G21	Plastic caps/lids from drinks	373
3	SANITARY WASTE	G95	Cotton bud sticks	292
4	ARTIFICIAL POLYMER MATERIALS	G10	Food containers incl. fast food containers	273
5	GLASS/CERAMICS	G208	Glass or ceramic fragments >2.5cm	267
6	ARTIFICIAL POLYMER MATERIALS	G53	Nets and pieces of net < 50 cm	220
7	ARTIFICIAL POLYMER MATERIALS	G4	Small plastic bags, e.g. freezer bags, including pieces	101
8	ARTIFICIAL POLYMER MATERIALS	G67	Sheets, industrial packaging, plastic sheeting	89
9	ARTIFICIAL POLYMER MATERIALS	G73	Foam sponge	89
10	ARTIFICIAL POLYMER MATERIALS	G124	Other plastic/polystyrene items (identifiable)	85
Top 10 Items Seafloor				
TOP 10	Material type	Code	Items name	Number of items ( <b>∑</b> total)

101 10	waterial type	coue	items name	(Σ total)
1	GLASS/CERAMICS	G200	Bottles, including pieces	202
2	GLASS/CERAMICS	G208	Glass or ceramic fragments >2.5cm	57
3	ARTIFICIAL POLYMER MATERIALS	G10	Food containers incl. fast food containers	47
4	ARTIFICIAL POLYMER MATERIALS	G79	Plastic pieces 2.5 cm > < 50cm	44
5	ARTIFICIAL POLYMER MATERIALS	G2	Bags	36
6	ARTIFICIAL POLYMER MATERIALS	G53	Nets and pieces of net < 50 cm	31
7	ARTIFICIAL POLYMER MATERIALS	G67	Sheets, industrial packaging, plastic sheeting	19
8	ARTIFICIAL POLYMER MATERIALS	G7	Drink bottles <=0.5l	15
9	ARTIFICIAL POLYMER MATERIALS	G13	Other bottles & containers (drums)	12
10	ARTIFICIAL POLYMER MATERIALS	G124	Other plastic/polystyrene items (identifiable)	10



items, green the least abundant items

Fig 3. Percentage of number of items by type of material (Beach and underwater seafloor macrolitter) acording to "Join List of Marine Litter Items Categories" in Cabrera National Park



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Fig 4. Spearman's correlation of total number of items on the seafloor and beaches of Cabrera National Park

#### (Figure 4). References

seafloor)

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### Session 23.9\_Ma. Chaired by Mateo Cordier, Guyancourt

Single and joint effects of chronic exposure to polyethylene microplastics and chlortoluron on oyster, *Crassostrea gigas*: biomarkers and metabolomics approaches *Bringer Arno*. Paper number 334406

# Investigation of microplastics ingestion and effects in striped red mullet in east Mediterranean Sea

Patsiou Danae, Digka Nikoletta, Tsangaris Catherine. Paper number 334424

**Physical and chemical effects of microplastics on the marine polychaete** *Capitella spp. Lyngstad Inger, Booth Andy, Farkas Julia, Igartua Amaia, Wagner Martin, Sørensen Lisbet.* Paper number 334508

# Single and joint effects of chronic exposure to polyethylene microplastics and chlortoluron on oyster, *Crassostrea gigas*: biomarkers and metabolomics approaches

Bringer Arno.

Microplastics (MPs) are a pollutant of increasing environmental concern based on their ubiquitous and persistent nature. Pesticides are widely used in agricultural settings as well as in many homes and residential gardens and may enter estuarine and coastal environments. Microplastics and pesticides are identified as two environmental pollutants that have an adverse impact on the environment. The knowledge about the combined exposure of pesticides and MPs may facilitate further assessment of their ecotoxicity. In this study, a 24-day exposure was set up on juvenile oysters (C. gigas) to various contaminants potentially present in the natural marine environment. Four experimental conditions were tested: control, polyethylene MPs (HDPE 20-25 µm, 10 µg.L-1), chlortoluron (herbicide, 30 µg.L-1) and cocktail mixture (MPs + chlortoluron). Bivalve (juvenile oysters) samples were taken at two sampling dates (D5 and D25) in order to perform various molecular defence biomarker assays: SOD (oxidative stress), GST (detoxification process), MDA (lipid peroxidation), and Laccase (immune reaction). Measures were conducted in the digestive glands due to their bioaccumulation properties. Biochemical analysis showed that the oxidative damage of oysters exposed to the cocktail could be observed on the D25. The condition with MPs alone, on the other hand, shows a higher activity in Laccase in response to environmental stress on the D25. In addition, we propose to evaluate the risk assessment of the effects of MPs and chlortoluron on the bioaccumulation, biomarkers and metabolic profile (metabolic and xenobiotic networks) of juvenile oysters. As part of a modelling approach, various parameters will be assimilated in order to develop sensitive and predictive responses for improving the diagnosis of the quality of the marine environment.

Keywords : Biomarkers & metabolomics approaches , Microplastic , Oysters

Single and joint effects of chronic exposure to polyethylene microplastics and chlortoluron on oyster, *Crassostrea gigas*: biomarkers and metabolomics approaches

Arno Bringer <sup>a</sup>, Emilien Jamin <sup>b</sup>, Emmanuel Dubillot <sup>a</sup>, Valérie Huet <sup>a</sup>, Jérôme Cachot <sup>c</sup>, Hélène Thomas <sup>a</sup> <sup>a</sup> Littoral Environnement et Sociétés (LIENSs), UMR 7266, CNRS- La Rochelle Université, F-17042 La Rochelle Cedex 01, France. <sup>b</sup> MetaToul-MetaboHUB, National infrastructure of metabolomics and Fluxomics, Toulouse, France – MetaToul, France. <sup>c</sup> Laboratoire EPOC, UMR 5805, CNRS Université de.Bordeaux, F-33600 Pessac, France.

#### CONTEXT

In this study, a 25-day exposure was set up on juvenile oysters (*C. gigas*) to various contaminants potentially present in the natural marine environment. Four conditions were tested: control (without pollutants), polyethylene (20-25  $\mu$ m, 10  $\mu$ g.L<sup>-1</sup>) microplastics (MPs), chlortoluron (herbicide, 30  $\mu$ g.L<sup>-1</sup>) and cocktail mixture (MPs + chlortoluron). Two sampling times (D<sub>5</sub> and D<sub>25</sub>) made it possible to study the metabolic responses in all tissus of oyster and the activities of biomarkers in response to stress, in digestive gland only. Environmental metabolomics is a holistic approach that provides insight into the metabolic status of an organism, while the biomarkers approach is more integrative.



### Investigation of microplastics ingestion and effects in striped red mullet in east Mediterranean Sea

Patsiou Danae, Digka Nikoletta, Tsangaris Catherine.

Marine plastic debris is increasing and so does the microplastic (MP) ingestion by marine organisms. Detection of MPs in the marine environment is ongoing, however, there is lack of consensus regarding the effects of MP ingestion in marine organisms. The aim of the present study is to assess MP ingestion together with cellular and biochemical biomarkers in native Mediterranean fish species proposed as bioindicators of MPs, at coastal sites under different human pressure intensity in the Ionian Sea, and to investigate whether biomarker variations can be related to the occurrence of MPs in their gastro-intestinal (GI) tract. Striped red mullets Mullus surmuletus, were sampled along the tourist coast of Zakynthos island and inside the Zakynthos National Marine Park. The fish GI tracts were digested using 15% H2O2 at oC and then filtered to detect ingested MPs. The polymer identification analysis using Fourier-Transform Infrared Spectroscopy analysis of the samples is ongoing. Blood samples were collected from alive M. surmuletus to evaluate DNA damage, and the results so far indicate no differences in the micronuclei occurrence in fish blood cells among sampling sites. The investigation of biochemical biomarkers includes: acetylocholinesterase in muscle tissues to evaluate neurotoxicity, and catalase and glutathione S-transferase in the liver tissues to assess antioxidant defence and biotransformation responses. The present study carried out in the framework of the Plastic Busters MPAs project intends to measure the extend of MPs pollution by in situ investigation of cellular and biochemical parameters of native Mediterranean species, to attribute the animal health status directly to the presence of MPs in the animal.

Keywords : ecotoxicological biomarkers , Greece , Ionian Sea , microplastic ingestion , Mullus surmuletus



# INVESTIGATION OF MICROPLASTIC INGESTION AND EFFECTS IN STRIPED RED MULLET IN EAST MEDITERRANEAN SEA





#### Danae Patsiou\*, Nikoletta Digka\*, Catherine Tsangaris

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#### Introduction

Marine plastic debris is increasing and so does the microplastic (MP) ingestion by marine organisms. Detection of MPs in the marine environment is ongoing, however, there is lack of consensus regarding the effects of MP ingestion in marine organisms. The striped red mullet, *Mullus surmuletus*, is a native Mediterranean fish species proposed as bioindicator for benthic MP monitoring (1,2).

#### Aims of the study

1) Assessment of MP ingestion by fish collected in coastal sites of different human pressure intensity, the Marine National Park of Zakynthos island and coastal sites outside the marine protected area (MPA, East Ionian sea, Greece) and;

2) investigation of whether cellular and biochemical biomarker variations can be attributed to the occurrence of MPs in fish gastro-intestinal (GI) tract.

#### **Results**

Table 1: Number of individuals examined, occurrence of ingested microplastics (% of individuals containing microplastics) and abundance of microplastics in *M. surmuletus* GI. Microplastic abundance (mean ± SD) is expressed as the average number of microplastic items per individual in all individuals examined

	MPA	outside MPA
number of individuals examined	31	62
number of individuals containing MPs	15 (48.4 %)	20 (32.3 %)
number of MPs per individual	0.84 ± 1.2	0.97 ± 1



Figure 1: Shape of microplastics (%) detected in *M. surmuletus* GI.



Figure2: A) Micronuclei frequencies (‰) as recorded in 4000 red blood cells for each *M. surmuletus* alive individual sampled outside the MPA (orange triangles and curve) and inside the MPA (blue circles and curve). B, C and D) ACHE (in nmoles ACTC/min/mg protein), GST (in nmoles CDNB/mg protein) and CAT (in units/mg protein) activities, respectively. No significant differences were observed for micronucleus frequency or ACHE, GST and CAT activities between the MPA and areas outside the MPA that are related to the MP content in the GI tract of *M. surmuletus* (glm, p>0.05).

#### Materials & Methods

Striped red mullets *Mullus surmuletus* were sampled along the tourist coast of Zakynthos island (outside MPA) and inside the Zakynthos National Marine Park (MPA) in June 2019.

- Blood samples were collected from alive individuals to evaluate micronuclei presence
  - Biochemical biomarkers analyses included:
    - 1. acetylocholinesterase (ACHE) in muscle tissues (3)
    - 2. glutathione S-transferase (GST) and catalase (CAT) in the liver tissues (4,5)
  - 3. protein determination conducted following the Bradford method (6)
  - The fish GI tracts were digested using 15%  $\rm H_2O_2$  at <50 °C and then filtered to detect ingested MPs under stereomicroscope.
  - The polymer identification analysis using ATR Fourier-Transform Infrared Spectroscopy (FTIR) and the analysis of smaller items (down to 100 μm) using FTIR microscopy is ongoing.

#### **Statistical analysis**

The micronuclei frequencies (‰) and enzyme activity levels (dependent variables) were modelled by general linear model (GLM) according to the independent variable of MPs content in GI and the addition of sampling site factor (e.g. MPA or outside MPA; p-value of < 0.05). Differences between MPA and outside MPA sites were determined by pair-wise contrast statements. The statistical analyses were conducted using R statistics (version 3.4.0).



#### **Discussion – Conclusions**

- No differences were observed on the occurrence of MPs in *M. surmuletus* GI tract, neither on abundance of MPs per individual (t-test, chi square, p>0.05).
- 2. Inside the MPA, the mean number of MPs ingested per individual was 0.84, and outside the MPA was 0.97 MPs.
- 3. The results so far indicate no differences in micronuclei occurrence in fish blood cells between sampling sites.
- 4. Similarly, no effect of the sampling site was observed in the activity of ACHE, GST and CAT enzymes.
- No differences were observed in micronuclei presence or the enzyme activities between the two sampling sites (MPA vs outside MPA) that can be attributed to MP presence in fish GI.
- 6. However, a negative relation was observed between GST and CAT activities, and the increasing presence of MPs in the fish GI tract in both sampling sites.
- 7. Further work on MP polymer type identification and additional biomarker effects on toxicology is ongoing.

#### Acknowledgements

The authors would like to thank the HCMR Marine Litter Research Group, the HCMR technician Yiorgos Papas for his help during the sampling cruises and the placement student Anna Misalidi for her help in the analysis of the samples.

This study was funded by the INTERREG MED project "Plastic Busters MPAs: preserving biodiversity from plastics in Mediterranean Marine Protected Areas", co-financed by the European Regional Development Fund (grant agreement No 4MED17\_3.2\_M123\_027).

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# Physical and chemical effects of microplastics on the marine polychaete *Capitella spp*.

Lyngstad Inger, Booth Andy, Farkas Julia, Igartua Amaia, Wagner Martin, Sørensen Lisbet.

Although numerous studies have documented the effects of microplastics (MP) exposure on marine organisms, there is still uncertainty regarding to what degree such effects are caused by physical toxicity from particles or chemical toxicity from leaching chemicals associated with MP. In this study, we investigate the contribution of these two types of effects through a sublethal exposure study on the benthic polychaete deposit feeder Capitella spp. The study is conducted with MP produced from several materials, in order to see how effects might vary as a function of polymer type and additive chemical content. Firstly, solvent extracts from 50 plastic consumer products will be screened using the Bacteria Luminescence Toxicity (BLT) test to establish the baseline toxicity induced by the additive chemicals present. Based on this, a sub-group of materials representing both high and low chemical toxicity will be selected. The selected material will be cryomilled into irregular, polydisperse MP. Capitella spp. will be exposed for a full life cycle (appr. 21 d) to four treatments: MP with associated chemicals, the chemical leachate from the same material, cleanMP with the leachate removed and a control with natural silica particles. For this ongoing work, we hypothesise (i) that exposure to MP with high levels of associated chemicals and MP leachates derived from those materials will affect sublethal parameters of Capitella spp. to a higher degree than the cleanMP, and (ii) that cleanMP will have the same effect as the natural particles. The study also aims to assess with the BLT screening test is suitable for predicting toxicity in higher trophic levels.

Keywords : BLT , Capitalla , Leachate , Microplastics
# Physical and chemical effects of microplastics on the marine polychaete *Capitella* spp.

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icroLEA

#### Introduction

- Plastic additives and non-intentionally added substances may alter microplastic (MP) toxicity
- The number of chemical features varies greatly in different plastic materials
- Combining chemical analysis and baseline toxicity screening can help predict which plastic materials will have a higher toxic effect on higher organisms
- As MPs accumulate in sediments, benthic deposit feeders might be of high risk to MP and plastic additive exposure

#### Goals

- Determine sublethal endpoints of longterm MP and leachate exposure in the marine polychaete *Capitella spp.*
- Identify the respective contribution of physical particle and chemical content on observed toxicity
- Elucidate the difference in toxicity from "highly toxic" and "less toxic" plastic products



Day 2, Tuesday 24th. November 2020			
9h-10h	24.1_O	24.1_Me	24.1_Ma
10h-10h15	24.1_Gaia: 3 sessions brief		
10h30-11h30	<b>24.2_O</b>	24.2_Me	24.2_Ma
11h30-11h45	24.2_Gaia: 3 sessions brief		
12h-13h	<b>24.3_O</b>	24.3_Me	24.3_Ma
13h-13h15	24.3_Gaia: 3 sessions brief		
13h15-13h45	Poster.24.4_O	Poster.24.4_Me	Poster.24.4_Ma
14h-15h	24.5_O	24.5_Me	24.5_Ma
15h-15h15	24.5_Gaia: 3 sessions brief		
15h30-16h30	24.6_O	24.6_Me	24.6_Ma
16h30-16h45	24.6_Gaia: 3 sessions brief		
17h-18h	24.7_O	24.7_Me	24.7_Ma
18h-18h15	24.7_Gaia: 3 sessions brief		
18h30-19h	Poster.24.8_O	Poster.24.8_Me	Poster.24.8_Ma
19h-19h30	Poster.24.9_O	Poster.24.9_Ma	
19h30-20h	Poster.24.10_O	Poster.24.10_Me	Poster.24.10_Ma

### Day 2/5, Tuesday 24<sup>th</sup> November 2020

#### Session 24.1\_O. Chaired by Matthew Cole and Zara Botterell, Plymouth

**Influence of pH and temperature on the adsorption/desorption capacity of microplastics** *Godoy V., Martín-Lara M.a., Blázquez G., Calero M..* Paper number 333848

Effects of stratified water column on settling dynamics of microplastics and implications for interactions between microplastics and marine ecosystem *Mrokowska Magdalena*. Paper number 334377

**Exposure to microplastics can cause stress-induced spawning on arctic copepods** *Rodriguez Torres Rocío, Almeda Rodrigo, Kristiansen Michael, Rist Sinja, Winding Mie S., Nielsen Torkel G.*. Paper number 334472

# Influence of pH and temperature on the adsorption/desorption capacity of microplastics

Godoy V., Martín-Lara M.a., Blázquez G., Calero M.

Microplastics have a demonstrated capacity to adsorb chemical substances (metals, pharmaceuticals and others) that are frequent in seawater, freshwater or wastewater. This poses a high risk for fauna that ingest microplastics accidentally, due to the fact that, under acidic pH and high temperature conditions, microplastics can release the adsorbed contaminants into the organism. In the present study, kinetic and equilibrium tests were carried out with lead and amoxicillin in Milli-Q water, using microplastics of the most common polymers (polypropylene and polyethylene). The selected concentration for kinetic tests was 1 mg/l, whereas in equilibrium tests concentrations of 0.5, 1, 2, 4, 16 and 32 mg/l were used. All results were fitted to Langmuir and Freundlich models. Also, desorption tests were carried out in Milli-Q water at three pH values (2, 4, 6) and two temperatures (25 and 40°C). The results showed large differences in adsorption behavior between metal and pharmaceutical, and also between polymers. In the case of lead with polypropylene, the equilibrium was reached after 5 days of contact with the microplastics, whereas in the case of amoxicillin with polyethylene, equilibrium was not reached after 28 days of contact. The desorption tests with amoxicillin and lead showed, in general, a great release at the lowest tested temperature (25°C) and pH (2). In desorption tests with lead, the greatest desorption occurred at pH 4 at both temperatures. The temperature of 40°C and pH 6 showed the lowest rate of desorption. In desorption tests with amoxicillin, the greatest desorption ocurred at pH 6 and temperature of 40°C, just the opposite of the results obtained for lead. However, a combined phenomenon of adsorption-desorption was observed, which indicate the influence of several factors in this case.

Keywords : Adsorption , Kinetic , Microplastics , Polyethylene , Water pollutants

# **Effects of stratified water column on settling dynamics of microplastics and implications for interactions between microplastics and marine ecosystem** Mrokowska Magdalena.

Recent studies have indicated that microplastics are likely to accumulate at pycnoclines, that is the regions of sharp density gradients induced by the vertical variation of salinity and/or temperature. Moreover, pychoclines present in the ocean, coastal regions, and estuaries cause a pronounced decrease in settling velocity of natural marine particles such as plankton, faecal pellets, and detritus causing their accumulation at density transitions. In such conditions, intensified interactions between microplastics and natural marine particles may occur. The knowledge on the interactions between microplastics and marine biological components in stratified systems is poor and research on this topic poses a challenge, since interdisciplinary studies are necessary to gain understanding of these processes. Results shown in this presentation may help understand the role of physical aspects of microplastic settling that could be used in further studies on their fate in marine system. I demonstrate the effects of water column stratification on settling dynamics of plastic non-spherical particles. I present laboratory experiments in which disks made of ABS were settling in a column of salt-water solution. Disks were of diameters less than 5 mm, and showed a complex pattern of reorientations and velocity variation during settling through density transition. Visualization (cameras with macro lenses and backlight) combined with image analysis methods were used to measure the pattern of trajectory, variation of particle orientation, and settling velocity. The results indicate that disks decelerate significantly in the pycnocline, and existing methods of particle flux assessment overestimate settling velocities. Consequently, microplastics reside in the transition layer much longer than predicted, which may explain to some extent enhanced interactions with biota such as ingestion by zooplankton, adsorption of heavy metals, biofouling, aggregation, and incorporation of microplastics into marine snow in stratified aquatic systems. Reorientations may additionally induce interactions between microplastics and other types of marine particles.

Keywords : density transition , disks , microplastics , particle settling , stratification

**Exposure to microplastics can cause stress-induced spawning on arctic copepods** Rodriguez Torres Rocío, Almeda Rodrigo, Kristiansen Michael, Rist Sinja, Winding Mie S., Nielsen Torkel G.

The Arctic is composed of sensitive ecosystems that are currently experiencing enormous changes. Knowledge about the effects of microplastic pollution on the arctic ecosystems is still scarce despite the extremely high concentrations of microplastics (MPs) currently found in arctic sea ice. We experimentally investigated the ingestion and impact of virgin MPs (20um polvethylene spheres) on three key arctic copepods: Calanus finmarchicus, C. glacialis and C. hyperboreus. Copepod females were exposed to two MP concentrations (200 and 20000 MP L-1) in combination with different food concentrations (from 50 cells mL-1 to 5000 cells mL-1) for 6 days. We found that the three copepod species ingested MPs, up to 900 particles cop.-1 d-1. The amount of MPs inside the fecal pellets increased exponentially with decreasing food concentration. Fecal pellet production rates were not affected by the ingestion of MPs. However, egg production rate increased when copepods were exposed to MPs, suggesting that MPs causes stress-induced spawning. The presence of MPs inside the fecal pellets did not affect their sinking velocities. Overall, our results indicates that: (1) ingestion of virgin MP microspheres have a low impact on fecal pellet production rates of arctic Calanus species, (2) exposure to MPs can induce behavioral stress responses (e.g., increased spawning) and, (3) copepod fecal pellets may play a crucial role on the fate of virgin MP in the ocean by packing and vertically transporting the plankton-size particles to the sea floor.

Keywords : arctic , copepods , egg production , fecal pellet production , ingestion

#### Session 24.1\_Me. Chaired by Christian Laforsch, Bayreuth

**Microplastics: laws of formation, transport, physical-chemical behaviour and biological effects** *Laforsch Christian.* Paper number 334583

Small(er) particles, big(ger) problems? Fate, transport and implications of nanoplastic in the environment *Mitrano Denise*. Paper number 334123

### LBM simulations of raindrop impacts demonstrate microplastic transport from the ocean into the atmosphere

*Lehmann Moritz, Gekle Stephan.* Paper number 334143

**Environmental exposure enhances the internalisation of microplastic particles into cells** *Ramsperger Anja Frm, Bangaloore Narayana Vinay K, Gross Wolfgang, Mohanraj John, Thelakkat Mukundan, Greiner Andreas, Schmalz Holger, Kress Holger, Laforsch Christian.* Paper number 333340

# **Microplastics:** laws of formation, transport, physical-chemical behaviour and biological effects

Laforsch Christian.

The CRC1357 Microplastics at the University of Bayreuth is a large interdisciplinary collaborative research centre funded by the DFG. The speakers are Christian Laforsch (Animal Ecology I) and Andreas Greiner (Macromolecular Chemistry II). This special session highlights new CRC1357 projects results of the main research areas: Biological Effects, Environmental behaviour and migration in and between ecosystems, Mechanisms of degradation of plastics in natural and technical systems. Guest keynote lecturer of the CRC1357 session is Denise Mitrano (ETH Zürich). The special session will be live-streamed on Tuesday 24th November 2020 and available in recorded formats (full session & single talks). 09:00 Christian Laforsch, UBT Welcome & "CRC1357 Microplastics-Fundamental Questions in Microplastics Research" 09:20 Keynote Denise Mitrano, ETH "Small(er) particles, big(ger) problems? Fate, transport and implications of nanoplastic in the environment" [micro2020:334123] 10:15 Keynote Seema Agarwal, UBT "Are Biodegradable Polymers a solution to the Microplastic issue?" [micro2020:334142] 10:50 WORLD CAFÉ / COFFEE BREAK 11:15 Anja Ramsperger, UBT "Environmental Exposure Enhances the Cellular Internalisation of Microplastic Particles" [micro2020:333340] 11:35 Anja Holzinger, UBT "Comparing the effects of multiple microplastic polymer types and shapes on the earthworm Eisenia fetida" [micro2020:334379] 11:55 Sven Frei, UBT "Occurrence of microplastics in the hyporheic zone of rivers" [micro2020:334135] 12:15 Moritz Lehmann, UBT "LBM Simulations of Raindrop Impacts demonstrate Microplastic Transport into the Atmosphere" [micro2020:334143] 12:35 Makis Gkoutselis Microplastic as Spreader of Fungal Pathogens [micro2020:334558] 12:55 Hannes Laermanns, Uni Köln "Tracing the lateral transport of microplastic particles with an advancedimaging sCMOS camera" [micro2020:334274] 13:15 Martin Löder, UBT "Deeper insights: The potential of X-ray micro-computed tomography (µCT) in microplastic studies" [micro2020:334425]

Keywords : biological effects , chemical properties , formation , limnic ecosystems , migration , physical , polymer chemistry , terrestrial ecosystems

# Small(er) particles, big(ger) problems? Fate, transport and implications of nanoplastic in the environment

Mitrano Denise.

Numerous studies have made the ubiquitous presence of plastic in the environment undeniable, and thus it no longer comes as a surprise when scientists monitor the accumulation of microplastic litter and microplastic fragments in both urban and remote sites. Ultimately, the different physical and chemical characteristics of the different size classes of plastic pollution (macroplastic, microplastic and nanoplastic) will result in divergent fate and hazards. Quantitative data are still limited due to analytical difficulties to detect nanoplastics in complex matrices, and thus mechanistic studies to understand the fate, transport and biological interactions of these materials are limited. While progress is still ongoing to develop protocols to measure particulate plastic in field studies, researchers who study these processes in bench top or pilot scale studies can take advantage of an entirely different approach. In the last years, we have synthesized a variety of particulate plastics with an embedded inorganic fingerprint which can be used as a proxy to detect plastic by common analytical techniques for metals analysis. In practice, this affords for quicker and more accurate sampling and subsequently allows us to investigate the basic processes and pathways which control particulate plastic fate and impacts. To highlight the utility of this approach, we have used these materials in a number of different test systems including, 1) mass balance and flux of plastic through pilot-scale wastewater and drinking water treatment plants, 2) application of sewage sludge in agriculture and nanoplastic mobility through porous media and 3) the interaction and uptake of nanoplastics with plants and organism. As human and environmental nanoscientists, we should try to place nanoplastic in the context of global plastic pollution by assessing its source and risk, but also by assessing commonalities nanoplastics may share with other nano-sized objects in environmental systems, such as engineered nanomaterials and natural colloids.

Keywords : fate , nanoplastic , risk assessment , transport , uptake

## LBM simulations of raindrop impacts demonstrate microplastic transport from the ocean into the atmosphere

Lehmann Moritz, Gekle Stephan.

It is a common misbelief that microplastics getting into the ocean stay there indefinitely. Recent observations detected elevated levels of microplastics in the air near the coastline [Allen et al. Examination of the ocean as a source for atmospheric microplastics. In: PloS one 15.5 (2020), e02327469]. Here we show that raindrop impacts can possibly explain these observations as they constitute a mechanism for microplastic transport from the ocean into the atmosphere. We model these impacts with the Volume-of-Fluid lattice Boltzmann method extended by the immersed-boundary method. We use typical sizes and velocities of real-world raindrops - a parameter range previously inaccessible to 3D simulations. We find that a 5mm diameter raindrop impact ejects more than 160 droplets and that at least 75 of them are so fast that they reach an altitude above half a meter in the air. We further show that the droplets indeed contain microplastic concentrations similar to the ocean a few millimeters below the surface.

Keywords : droplet , fluid dynamics , immersed boundary method , impact , lattice Boltzmann method , LBM , particle , raindrop , transport , VoF

## Environmental exposure enhances the internalisation of microplastic particles into cells

Ramsperger Anja Frm, Bangaloore Narayana Vinay K, Gross Wolfgang, Mohanraj John, Thelakkat Mukundan, Greiner Andreas, Schmalz Holger, Kress Holger, Laforsch Christian.

Research efforts and public attention on microplastic pollution was exponentially increasing during the last years due to the observed variety of effects on an organismal and environmental level. Plastic introduced to the environment undergoes processes of degradation and fragmentises down to microplastic. Furthermore, the colonisation by microbes, together with biomolecules, forms an ecocorona on microplastic particle surfaces that enhances the ingestion of microplastic by organisms. Once ingested there is evidence that microplastics harm organisms and translocate in tissues causing histological changes and inflammatory responses. The reason for cellular internalisation is unknown, since this has only been shown for specifically surface-functionalised particles. The translocation of environmentally relevant microplastics into tissues may occur via the paracellular or transcellular pathway. Paracellular transport occurs in between cells by gap junctions for instance, whereas the transcellular transport occurs via the internalisation into cells directly. Due to the size of microplastics the transcellular pathway into tissues seems more realistic. Therefore, we investigated the cellular internalisation of environmentally relevant microplastic particles. We show for the first time, that the exposure of microplastic particles in the aquatic environment significantly enhances the internalisation into macrophages compared to pristine microplastic particles. We identified biomolecules forming an ecocorona on the surface of microplastic particles from both, fresh- and saltwater by using scanning electron microscopy, µ-Raman spectroscopy, and X-ray photoelectron spectroscopy. Our findings suggest that the environmental exposure of microplastic particles promotes their cellular internalisation. Our results provide new insights that the transcellular pathway via cellular internalisation is a key route by which microplastic particles can translocate into tissues. These findings will help to better understand the risks deriving from plastic pollution.

Keywords : biofilm , cellular internalisation , cellular uptake , ecocorona , microplastic , Raman , translocation , XPS

#### Session 24.1\_Ma. Chaired by Martin Wagner, Trondheim

Microplastic contamination of packaged meat: occurrence and associated risks

Kedzierski Mikaël, Lechat Benjamin, Sire Olivier, Le Maguer Gwénaël, Le Tilly Véronique, Bruzaud Stéphane.

Paper number 332471

Nanoplastics toxicity to *Philodina roseola* a fresh water rotifer *Natarajan Chandrasekaran*. Paper number 332563

### Nanoplastics accumulate in zebrafish liver cells and larvae after an acute exposure but do not affect larval survival after bacterial infection

Brandts Irene, Garcia-Ordoñez Marlid, Tort Lluís, Teles Mariana, Roher Nerea. Paper number 333145

### Development of a novel technological approach for removal of microplastics from water – comparison of environmental factors and different polymer types.

Strum Michael, Schuhen Katrin. Paper number 331996 **Microplastic contamination of packaged meat: occurrence and associated risks** Kedzierski Mikaël, Lechat Benjamin, Sire Olivier, Le Maguer Gwénaël, Le Tilly Véronique, Bruzaud Stéphane.

Food trays, one of the most commonly used packaging, are often made from extruded polystyrene (XPS). Recently, quantities of millimetre-sized particles of this material are trapped between the meat they contain and the sealing film. The purpose of this study is to identify the chemical nature of these particles and to quantify them. For this purpose, this study focuses on meat products (chicken) packed in extruded polystyrene trays (230x140x20mm). Products from four different brands (named A, B, C, and D) were purchased at a local supermarket (n=3 by brand). A dissection microscope (30X magnification) was used to count fibres and potential MP-XPS. The spectra of all recovered fragments were acquired using an Attenuated Total Reflection Fourier Transform Infrared microspectrometer (ATR-FTIR Lumos, Bruker). The results showed that XPS microplastics (MP-XPS) contaminate food products at a level ranging from 4.0 to 18.7 MP-XPS/kg of packaged meat. Analysis showed that these microplastics were likely to come from the XPS trays. These particles are difficult to remove by mere rinsing and are probably cooked before being consumed. However, at this stage, it is not clear from the scientific literature whether there is a potential risk to humans associated with the ingestion of MP-XPS. Based on the average French yearly consumption of meat, the mass of XPS observed inside the packaging and potentially ingested per year was calculated, the potentially ingested mass of XPS could reach a maximum between 19.7 mg/y and 511 mg/y. Only a few scientific studies report the presence of microplastics that contaminate food. This study, by assessing the presence of microplastics on the surface of meat products, extends the state of knowledge on the contamination of human food by microplastics.

Keywords : contaminant , extruded polystyrene , fibres , human alimentation , Microplastics , packaging

#### Nanoplastics toxicity to *Philodina roseola* a fresh water rotifer

Natarajan Chandrasekaran.

Micro and nanoplastics are emergent pollutants of the modern world. Since the finding of new synthetic polymers and making into different usable products, its durability has made it so attractive that it is being used in many consumer products. The weathering of plastics leads to microplastics and then to nanoplastics. Moreover these particles acts as a vector for other pollutants, by carrying other xenobotics like heavy metals and pesticides, at the same time these particles haing other addtives inherently like bisphenol, phthalates and other organics will also get leached during weathering process. From house hold drains to treatment plants and finally to aquatic system. ultimately reaching organisms dwelling there. Rotifers forms one of the basic component in the aquatic food chain. Nanoplastics of two different sizes 50 and 100nm polystyrene spherules were treated to Philodina roseola a fresh water rotifer. The LC50 for 100 and 50 nm polystyrene spherules were 22.94 ppm and 16.36 ppm respectively. Rotifers caused diapauses and failed to produce F2 generation. The biochemicals and antioxidant assays decreased when the concentration of nanoplastics increased. Eggs treated with nanoplastics showed 89 % mortality at 72 hrs with LC50 of 14.72 ppm, no hatching was observed upto 180 minutes, all the eggs went into diapause. After 180 minutes neonates developed leading to F1 generatio. The proportion of neonates transforming into aduls was least. Finally the adults could not gave raise to F2 generation. Nanoplastics has an impact on to fresh water rotifers.

Keywords : generation , nanoplastics , nanospherules , Philodin roseola , polystyrene , toxicity

# Nanoplastics accumulate in zebrafish liver cells and larvae after an acute exposure but do not affect larval survival after bacterial infection

Brandts Irene, Garcia-Ordoñez Marlid, Tort Lluís, Teles Mariana, Roher Nerea.

The presence of small-sized plastic particles in marine and freshwater environments is a global problem but their long-term impact on ecosystems and human health is still far from being understood. Nanoplastics (and ability to penetrate living organisms at any trophic level. Few studies evaluate the impact of nanoplastics in vivo on the immune system of aquatic organisms, while most of them assessed the impact on indirect markers of immune response such as regulation of gene expression, ROS production or DNA genotoxicity. Moreover, the study of the effects of nanoplastics on aquatic vertebrate species in vivo is still scarce. We seek to shed light on the underlying effects of polystyrene nanoplastics (PS-NPs) on the immune response in a model fish species (Danio rerio, zebrafish) after an acute exposure, with a combination of in vitro and in vivo experiments. Our results show that PS-NPs (65 nm) are efficiently taken up by zebrafish liver cells, accumulating mainly in lysosomes. Furthermore, the expression of immune genes presents a synergy when cells were simultaneously exposed to PS-NPs, at a low dose and early time point (12 h) and challenged with a viral stimulus (poly(I:C)). Moreover, zebrafish larvae also internalize PS-NPs, accumulating them in the gut and pancreas. However, at concentrations of up to 50 mg/L in an acute exposure, PS-NPs do not interfere with the survival of the larvae after a lethal bacterial challenge (Aeromonas hydrophila). This study addresses the relevant environmental question of whether a living organism exposed to PS-NPs can cope with a real immune threat. We show that, although PS-NPs induce an immune response, the survival of zebrafish larvae challenged with a bacterial infection after exposure to PS-NP is not decimated with respect to unexposed larvae.

Keywords : immune system , infection , nanoplastics , zebrafish , ZFL

#### **Development of a novel technological approach for removal of microplastics from water – comparison of environmental factors and different polymer types** Strum Michael, Schuhen Katrin.

An increasingly serious and widespread problem is the introduction of plastics into the water cycle. The poor degradability leads to the plastic waste remaining in the water for a long time and over time it fragments into smaller and smaller plastic particles, so called microplastics. Fields of application are microplastic sensitive seawater using process as membrane based sea water desalination, where microplastics poses a risk of membrane fouling or near food chain processes as sea salt production [1,2]. Another example is municipal and industrial wastewater, which is an important point source for microplastics in the environment. In a comparative study the effect of different types of water on and temperatures the fixation process using the three in previous studies best performing organosilanes were examined and compared with our results of further lab scale studies . We compared rinsed municipal wastewater, seawater and demineralized water at temperatures ranging from 7.5 - 40 °C. The residues of the organosilanes remaining in the water after the fixation process were monitored using ICP-OES and DOC measurements. Due to the fact, that microplastic encompasses a multitude of different types of polymers with different properties and surface properties, we compared the efficiency of the process for polyethylene, polypropylene, polyamide, polyester and polyvinylchloride as examples for common polymer types with different heteroatoms and polarities. The results will be presented in our talk. References [1] K. Schuhen, M.T. Sturm, Microplastic Pollution and Reduction Strategies, in: T. Rocha-Santos, M. Costa, C. Mouneyrac (Eds.), Handbook of Microplastics in the Environment, Springer International Publishing, Cham, 2020, pp. 1–33. [2] K. Schuhen, M.T. Sturm, A.F. Herbort, Technological Approaches for the Reduction of Microplastic Pollution in Seawater Desalination Plants and for Sea Salt Extraction, in: A. Gomiero (Ed.), Plastics in the Environment, IntechOpen, 2019.

Keywords : agglomeration , microplastics removal , organosilanes , seawater , sol gel chemistry , wastewater

#### Session 24.2\_O. Chaired by Chelsea M. Rochman, Toronto

Water assessment in the Bunaken-Tangkoko- Minahasa Biosphere Reserve proposal site and its impact to improve participatory management and co-creation of knowledge *Hernandez-Salinas Alberto*. Paper number 334501

**Microplastic pollution in surface water of the Lower Weser and Weser estuary** *Roscher Lisa, Halbach Maurits, Scholz-Böttcher Barbara, Gerriets Michaela, Shiravani Gholamreza, Primpke Sebastian, Gerdts Gunnar.* Paper number 334524

**Beached microlitter on the lagoon coasts of the Russian part of the Baltic Sea** *Ershova Alexandra, Eremina Tatjana, Khatmullina Lilia, Esyukova Elena, Chubarenko Irina, Gyraite Greta.* Paper number 334590

**Meso to micro: Plastics in sandy beaches of Santa Catarina Island, Brazil** *Zanetti Daniela, Leonel Juliana, Andreussi Camila.* Paper number 334597

#### Water assessment in the Bunaken-Tangkoko- Minahasa Biosphere Reserve proposal site and its impact to improve participatory management and cocreation of knowledge

Hernandez-Salinas Alberto.

Polluted urban storm water runoff, industries spills and waste, and sediment loading and solid waste transport into water bodies of great rivers. These have direct impacts on the quality of surface waters and groundwater. Emerging pollutants present new water quality challenges (UN-water report 2018). Rivers and lakes can play the role of green and blue corridors to let the biodiversity moves and become more resilient. By improve the ecological resilience and the environmental health, we will ensure a better quality of their ecosystem services that prevent human population diseases, mitigate climate change impact and soon. Under a landscape perspective, this study will try to provide a general view of the existing knowledge of the water, including surface and groundwater. It is essential to understand the water situation in the Biosphere Reserve and to integrate it in the management of this site as a holistic approach. The study will be carried out within the framework of the nomination of the Bunaken-Tangkoko-Minahasa Biosphere Reserve (BR) (North Sulawesi, Indonesia). The process of recognition of the BR has been delayed as a result of the global pandemic by COVID-19. It comprises an area of 746,412.54 Ha that contributes significantly to the conservation of the landscape, ecosystems and biodiversity at a regional scale. It is made up of five core areas that together encompasses the variety of highly biodiverse tropical coastal and marine ecosystems within the Wallacea Biodiversity Hotspot. This site provides a very suitable place to test novel management for the conservation of natural resources, including water, as well as sustainable practices at the regional scale (MAB Indonesia-LIPI, 2019). It will highlight local activities from different sectors to promote co-knowledge and community of practices (Onaindia et al., 2020) and University cooperation.

Keywords : Biosphere Reserve , co-creation of knowledge , North Sulawesi , Water body

Microplastic pollution in surface water of the Lower Weser and Weser estuary

Roscher Lisa, Halbach Maurits, Scholz-Böttcher Barbara, Gerriets Michaela, Shiravani Gholamreza, Primpke Sebastian, Gerdts Gunnar.

Microplastics (MPs) are defined as synthetic organic pollutants with a size ; 5 mm, and have been recorded in various environments across the world. Due to their small size they pose a potential risk for many organisms throughout the food web. Yet, little is known about MP-distribution patterns and associated transport mechanisms into the oceans. River systems may act as pathways for MPs into marine environments, and especially the transition zone between these two environments is of great importance. In this study, we investigated the estuary and lower stretch of the second-largest German River Weser for the occurrence of MPs, being an important interface of fresh water and marine environments. Hereby, we are aiming at enhancing the general understanding by providing novel, comprehensive data and suggestions for future studies on riverine systems. Surface water samples of two different size classes (11-500 µm and 500-5000 µm) were collected by means of an on-board filtration system as well as net sampling. After a thorough sample preparation using state-of-the-art methods, MP samples were analyzed with µFTIR as well as FTIR-ATR in order to obtain information on concentrations, polymer composition and size distribution. Highest concentrations of MP were revealed in the sample fraction 11-500 µm, with the polymer cluster acrylates/PUR/varnish being dominant. Further, data showed that MP concentrations generally decreased from the river mouth towards the open sea. In the sample fraction 500-5000 µm, MP concentrations were lower, with polyethylene as dominant polymer type. This study contributes to the current MP research by providing novel insights into MP pollution of the estuary and lower stretch of an important German River, and provides implications for future MP monitoring and conservation measures.

Keywords : infrared spectroscopy , microplastics , river systems , surface water sampling

**Beached microlitter on the lagoon coasts of the Russian part of the Baltic Sea** Ershova Alexandra, Eremina Tatjana, Khatmullina Lilia, Esyukova Elena, Chubarenko Irina, Gyraite Greta.

The coasts of the largest inland marine areas (lagoons) of the Russian part of the Baltic Sea were studied in summer 2018-2019. "Marine litter sampling method" (IOW) was tested on the sandy beaches, including the assessment of macro-, meso- and large microlitter (¿ 2 mm) in the wreckline zone. This study was supported by the RFBR grant 18-55-76001, project "Litter rim of the Baltic coast: monitoring, impact and remediation" of the Project "ERA.Net Rus Plus". In total, 14 beaches in the Kaliningrad region and 7 beaches in the Neva Bay (St. Petersburg) were examined, over 200 sand samples were analyzed where objects of anthropogenic origin were found (cigarette filters, ear sticks, foamed plastic, fragments of bricks/ceramics, plastic pellets and fragments, glass, rubber, metal). Plastic waste amounted to 67% on the beaches of the Kaliningrad region and up to 80% of the composition of microparticles in the NevaBay. The average concentrations of anthropogenic microlitter on the beaches of the Kaliningrad region were: Curonian Lagoon and Vistula Lagoon - 16 items/m2 and 6.3 items/ m2 of microlitter respectively. In the NevaBay, the concentration of marine litter is much higher: from 25-55 items/m2 of microlitter on the northern coast to 6-15 items/m2 on the southern coast, with microplastics dominating. In general in 2018-2019 the largest amount of litter of all fractions is found in the inner part of the estuary in the NevaBay despite the regular municipal cleanings of beaches. The remoteness of the beach from settlements is not associated with the amount of litter on the beaches. The main critical factors of litter migration in the coastal zone are wind speed and direction, wave heights, and shore exposure to wind. The studies confirmed the Frame method is applicability on the coasts of the Baltic Sea lagoons, but with limitations.

Keywords : Baltic Sea , beach litter , lagoons

#### Meso to micro: Plastics in sandy beaches of Santa Catarina Island, Brazil

Zanetti Daniela, Leonel Juliana, Andreussi Camila.

Microplastics (MPs) and mesoplastics (MSs) have received considerable attention due to their high dispersibility and accumulation capacity in various environments. They may pose a risk to biota due to their size and consequently ingestion as food. Moreover, MPs are also capable of concentrating contaminants. MPs and MSs were sampled in ten sandy beaches along Santa Catarina Island (Brazil) to study spatial distribution and composition. All particles were analyzed for shape, color, size and degradation stage by processing images obtained by a binocular microscope. Concentration of MPs ranged from 1.33 to 127.3 particles m-2 in the strandline and from 0 to 37.33 particles m-2 in the backshore portion. Concentration of MSs ranged from 0 to 33,32 particles m-2 in the strandline and from 0 to 11,44 particles m- in the backshore portion. Natural characteristics of each beach, such as exposure to wind and wave energy, appeared to be as important for MP and MSs occurrence as anthropogenic factors.

Keywords : coastal pollution , pellets , plastic , weathering processes

#### Session 24.2\_Me. Chaired by Stefan Peiffer, Bayreuth

Are biodegradable polymers a solution to the microplastic issue? Agarwal Seema. Paper number 334142

**Occurrence of microplastics in the hyporheic zone of rivers.** *Frei Sven, Piehl Sarah, Gilfedder Benjamin, Löder Martin, Laforsch Christian.* Paper number 334135

### Comparing the effects of multiple microplastic polymer types and shapes on the earthworm *Eisenia fetida*

*Holzinger Anja, Hink Linda, Rothmaier Melanie, Döring Max, Horn Marcus, Feldhaar Heike.* Paper number 334379

#### Are biodegradable polymers a solution to the microplastic issue?

Agarwal Seema.

The stability of polymers against chemicals, hydrolysis, temperature, light, and microbes has challenged society to accumulate plastic waste and its management worldwide. Large amounts of plastic litter accumulate in the environment and disintegrate into microplastic (MP) (small pieces in size less than 5mm), a topic of actual concern for products and applications where the plastics are used for a short time before becoming waste and where they are difficult to recover after use and stay/arrives in the environment. Can biodegradable polymers be one of the solutions to the problem of plastic waste/microplastic issue is a question very often raised in this context. Although the use of biodegradable polymers appears to be highly promising based on recent and past studies, but several aspects need to be considered further regarding environmental sustainability, acceptability, and degradability in the complex natural environment. Intensive efforts need to be invested in developing new, environmentally degradable biodegradable polymers and smart mechanisms of degradation after use in the environment. I want to discuss in the talk the potential role of the classified biodegradable polymers in creating and solving the microplastic issue, their impact on the environment based on the experimental data.

Keywords : Biodegradable polymers , microplastic , synthesis

#### **Occurrence of microplastics in the hyporheic zone of rivers**

Frei Sven, Piehl Sarah, Gilfedder Benjamin, Löder Martin, Laforsch Christian.

Although recent studies indicate that fluvial systems can be accumulation areas for microplastics (MPs), the common perception still treats rivers and streams primarily as pure transport vectors for Mps. In this study we investigate the occurrence of MPs in a yet unnoticed but essential compartment of fluvial ecosystems - the hyporheic zone (HZ). Larger MP particles (500–5,000 µm) were detected using attenuated total reflectance (ATR) - Fourier-transform infrared (FTIR) spectroscopy. Our analysis of MPs(500–5,000 µm) in five freeze cores extracted for the Roter Main River sediments (Germany) showed that MPs were detectable down to a depth of 0.6 m below the stream bed in low abundances ( $\ll 1$  particle per kg dry weight). Additionally, one core was analyzed as an example for smaller MPs (20–500 µm) with focal plane array (FPA)- based µFTIR spectroscopy. Highest MP abundances (30,000 particles per kg dry weight) were measured for pore scale particles (20–50 µm). The detected high abundances indicate that the HZ can be a significant accumulation area for pore scale MPs (20–50 µm), a size fraction that yet is not considered in literature. As the HZ is known as an important habitat for invertebrates representing the base of riverine food webs, aquatic food webs can potentially be threatened by the presence of Mps in the HZ. Hyporheic exchange is discussed as a potential mechanism leading to a transfer of pore scale MPs from surface flow into stream bed sediments and as a potential vector for small MPs to enter the local aquifer. MPs in the HZ therefore may be a potential risk for drinking water supplies, particularly during drinking water production via river bank filtration.

Keywords : freshwater systems , hyporheic zone , microplastic , rivers and streams

# Comparing the effects of multiple microplastic polymer types and shapes on the earthworm *Eisenia fetida*

Holzinger Anja, Hink Linda, Rothmaier Melanie, Döring Max, Horn Marcus, Feldhaar Heike.

Public concerns of microplastic pollution have spread from aquatic environments to terrestrial ecosystems. Large amounts of microplastics have recently been detected in terrestrial environments. Until now, the effects of microplastic on the soil fauna still remain largely unexplored. Microplastics occur in various shapes, sizes and polymer types. The influence on soil organisms might depend on one of these parameters. In particular, the earthworm Eisenia fetida is likely to come into contact with microplastics while foraging. Therefore, we assessed the direct effects of three representative polymer types (PS, PA, and the biodegradable PLA) as particles and fibres (2% v/v in soil) on the life-history and oxidative stress level of the earthworm E. fetida over a period of 56 days. The ecotoxicological responses were evaluated by analysing various oxidative stress biomarkers to uncover potential direct effects on the earthworm. Also, the gut microbiome plays an important role in an organism's health. By quantifying the gut microbiome composition and activity we studied the indirect effects of microplastic exposition on the worm. As studies have shown that fibres cause severe tissue damage and increase oxidative stress, we hypothesize that worms exposed to microplastic fibres will have a reduced growth rate, reproductive fitness and health. In addition, different polymer types can cause shifts in the composition of the gut microbiome. The findings will be beneficial in estimating the effect, mechanism and hazard potential of microplastics of a specific shape and polymer on soil biota.

Keywords : Earthworms , gut microbiome , microplastic fibres , microplastic fragments , oxidative stress , soil macrofauna

#### Session 24.2\_Ma. Chaired by Andy Booth, Trondheim

**Occurrence of microplastics in filet and organs of farmed and wild salmon** *Gomiero Alessio, Haave Marte, Bjorøy Ørjan, Herzke Dorte, Kögel Tanja, Nikiforov Vladimir, Øysæd Kjell Birger.* Paper number 334370

**Critical evaluation of Nile Red for the detection of Microplastics in environmental samples** *Stutzinger Laura, Primpke Sebastian, Mackay-Roberts Nicholas, Gerdts Gunnar.* Paper number 334411

#### Can polystyrene nanoplastics be hazardous to polychaetes?

Silva Marta, Oliveira Miguel, Figueira Etelvina, Martins Manuel, Pires Adília. Paper number 334442

### Microplastics in transitional systems, occurrence in fish aquaculture and potential impacts in human health

Rocha Carolina, Marques João Carlos, Gonçalves Ana Marta. Paper number 334474 **Occurrence of microplastics in filet and organs of farmed and wild salmon** Gomiero Alessio, Haave Marte, Bjorøy Ørjan, Herzke Dorte, Kögel Tanja, Nikiforov Vladimir,

Øysæd Kjell Birger.

Microplastic is of growing concern to environmental and consumer health. This study aimed at quantifying MP in a relevant selection of tissues of farmed and wild salmon to establish likely indicator organs for future documentation purposes. Three analytical methods were tested for the characterization of microplastic (MP). Vibrational spectroscopy-microscopy and thermal degradation mass spectrometry were successfully applied to detect and quantify MP in the fish tissues. Highresolution mass spectrometry (HRMS-Orbitrap) was successful in quantifying phthalates and organophosphorus flame retardants indicating plastic contamination in tissues. 14 pooled samples of muscle and 14 pooled samples of liver from 70 fish were altogether analyzed. The outcomes of the present study point plastics contamination present in both wild and farmed salmon as well as wild mountain lake trout. Particles of minor dimension length 11-40 µm are most prevalent. The most frequently detected polymer types are PS, PP and PE, some of which were also detected in wild trout from remote mountain lakes. There are no observed differences between MP in liver and muscle tissues, per 100 g. Therefore, from the data of this study it is not possible to recommend one over the other for surveillance or effect studies. For monitoring purposes, muscle samples are routinely obtained, and may therefore be suitable for further investigation of the occurrence of MP in fish. Muscle samples may also be used for investigation of the correlation between exposure and uptake of MP.

Keywords : GCMS , human health , Microplastics , pyr , salmon filet , seafood safety ,  $\mu FTIR$ 

# **Critical evaluation of Nile Red for the detection of Microplastics in environmental samples**

Stutzinger Laura, Primpke Sebastian, Mackay-Roberts Nicholas, Gerdts Gunnar.

The widespread use of plastics in this day and age has resulted in the accumulation of these synthetic polymers in the environment. Upon exposure to physical forces and UV radiation, larger plastic debris have been found to break down into smaller fragments known as microplastics (MP). The pervasive nature of MP and their potential impacts on the biosphere have become cause for growing concern. In an effort to better understand the occurrences of MP in nature, several techniques have been employed throughout existing studies for the detection and identification of MP. Among the most commonly used approach is visual sorting. However, as synthetic and non-synthetic materials can be difficult to distinguish visually, this approach is prone to human error and can result in the misreporting of MP concentrations in nature. As such, this can be circumvented through the use of a staining dye Nile Red (NR). In this study, a critical evaluation of NR staining coupled with quantitative fluorescence imaging as an easy-to-use detection method for MP.

Keywords : environmental samples , Nile Red staining , quantitative fluorescence imaging

#### Can polystyrene nanoplastics be hazardous to polychaetes?

Silva Marta, Oliveira Miguel, Figueira Etelvina, Martins Manuel, Pires Adília.

Plastics are recognized as persistent pollutants in aquatic environments. Their increased production and environmental release raised concerns about their impact on marine life. Polystyrene (PS) is among one of the most frequently found in the environment since many single use products are made with this polymer. PS, like other polymers, under environmental conditions becomes brittle and breaks down into smaller particles, becoming more available to biota and thus threatening organisms in the water column and sediment, as they gradually sink to the sediments. Polychaetes are the most abundant group in marine ecosystems and create conditions for a more diverse fauna in sediments. As benthic organisms they are exposed to contaminants present in the sediment and as well as in the water. Thus, this study aimed to assess the effects of PS nanoplastics (PS NPs) on biochemical endpoints associated with oxidative status and damage, behavior and regenerative capacity of Hediste diversicolor. H. diversicolor specimens were collected at a reference site in the Aveiro lagoon (Portugal), and after depuration and acclimatization, they were exposed, for 28 days, to five different concentrations of 100 nm PS NPs (0.0; 0.005; 0.05; 0.5; 5.0; 50.0 mg/L). Results demonstrated the capacity of PS NPs to impact the activities of important antioxidant enzymes such as glutathione peroxidase (0.005 and 0.05 mg/L) and catalase (0.05 to 50 mg/L), and enzymes of phase II of biotransformation glutathione S-transferases (0.05 to 50 mg/L). The ability of these particles to induce oxidative damage was also observed. Protein oxidation occurred in organisms exposed at 0.05 to 50 mg/L. Effects were also reported at the individual level. Organisms took longer to bury (0.005 to 0.5 mg/L) and presented a decreased body regeneration (0.005 and 5 mg/L). Overall, the results highlight that PS NPs may have an impact on the population of polychaetes.

Keywords : Biomarkers , Body regeneration , Burrowing , Effects , Invertebrate , Waterborne nanoplastics

### Microplastics in transitional systems - occurrence in fish aquaculture and potential impacts in human health

Rocha Carolina, Marques João Carlos, Gonçalves Ana Marta.

For the past decades, the ubiguity of plastic pollution has raised the concern of all layers of the society towards the consequences of this synthetic material in the environment. However, the focus of the scientific community concerning plastic pollution has mainly turned to marine environments. while other environments have been disregarded. Estuaries are particularly pressured by polluting human activities. Aquacultures are among the main activities in estuaries and are quickly expanding. to match the market demand for alternatives to capture fisheries. Fish production is particularly important in Southern European countries, and in some, as Portugal, fish are reared in semi-intensive regimes, using estuarine water for the rearing tanks. The water is kept in the tanks roughly two weeks, after which it is released to the estuary, while water with lower matter load is brought in. Due to the worldwide importance of fish as a food source and given the particularities of semi-intensive practices, there is an interest in understanding the pressure of plastic pollution in these systems. Thus, the present study aims to understand the path of plastic input in estuaries, particularly in aquacultures. Therefore, water, sediment and fish were sampled from four Portuguese aquacultures, located in two estuaries – Mondego and Sado –, subjected to different polluting pressures. Results show i) water released from the rearing tanks carries higher microplastic load, compared to incoming water; ii) water from the Sado estuary, which supports more human activities compared to the Mondego estuary, presents a higher count of microplastic particles; iii) sediments enclose microplastics of higher density, that sink due to low water dynamics; iv) fish tissue accumulates different types of microplastics. This study provides further supporting material to enhance strategies to food safety and security and to the economic development, highlighting the potential impacts to human diet and health.

Keywords : Aquaculture , Contamination , Estuary , Fish , Human Health , Plastic

#### Session 24.3\_O. Chaired by Elke Brandes, Braunschweig

**BMBF Plastics in the environment. Sources and pathways of micro-plastic in inland, coastal and marine waters** *Knoblauch Doris, Brandes Elke.* Paper number 334623

### Numerical modelling of microplastic interactions with fine sediments in the Weser estuary and the German Wadden Sea

*Shiravani Gholamreza*, *Oberrecht Dennis*, *Wurpts Andreas*. Paper number 332648

#### From Land to Sea. Model for the documentation of land-sourced plastic litter

*Cieplik Stephanie.* Paper number 334295

#### **Model based estimation of regional microplastic emissions from agriculture in Germany** *Henseler Martin, Brandes Elke, Kreins Peter.* Paper number 333888

# **BMBF** Plastics in the environment - Sources and pathways of micro-plastic in inland, coastal and marine waters

Knoblauch Doris, Brandes Elke.

In the past, microplastic (MP) research has mainly focused on marine systems. The land-based discharges of MP into the oceans have been poorly studied so far, although they are assumed to represent an important contribution to the marine MP pollution. The significance of the various MP sources, sinks, pathways, and corresponding transport mechanisms has not been systematically assessed to date. Due to the methodological challenges, MP analysis will continue to provide mainly point information in the foreseeable future. To quantify the various MP sources, pathways and fates within and across ecosystems, data-driven models present crucial tools to connect field work results, scale up transport and movement processes to a landscape level, and simulate effects of mitigation measures. This session will give an insight into several spatial and temporal MP modelling approaches covering three exemplary river catchments in Germany that, as a whole, provide a framework to improve our systems understanding of the sources, pathways and fates of MP in the environment. Scope, boundaries, and mechanisms of the models, state of model development, first results, as well as data deficiencies and missing links will be presented and discussed. The aim is to highlight how field work analysts and modelers can shape their collaboration in the future to streamline mutual information exchange and create synergies between the disciplines.

Keywords : analysis , hotspots , modelling , models , MP sources , pathways , sampling , sinks

#### Numerical modelling of microplastic interactions with fine sediments in the Weser estuary and the German Wadden Sea

Shiravani Gholamreza, Oberrecht Dennis, Wurpts Andreas.

Plastic litter has drastically increased since 1950s to around 359 million tons in 2018 (Plastic-the facts, 2019). Microplastic (MP) is an important part of plastic litter, which is present in marine waters and rivers. MP due to its small size (< 5 mm diameter) is more problematic than other plastic litter to the ecosystems and organisms. Therefore, extensive research is conducted to improve the understanding of MP effects, distribution, pathways, sinks and sources. However, quantitative studies on the MP-transport along rivers and estuaries are rare. Sediments are one of the major sinks of MP in rivers and marine waters. In addition, sediments could force the floating MP to settle and change their pathway and physical properties; especially when cohesive fine sediments at higher concentrations are involved (e.g. estuarine turbidity zone). MP found in bottom sediments, particularly fine MP of light polymers (e.g. PE, PP) pronounce the importance of further research on MP-sediment interaction. Above all, floating MP like PE and PP are the most frequently used polymers in everyday life, and therefore the main polymer-type of plastic litter in contact with suspended fine sediments. To understand the fate of MPs and associated transport mechanisms more accurately, a BMBF(Federal Ministry of Education and Research of Germany) supported research program about MP- contamination in the Weser river catchment and the German Wadden Sea (PLAWES) is currently under investigation. As a part of the PLAWES model chain, an Eulerian model approach for the overall estuarine MP-budget is set up and, among others, sediment-MP interaction is developed. All processes are implemented into a coupled hydro-morphodynamic model (using Delft3D) with water quality and ecology module (using Delft3D-WAQ). The presentation shows results of the numerical modelling of MP-transport with a focus on MP-fine sediment interaction and its effects on the fate of MP under estuarine dynamical conditions.

Keywords : fine sediment , Microplastic , numerical modelling

#### **From Land to Sea - Model for the documentation of land-sourced plastic litter** Cieplik Stephanie.

The model From Land to Sea – Model for the documentation of land-sourced plastic litterwas developed on behalf of BKV GmbH, Frankfurt, Germany. This model systematically records for the first time discharges of improperly disposed-of plastic litter from Germany that gets into the North Sea, the Baltic Sea and the Black Sea. All discharge pathways and sources are taken into account. A distinction is made between discharges of microplastic and macroplastic. The main emphasis of this project is on the collection, compilation and processing of facts and data, especially with regard to mass flows and the routes by which the plastics are transported into the sea. The aim of the project is, based on the methodical approach, to systematically record, structure and quantify the main discharge pathways and sources for plastics. Only if the discharge pathways and sources as well as the corresponding mass flows of the plastics into the sea are identified and analysed, it will be possible to make a useful contribution to the prevention of further input of litter into the seas. The report and handbook belonging to the model are continuously updated and further revised. In addition to continuously adapting the above-mentioned framework conditions, the model parameters are also checked and, wherever necessary, modified. The evaluation of the model is accompanied by consultations with external experts. The open structure of the methodology makes it possible to also apply it to other regions or countries and to incorporate further discharge pathways. The advantage of the model lies, in particular, in the easy and flexible adaptation of variables and calculations. The aim is, therefore, to transfer the model to other regional conditions. The report and handbook for the model are available in German and English and can be obtained free of charge via the BKV website.

Keywords : Discharge pathways/discharge sources , Germany's contribution , Microplastic/Macroplastic , New methodology , Volume estimate

# Model based estimation of regional microplastic emissions from agriculture in Germany

Henseler Martin, Brandes Elke, Kreins Peter.

The topic of microplastic (MP) contamination in agricultural soils has recently gained attention in science and society. Experimental studies indicate that microplastic (i.e., plastic particles ; 5mm) can have negative effects on soil physical properties and ecology, but an actual impairment of soil functions at current concentration levels in agricultural soils has yet to be shown. The most discussed agricultural sources for MP contamination of cropland are biosolids (sewage sludge and compost) applied as soil amendment to fields, as well as plastic film used in plasticulture. However, knowledge about how much MP has been accumulating in agricultural soils is scarce. Due to methodological challenges, MP analysis of field samples will continue to provide mainly point information in the foreseeable future. To quantify the various MP sources and pathways within and across ecosystems, data-driven models present crucial tools to scale up these analytic results to a landscape level and to simulate effects of mitigation measures. Some recent modelling studies have estimated MP emissions based on production and consumption statistics at national level. However, spatially explicit regional quantification of microplastic immissions into agricultural soils are virtually missing in the scientific literature. Using data on MP concentrations in biosolids from the literature in combination with national and regional statistics on sewage sludge, compost and organic waste production, as well as specialty crop areas, we estimated the spatial distributions of MP mass contents in agricultural soils in Germany originating from agricultural activities. Although these estimates are based on limited data availability, our results provide first indications about locations where detailed soil analysis could be useful to investigate in situ processes and impacts. The methodology can be applied to other regions and continuously adapted when more knowledge on relevant sources, transport, accumulation, and degradation rates of MP in soils will be gained in the future.

Keywords : agriculture , land based emissions , modelling , regional distribution , soils

#### Session 24.3\_Me. Chaired by Seema Agarwal, Bayreuth

### Tracing the lateral transport of microplastic particles with an advanced-imaging sCMOS camera

*Laermanns Hannes, Klee Marcel, Steininger Florian, Löder Martin, Bogner Christina.* Paper number 334274

### From macro- to microplastic: Accelerated weathering of polystyrene by simulated solar radiation and mechanical stress

Meides Nora, Menzel Teresa, Pötzschner Björn, Strohriegl Peter, Altstädt Volker, Senker Jürgen. Paper number 334281

### Deeper insights: The potential of X-ray micro-computed tomography ( $\mu$ CT) in microplastic studies

Löder Martin G. J., Wittmann Marie, Teichert Sebastian, Schulbert Christian, Oster S. D. Jakob, Laforsch Christian. Paper number 334425

#### Microplastic as spreader of fungal pathogens

Gkoutselis Gerasimos, Rohrbach Stephan, Harjes Janno, Brachmann Andreas, Horn Marcus, Rambold Gerhard. Paper number 334558
# Tracing the lateral transport of microplastic particles with an advanced-imaging sCMOS camera

Laermanns Hannes, Klee Marcel, Steininger Florian, Löder Martin, Bogner Christina.

The impact of microplastic particles (MPs) in different ecosystems has recently become subject of numerous studies. However, the research of the last years has focused mainly on marine ecosystems and neglected terrestrial environments so far. This has led to a substantial lack of knowledge about the transport mechanisms of MPs in soils and sediments. While first studies in this field investigated the abundance of MPs in soils, only little is known about their surface transport. In this study we investigate surface transport mechanisms and patterns through a new approach using images of advanced scientific complementary metal–oxide–semiconductor (sCMOS) high-resolution camera. The experimental set-up includes a flume box with an inclined surfaces of different roughnesses and an artificial irrigation system installed at the top end. Transport pathways of MP are studied under different irrigation rates, inclinations and surface roughnesses using fluorescing amorphously shaped polystyrol particles. Time series of the images are analysed using a combination of R and Python packages (Hardy et al., 2017. https://doi.org/10.1016/ j.catena.2016.11.005). Preliminary results suggest that the MPs are transported along preferential pathways depending on the microrelief. While higher inclination probably leads to a shift from laminar to turbulent flow, increased irrigation rates seem to have only a minor influence on the MP transport.

Keywords : flume box , high , polystyrol , resolution camera , surface transport , time series analysis

# From macro- to microplastic: Accelerated weathering of polystyrene by simulated solar radiation and mechanical stress

Meides Nora, Menzel Teresa, Pötzschner Björn, Strohriegl Peter, Altstädt Volker, Senker Jürgen.

To date, the presence of microplastics (MP) is verified in almost every environmental compartment. The fragmentation of macro- into microplastic is rated as one of the most important pathways for secondary MP into the environment. As natural degradation is challenging to monitor over long time scales, artificial weathering represents a promising, but still hardly recognized, alternative. Here, we present a long-term accelerated weathering study on the Polystyrene (PS) grade PS 158 N. Tensile bars were exposed to simulated solar radiation and MP particles (125-200 µm) additionally to mechanical abrasion under laboratory-controlled conditions in a commercial weathering chamber. We studied the mechanical and chemical degradation of PS materials using a variety of analytical techniques such as GPC, SEM, EDX and tensile testing as a function of exposure time. Additionally, solid state 13C MAS NMR spectroscopy was used to investigate chain defects induced by photooxidation. Furthermore, Monte Carlo simulations of the chain scission and crosslinking processes were performed. A quantification of the reaction products as a function of exposure time could be completed. Our results helped us to reconstruct the process of PS degradation, coupling both geometries investigated and methods utilized. We identified two stages, ultimately leading to the formation and degradation of MP. Stage I is dominated by photochemical oxidation in a nearsurface layer. In stage II, the degradation is accelerated by the formation of microcracks and particle rupturing. The ratio and intensity of both environmental stress factors influence the life times of both morphologies dramatically. Small MP particles with high proportions of carboxyl, peroxide and keto groups are constantly released into the environment. Their increasing polarity facilitate biofilm formation and the uptake by and interaction with organisms. We expect this two-stage model to be general, allowing a transfer of the degradation mechanisms to other commodity plastics.

Keywords : chain defects , microcrack formation , Monte Carlo simulations , particle fragmentation , photochemical oxidation , polymer degradation , quantitative 13C MAS NMR spectroscopy , two stage degradation model

# Deeper insights: The potential of X-ray micro-computed tomography (µCT) in microplastic studies

Löder Martin G. J., Wittmann Marie, Teichert Sebastian, Schulbert Christian, Oster S. D. Jakob, Laforsch Christian.

Since the topic of microplastics and the detection of these small plastic particles is becoming increasingly important in science, it is necessary to establish methods that allow for a non-destructive detection of microplastics in research objects. Especially with respect to the detection of environmentally relevant concentrations of microplastics in experimental organisms without destroying their body structures X-ray micro-computed tomography ( $\mu$ CT) is a very promising technique. We report first results of experiments with radiopaque polyurethane microplastics. Ten terrestrial and aquatic invertebrate species were selected as experimental organisms to provide a cross-section of different invertebrate taxa. In nine of the ten organisms used, microplastic particles could be easily detected by  $\mu$ CT, and in most cases the exact position of the plastic in the digestive tract of the individual organisms could also be determined. In animals with a solid cuticle, the method is also well suited for detecting external structures. In order to be able to make more precise statements about the position of the microplastics in the body structure of the animals radio-opaque staining methods are available and have to be tested in future studies. The method described here is particularly suitable for non-destructive detection of microplastics in organisms at low natural microplastic concentrations in experiments on the uptake and trophic transfer of microplastics.

Keywords : detection of environmental relevant MP concentrations in organisms , trophic transfer ,  $\mu CT$ 

### Microplastic as spreader of fungal pathogens

Gkoutselis Gerasimos, Rohrbach Stephan, Harjes Janno, Brachmann Andreas, Horn Marcus, Rambold Gerhard.

Microplastic (MP) has become a pervasive and persistent pollutant in nature, where it constitutes a micro-ecosystem for a diversity of microorganisms, the so-called plastisphere. Through the attachment of microbes, these floating artificial substrates implicate the enrichment and vectoring of potential pathogens, thereby interfering with the ecosystem functioning and threatening human health. However, microbial ecotoxicology of MP has been exclusively investigated in aquatic systems, although soil is the major sink for plastics and the main reservoir for a variety of microbes. Developing countries suffer particularly from environmental plastic pollution. Aggravatingly, destructive fungal pathogens are highly abundant in tropical soils, where they contribute substantially to human morbidity and mortality. Although developing countries in tropical regions, i.e. Subsaharan Africa, are particularly vulnerable to microbial ecotoxicology of MP, epiplastic communities remain alarmingly underexplored in those areas. In fact, no studies of plastic-associated microbial consortia from African environmental samples exist to date, nor molecular studies of the terrestrial plastisphere with a focus on fungi. Here, we grant first insights into the plastisphere of terrestrial MP and explore the role of MP debris as selective micro-habitats of soil fungi within the city boundary of Siava, Kenya. Soil samples were collected from plastic waste accumulations from landfills and MPs separated from soil particles in an interposed subsampling step. ITS metabarcoding and multivariate analysis indicated a diverse fungal plastisphere compositionally distinct from the surrounding soil. Scanning electron microscopy (SEM) revealed massive quantities of plasticattached spores and hyphae. Based on assignment analyses of trait data obtained from various sources, we highlight that soil-borne MP hosts and even concentrates a variety of phyto-, zoo- as well as opportunistic human pathogenic fungi. Our findings emphatically substantiate the need for a deeper and more comprehensive understanding of plastic pollution and its ecological implications in terrestrial ecosystems and for human health.

Keywords : DNA metabarcoding , human pathogens , microbial ecotoxicology , microplastic , plastisphere , soil fungi

# Session 24.3\_Ma. Chaired by Bénédicte Morin and Xavier Cousin, Palavas

Does triclosan adsorption on polystyrene nanoplastics modify the toxicity of single contaminants?

Binelli Andrea, Parenti Camilla Carla, Magni Stefano, Della Torre Camilla. Paper number 333153

# Microplastic distributions in a domestic wastewater treatment plant: Removal efficiency, seasonal variation and influence of sampling technique

Ben-David Eric, Habiby Maryana, Haddad Elias, Hasanin Mahdi, Angel Dror, Booth Andy, Sabbah Isam. Paper number 334421

# A stable isotope assay for determining microbial degradation rates of plastics in the marine environment

Goudriaan Maaike, Hernando Morales Victor, Van Bommel Ronald, Van Der Meer Marcel, Ndhlovu Rachel, Hinnrichs Kai-Uwe, Niemann Helge. Paper number 334483

**Influence of simulated weathering on microsize polypropylene properties and quantification by pyrolysis gas chromatography mass spectrometry** *alajo tania.* Paper number 334602

# Does triclosan adsorption on polystyrene nanoplastics modify the toxicity of single contaminants?

Binelli Andrea, Parenti Camilla Carla, Magni Stefano, Della Torre Camilla.

Physical and chemical properties of nanoplastics make them potential carriers for some environmental contaminants, enhancing their biological effects. Nevertheless, the toxicity caused by pollutant adsorption on nanoplastics is still controversial, depending on the interactions between chemical and physical pollutants, the consequent change in bioavailability, the modification in the intake, transport and accumulation in the organisms and also on the characteristics of contaminants. Among the environmental pollutants, the triclosan is considered as a contaminant of emerging concern for aquatic ecosystems, and its adsorption capability on polymers has been already demonstrated. In this context, we evaluated the combined effects made by 0.5 µm nanobeads of polystyrene and triclosan adsorbed on their surface in comparison with those caused by the single contaminants. The systemic effects of 7-day exposure to nanoplastics, triclosan alone, and to the nanoplastics-triclosan complex have been analyzed employing zebrafish larvae and using a multi-tier approach. Firstly, confocal microscopy evidenced nanobeads ingestion and translocation in several tissues and organs to guarantee the goodness of the exposure results. Behavioral assays were then conducted to highlight larval swimming defects as a 'real-time' readout of the potential effects on the whole organism, while a suite of several biomarkers and functional proteomics were applied to investigate the effects at both cellular and molecular levels. The whole dataset pointed out a clear modification in the toxicological effects of the nanoplastics-triclosan complex in comparison with the single contaminants, proved by opposite behaviours in the larval swimming activity and modulation of diverse protein classes, some of which just related to the behavioural modifications, as well as by different effects on several biochemical endpoints. This means that the interaction between chemical and physical pollutants leads to more complicated responses than additive, synergistic or antagonist models, resulting in a change in toxicity instead of its increase or decrease.

Keywords : adsorption , carrier , combined effects , ecotoxicity , environmental pollutants , exposure , nanomaterials

# Microplastic distributions in a domestic wastewater treatment plant: Removal efficiency, seasonal variation and influence of sampling technique

Ben-David Eric, Habiby Maryana, Haddad Elias, Hasanin Mahdi, Angel Dror, Booth Andy, Sabbah Isam.

Wastewater treatment plants (WWTPs) serve as important routes for microplastics (MP) to the environment. However, more effective MP sampling and detection methodologies, as well as a better understanding of their influence on MP occurrence and distributions in WWTP effluents, are needed for better removal and control. In this study, the efficiency of a municipal WWTP to remove MP was assessed by collecting samples from raw to tertiary effluent during a 12-month sampling campaign (season-based) using different sampling methods (containers, 24-h composite and large grab samples). MP retrieved from different treatment units within the WWTP were identified and quantified using plastic/non-plastic staining followed by optical microscopy, SEM and µ-Raman microscopy. Overall, the mean removal efficiency of MP in the WWTP was 97%, with most MP removed by the secondary stage and a mean effluent concentration of 1.97 MP L-1 after sand filtration. The relative abundance of fibers increased 74% of the total MP to 91% between raw wastewater and treated effluent, with a corresponding decrease in particles. Taking seasonal variations into account is important as total MP concentration in the effluent was notably higher in winter compared with the other seasons. Increasing the sampled volume using large samples or 24-h composite samples significantly reduced the variability between replicates. However, MP concentration post the tertiary stage was significantly lower using morning sampling (9 am) by large grab sampling method (1.2 MP L-1) compared to 24-h composite sampling (3.2 MP L-1) possibly due to intra-daily changes. Using a finer mesh size (0.45 µm) to capture MP beyond the size range typically studied ( $\geq 20 \ \mu m$ ) effectively quadrupled the number of MP detected in the tertiary effluent and highlights the importance of standardizing sampling procedures.

Keywords : Composite sampling , Grab sampling , Microplastic capture efficiency , Rapid sand filter (RSF) , Seasonal variation , Wastewater effluent

# A stable isotope assay for determining microbial degradation rates of plastics in the marine environment

Goudriaan Maaike, Hernando Morales Victor, Van Bommel Ronald, Van Der Meer Marcel, Ndhlovu Rachel, Hinnrichs Kai-Uwe, Niemann Helge.

The global use of plastic as a cheap and versatile base material has been growing exponentially. This lead to a likewise increase in plastic waste of which a substantial fraction enters the ocean, but the further fate of plastic in the marine realm is not well constrained. Pathways of plastic degradation (physicochemical and biological) in the marine environment are largely unresolved; yet, microbial plastic degradation is a potential plastic sink in the ocean. However, there is a lack of methods to determine this process, particular if the overall turnover is in the sub-percent range. We developed a novel method for quantifying kinetics of microbial plastic degradation that is based on tracing isotopically labelled polymers through microbial food web structures. We tested our method with a Rhodococcus Ruber strain (C-208), a known plastic degrader, as a model organism. We used granular polyethylene (PE) (granule size 30 µm) that was almost completely labelled with the stable isotope 13C (99%) as a sole carbon source. We monitored CO2 concentration and its stable carbon isotope composition during 35-day incubations at atmospheric oxygen concentrations and found an excess production of 13C-CO2. This result provides direct evidence for the microbially mediated mineralization of carbon that was ultimately derived from the polymer. After terminating the incubation, we measured dissolved inorganic carbon (DIC) concentrations allowing us to determine the total excess production of 13C in the CO2 and DIC phase, and thus the rate of plastic degradation. Of the 2000 µg PE added, up to 1% was degraded over a time course of 35 days with a rate of up to 1.9 µg month-1, providing a first characterization kinetics of R. Ruber mineralizing PE. The results show that isotopically labelled polymers can be used to quantify plastic degradation at rates that are undetectable for classic gravimetrical methods.

Keywords : Microbial kinetics , Plastic degradation , Rhodococcus Ruber , Stable isotopes

# **Influence of simulated weathering on microsize polypropylene properties and quantification by pyrolysis gas chromatography mass spectrometry** Alajo Tania.

Plastic debris has become ubiquitous in terrestrial and marine environments, and its consequent degradation into microplastics has not been yet well understood. Although there are no standardized analytical methods, recent advancements have identified and quantified microplastics in environmental samples using Pyrolysis Gas Chromatography/Mass Spectrometry (Pyr-GC/MS). To assess the reliability of Pyr-GC/MS for measuring microplastics in environmental samples, studies have used virgin polymer standards to correct for matrix effects and to select markers specific to the polymers investigated. However, little is known about how natural weathering processes could possibly impact inherent properties of microplastics, and consequently their quantitative analysis using techniques like Pyr-GC/MS for environmental samples. This research aims to add the understanding of weathered microplastic properties and how they could possibly cause variability in quantitation due to different properties in comparison to the original standards used for quantitation. Based on this, the objectives of this study were therefore to perform accelerated weathering of isotactic polypropylene microplastics of two different forms (i.e. beads ( 5 mm) and irregular shaped particles (250-500 µm, 500-1000 µm)) via photo-oxidization using accelerated laboratory weathering, and examine whether this impacts their quantitative estimation using Pyr-GC/MS techniques. The progressive weathering and degradation until 80 days (beads) and 37 days (particles) of beads and particles respectively, was confirmed via techniques including Fourier-transform Infrared - Attenuated Total Reflection (FTIR-ATR) spectroscopy, scanning electron microscopy (SEM), and differential scanning calorimetry (DSC). Comparison of pyrograms from Pyr-GC/MS of the progressive weathering was used to identify any changes to the quantification of virgin and aged MPs, specifically the pyrolysis product, 2,4-dimethyl-1- heptene. Quantitative changes were significant in terms of peak areas and suggest that quantitation using Pyr-GC/MS can be affected by weathering processes.

Keywords : carbonyl index , crystallinity , DSC , FTIR , GC/MS , microplastics , photo degradation , Pyr , SEM

**Poster session 24.4** 

# Session 24.4\_O. Chaired by Maria Murcia, Menorca

**Microplastic particles in atmospheric deposition of Northern Germany** *Klein Malin, Brecht Torben, Fischer Elke.* Paper number 327843

### Towards revealing sources of atmospheric microplastic pollution using an innovative winddrift sampling device

Oster Jakob, Loeder Martin, Babel Wolfgang, Georgi Christoph, Laforsch Christian. Paper number 334321

Sampling airborne microplastics and fibers: lessons learned from Aveiro, Portugal C. Prata Joana, L. Castro Joana, P. Da Costa João, Duarte Carlos, Cerqueira Mário, Rocha-Santos Teresa. Paper number 334322

# Microplastic particles in atmospheric deposition of Northern Germany

Klein Malin, Brecht Torben, Fischer Elke.

Atmospheric microplastic particles play an important role in calculating the input of microplastics in the environment. While a rising number of studies stating their concern, the extent of input through the atmosphere has been investigated sparsely. Based on a pilot study we presented at the Micro 2018, we started a longtime-study of microplastic particles in the atmosphere of Northern Germany. The monitoring took place every four weeks over the course of 12 months in two different areas of Germany. The city of Hamburg and the rural area of the Lake Tollense in Mecklenburg-Vorpommern. All investigation sites were equipped with three bulk-deposition samplers and a meteorological station. Three sites were located in the rural south of Hamburg, four sites in urban areas with a focus on heavy population, traffic or industrial pressure. In Mecklenburg-Vorpommern, four investigation sites were positioned north, east, south and west of the Lake Tollense. The volume of the samples was reduced by vacuum filtration onto a PC filter (Whatman 7060-4715) and the resulting suspensions were pretreated with hydrogen peroxide (30%) and sodium hypochlorite (7-14%) in order to destroy biological organic material. The samples were transferred to cellulose filters and underwent staining with nile red solution subsequently. Particles and fibers down to a lower size limit of 10 µm were counted under a fluorescence microscope and verified using µRaman spectroscopy. The amounts of microplastics in the atmospheric deposition of Northern Germany were statistically investigated with a focus on differences between sampling locations. The microplastic count was compared to the small-scale meteorological data of the sampling location. At four of the sampling sites in Hamburg we were able to link the microplastic particles with 10 µm sized particulate matter (cooperation with Luftmessnetz Hamburg). Two sites were investigated with attention to the canopy cover and the related comb-out effect.

Keywords : air pollution , Atmosphere , deposition , fluorescence microscopy , Nile Red staining ,  $\mu$ Raman spectroscopy





# Microplastic particles in atmospheric deposition of Northern Germany – An overview of the methodology of a one year study

### Malin Klein<sup>1</sup>, Torben Brecht<sup>1</sup>, Elke K. Fischer<sup>1</sup>

<sup>1</sup>Centre for Earth Systems and Sustainability, Microplastic Research at CEN (MRC), University of Hamburg

#### Introduction

atmosphere of Northern Germany.

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Atmospheric microplastic particles play an important role in calculating the input of microplastics in the environment. While a rising number of studies are stating their concern, the extent of input through the atmosphere has been investigated sparsely. Based on a pilot study we presented at the Micro 2018, we started a longtime-study of microplastic particles in the

The study aims to link microplastic pollution with anthropogenic and meteorological location factors. A new sampling and laboratory protocol was developed, to limit contamination and identify small-scale particles down to 10 µm. Due to Covid-19 restrictions, the laboratory analysis hasn't been completed yet. Therefore, we focus on the methodical approach.

#### Study Area

The monitoring took place with eleven investigation sites in two different areas of Germany (Fig. 1). The city of Hamburg and the rural area of the Lake Tollense in Mecklenburg-Vorpommern. Three sites were located in the rural south of Hamburg, four sites in urban areas with a focus on heavy population, high traffic or industrial pressure. In Mecklenburg-Vorpommern four investigation sites were positioned north, east, south and west of the Lake Tollense.



#### Material & Methods



To limit the microplastic contamination of the samples, a new bulk-deposition sampler was developed. Unlike regular bulk samplers, all parts that could contaminate the sample are made completely plastic- and rubber-free (Fig. 2). Three of the self build bulk-deposition samplers and one meteorological station were installed at each of the eleven sampling sites. The height was set to 2 m above ground. Sample bottles were changed every four weeks over the course of one year and sealed with a stainless steel cap for transportation. The meteorological data (temperature, humidity, wind speed, wind direction, precipitation) were measured every 15 minutes over the whole sampling period. The volume of the 348 samples plus 12 procedural blanks was determined and then reduced by vacuum filtration onto a PC filter (Whatman 7060-4715). The resulting suspensions were pretreated with hydrogen peroxide (30%) and sodium hypochlorite (7-14 %) in order to destroy biological organic material. The samples were transferred to cellulose filters and underwent staining with nile red solution subsequently. Particles and fibers down to a lower size limit of 10 µm are counted and measured under a fluorescence microscope (Axioscope 7, Zeiss) and verified using µRaman spectroscopy.

#### **Background Contamination**

To ensure the quality of the study, twelve procedural blanks were analyzed along the samples. Precautions were taken by using stainless steel equipment and by rinsing all glassware with acetone first followed by MilliQ water. Samples were treated under a dedicated fume hood, surface were wiped down and laboratory coats worn at all times.



### Preliminary Findings

- The developed bulk-deposit samplers have been working reliable over the whole sampling period and the stainless steel bottles enabled fast and easy sampling.
- Samples are varying in their particle load depending on the sampling location and date. Especially in the summer months the amount of organic and mineral matter was very high and made the digestion steps substantially difficult.
- All samples that have already been investigated under the fluorescence microscope contain microplastic particles. With decreasing particle size, the amount of microplastic particles on the filter increases. The chosen microscope settings and the laboratory protocol allow an identification down to a lower size limit of 10 µm.



ig, 4: Images of the filters taken under the fluorescence microscope for March 2020 (from left: Hamb Altona, Hamburg-Habichtstrasse, Hamburg-Open Field; Mecklenburg-Vorpommern-East, Mecklenburg-Vorpommern South and Mecklenburg-Vorpommern-West)

#### Outlook

- Calculation of microplastic concentration per m<sup>3</sup>/day over a year long sampling period
- Determination of particle size and polymer composition of atmospheric microplastic
- Investigation of differences in microplastic abundance between sampling sites
- Comparison between amount of microplastic and small-scale meteorological data

#### **Cooperation Partners**

We thank the Luftmessnetz Hamburg and the Neubrandenburger Stadtwerke for granting access to the sampling locations.

# Towards revealing sources of atmospheric microplastic pollution using an innovative wind-drift sampling device

Oster Jakob, Loeder Martin, Babel Wolfgang, Georgi Christoph, Laforsch Christian.

Plastic is discussed as a stressor for the environment and health. Today there is a massive amount of literature regarding this issue for different environments. However, information about concentrations, behavior and especially sources of microplastics in the atmosphere is almost lacking completely. Investigation of the wind drift of microplastics can help closing the lack of knowledge about sources for atmospheric microplastics. Thus, in this study we investigated whether microplastics drifted by wind can be measured. Therefore, we sampled three sites with different anthropogenic influence. For that we designed a new passive wind drift sampling device. Additionally, we applied footprint analyses to these measurements to determine the origin of the particles found. We found 5.32·10–03 - 1.29·10–02 MPs/m3. The results show, that wind-drifted microplastics can be detected using the sampling device. The origin and concentration of microplastics correlate with the anthropogenic influence at the respective sampling site. These results present the proof of concept for measuring wind-drifted microplastics and will help to reveal sources of atmospheric microplastics.

Keywords : atmosphere , drift , environment , method , microplastic , pollution , sampling , sources , wind



# Measuring atmospheric microplastic pollution using an innovative wind-drift sampling device [334321]



#### S.D. Jakob Oster<sup>1</sup>, Wolfgang Babel<sup>1</sup>, Christoph Georgi<sup>2</sup>, Sarmite Kernchen<sup>1</sup>, Andreas Held<sup>2</sup>, Christoph Thomas<sup>1</sup>,

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#### BACKGOUND

There is microplastic (MP) in the air! But where is it coming from? → Wind can disperse MP into the atmosphere and further transport it. Is it possible to measure MP actively transported by wind? → If so, we could better understand sources, fate and transport of MP in the atmosphere.





#### METHODOLOGY

We developed an innovative passive sampler for sampling microplastic actively transported by wind (three nets with 5 μm mesh size in three different heights: ground level, 1.5 m and 4 m.
We sampled three sampling sites with different anthropogenic influence: Fichtelgebirge – FG; Ecological botanical garden – ÖBG and industrial estate – IE)

 $\succ$  We analysed samples via  $\mu$ FTIR spectroscopy down to 10  $\mu$ m.

Three sampling sites with different anthropogenic influence. SI = Sink, SO = Source

#### RESULTS > MP concentration significantly differed between FG and IE > Concentration of MP ranged between 5.32·10<sup>-03</sup> -1.29·10<sup>-02</sup> MPs/m<sup>3</sup> > Main size of MP was between 50 and 150 µm > 10 different polymers were identified, mainly fragments > Footprint analyses show the source areas of the particles found



Fichtelgebirge = FG; Ecological botanical garden = ÖBG and industrial estate = IE. Three heights = Bottom, middle and top. LU = Land use. The errorbars in A,B and D correspond to SDs. Statistics were carried out using t-test and ANOVA. The differently highlighted areas in F denote the areas contributing with 50, 80 and 95% to the concentrations measured.

# Assessing the amount of MP transported by wind is possible using our passive wind drift sampler

Deutsche Forschungsgemeinschaft German Research Foundation

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# Sampling airborne microplastics and fibers: lessons learned from Aveiro, Portugal

C. Prata Joana, L. Castro Joana, P. Da Costa João, Duarte Carlos, Cerqueira Mário, Rocha-Santos Teresa.

Airborne microplastics and fibers can easily be inhaled, which causes increasing concerns over their environmental concentrations and toxic effects. However, the difficulty of sampling air can be felt in the need for specific equipment (e.g. air samplers) as well as the presence of other particulate matter and potential contaminations from the laboratory air. The objective of this work is to identify potential solutions for routine quantification, based on works conducted in Aveiro, Portugal. A first challenge was separating natural from synthetic fibers. A myriad of laboratory tests showed this could not be easily achieved through density separation, chemical digestion, or the use of staining dyes. The identification of fibers as natural or synthetic can only rely on visual characteristics for routine quantification. Based on the observation of multiple textile fibers, a diagram of identification has been produced based on the more regular structure of synthetic fibers. However, the identification of fibers and microplastics was hindered by the presence of other particulate matter. A method based on the removal of organic matter by H2O2 and density separation by NaI was developed and tested, with recovery rates of 94%. The use of both developed techniques in real sampling efforts allowed further improvements in contamination control measures, identifying possible sources as unfiltered work solutions, air deposition of fibers released from paper towels and cotton lab coats, directly weighting filters, and accumulation of dust in the sample inlet requiring proper decontamination procedures. Together, these developments allow a simple, fast, and reliable procedure which can be applied to determine concentrations of airborne microplastics and fibers.

Keywords : airborne microplastics , contamination , sampling , synthetic fibers

# Sampling-airborne-microplastics-and-fibers:lessons-learned-from-Aveiro,-Portugal-

Joana C. Prata<sup>1</sup>, Joana Castro<sup>2</sup>, João P. da Costa<sup>1</sup>, Armando C. Duarte<sup>1</sup>, Mário Cerqueira<sup>2</sup>, Teresa Rocha-Santos<sup>1</sup> 1. Centre for Environmental and Marine Studies (CESAM) & Department of Chemistry, University of Aveiro, 3810-193 Aveiro, Portugal 2. Centre for Environmental and Marine Studies (CESAM) & Department of Environment and Planning, University of Aveiro, 3810-193 Aveiro, Portugal

Sampling airborne microplastics and textile fibers, in both indoor and outdoor environments, presents methodological challenges despite their wide distribution in the environment<sup>1</sup>. These stem from relatively low concentrations, confounding particles, difficult identification, and cross contamination of samples. From trial and error, improvements were achieved in three key levels: strict contamination control measures, sampling and sample preparation, and particle identification.

### **Contamination Control**



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Filters Burning glass fiber filters, storing and weighting in clean glass Petri dishes.

required to produce reliable results<sup>2</sup>, with the particularity of sampling equipment decontamination<sup>3</sup>.



Thorough cleaning of sample equipment between samples.

Working under the laminar flow hood, in a room with limited access, wearing



Solutions-and-materials

Filtering all working solutions, using all glass or metal materials with proper cleaning, covering with lids or aluminum foil



Conducting blanks throughout the process: field blanks, procedural blanks, and open filters for air deposition.

### Sampling and Sample Preparation

n lab coats.

#### Sampling Sampling indoor or outdoor air with a portable air sampler with quartz fiber filters at a flow rate of 5 L min<sup>-1</sup> over 48-h period<sup>4</sup>.

#### Removing-organic-matter-Transferring particles from the filter to a solution of 15% H<sub>2</sub>O<sub>2</sub> and incubating at room temperature (20°C) for 8

days<sup>4</sup>

#### Density-separation-

Filtering and transferring again to a solution of saturated Nal, vortexing, leaving to separate for 90 minutes, and filtering the supernatant<sup>4</sup>.



References:-1. Dris et al. 2017 <u>10.1016/j.envpol.2016.12.013</u>; 2. Prata et al. 2021 <u>10.1016/j.jhazmat.2020.123660</u>; 3. Prata et al. 2020 <u>10.1016/j.mazpolbul.2020.111522</u>; 4. Prata et al. 2020 <u>10.1016/j.mex.2019.11.032;</u> 5. Prata et al. 2020 <u>10.1016/j.schotenv.2020.137498</u>. Acknowledgement:-This work was funded by Poruguese Science Foundation (FCT) through scholarship PD/80/135581/2018 under POCH funds, co-financed by the European Social Fund an Portuguese National Funds from MEC: Thanks are due to FCT/MCTES for the financial support to CESAM (UID9/S0017/2020 + UID8/S017/2020) through national funds. FCT

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# Session 24.4\_Me. Chaired by Ignacio de Sobrino, Lanzarote

Response of common bean (*Phaseolus vulgaris L.*) growth to soil contaminated with microplastics Meng Fanrong. Paper number 332454

### Influence of microplastics on grassland plants

*Cornelsen Hanna*. Paper number 334101

# **MicroSof Project:** study of the microplastic contamination in soil samples from 42 different sites in France.

Palazot Maialen, Kedzierski Mikaël, Bruzaud Stéphane. Paper number 334202

# **Response of common bean (***Phaseolus vulgaris L.***) growth to soil contaminated** with microplastics

Meng Fanrong.

Although concerns surrounding microplastics (MPs) in terrestrial ecosystems have been growing in recent years, little is known about the responses of plant growth to MPs pollution. Here, we conducted a pot experiment in a net house under natural condition by adding two types of MPs, lowdensity polyethylene (LDPE-MPs) and polylactic acid (PLA) mixed with poly-butylene-adipate-coterephthalate (PBAT, Bio-MPs), to sandy soil at 5 doses (0.5%, 1.0%, 1.5%, 2.0%, 2.5% w/w dry soil weight). The effects of LDPE-MPs and Bio-MPs on common bean (Phaseolus vulgaris L) were tested. Compared to control (no MPs addition), LDPE-MPs showed no significant effects on shoot, root and fruit biomass while  $\geq 1.0\%$  LDPE-MPs showed significant higher specific root nodules (n·g-1 dry root biomass) and only 2.5% LDPE-MPs showed significant higher specific root length (cm·g-1 dry root biomass). 1.0% LDPE-MPs caused significant higher leaf area and 0.5% LDPE-MPs caused significant lower leaf relative chlorophyll content. For Bio-MPs treatment, compared to control, ≥1.5% Bio-MPs showed significant lower shoot and root biomass. ≥2.0% Bio-MPs showed significant lower leaf area and fruit biomass. All Bio-MPs treatments showed significant higher specific root length and specific root nodules as compared to control. The results of the current research show that both MPs induced the responses of common bean growth, and  $\geq 1.5\%$  Bio-MPs exerted stronger effects. Further studies of their ecological impacts on soil-plant systems are urgently needed.

Keywords : Biodegradable microplastics , Microplastics , Plant growth , plant system. , Soil

# Response of common bean (Phaseolus vulgaris L.) growth to soil contaminated with microplastics

Fanrong Meng & Xiaomei Yang & Michel Riksen & Violette Geissen Soil Physics and Land Management, Wageningen University, The Netherlands



# Introduction

Although concerns surrounding microplastics (MPs) in terrestrial ecosystems have been growing in recent years. The growing body of literature have indicated that MPs could affect the soil biophysical environments, i.e. decreased soil bulk density and soil microbial activities, increased soil evaporation and desiccation cracking. The increasing concerns surrounding plastic pollution in agriculture have led to the development of biodegradable materials. However, little is known about its ecological impacts. Here, we conducted a pot experiment to investigate the the responses of plant growth to MPs pollution.

# Experiment design







## Results

2.3

24

2.1

root length (×104 cm·g<sup>4</sup>)\_VS







Bio-MPs produced higher specific root length and specific root nodules while LDPE-MPs also showed significant impacts on specific root nodules.



- · Factor 1 was mainly associated with total plant biomass, thus defined as plant biomass.
- Factor 2 (F2) included PodNb and FruitB and defined as plant production.
- · Factor 3 included FRL and SRL and defined as root characteristics.

# Conclusion

- Bio-MPs have a stronger effect on the growth of common bean (Phaseolus vulgaris L.) than LDPE-MPs.
- Bio-MPs at higher concentrations capable of eliciting the responses of common bean growth.
- The results presented have demonstrated that the occurrence of MPs in soil are capable of changing the plant growth, this is a fundamental understanding for future efforts to assess risks of agricultural MPs pollution in soil-plant systems.

### Influence of microplastics on grassland plants

Cornelsen Hanna.

So far, the fate of freshwater and marine systems has been in the focus of societal and scientific attention with manifold negative effects of microplastics identified for aquatic biota. On the contrary, the characteristics of these contaminants in terrestrial ecosystems have not been dealt with in depth. Especially our knowledge on dynamics of microplastics in agricultural soils and crops is fragmentary. Agroecosystems are areas of high human influence, providing manifold input routes for microplastic particles. Potential pathways are agricultural practices like fertilization by sewage sludge, the usage of plastic mulching or irrigation with contaminated water. Beside these, littering behaviour, runoff from surrounding areas and aerial deposition are further entry points. Considerable parts of the global agricultural area are covered by grassland that is providing essential functions for lifestock and fodder supply, and thus also food production. Therefore, it is important to determine the so far unclear consequences of microplastics' presence for grassland soil and plants. In the initial stage of plant performance, potential effects of stressors can be observed more detailed. Therefore, the aim of this research was to investigate the influence of different forms of microplastics particularly on the germination process of grassland species. We assessed different sets of germination trials under laboratory conditions, using annual ryegrass (Lolium multiflorum Lam.var. westerwoldicum) as a model plant. We chose polyvinylchloride (PVC) and particles derived from truck tyres for simulation of common primary and secondary microplastics. Seeds were grown on moisturized filter paper in petri dishes with different amounts of microplastics. After a predefined growth period, above- and belowground biomass of the seedlings was separately analysed. Dried shoot material was analysed for d13C as an indicator of drought stress using an isotope ratio mass spectrometer (IRMS), and dried roots were analysed with a scanning instrument. Results will be shown and discussed.

Keywords : Agriculture , germination trials , polyvinylchloride , terrestrial microplastics

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Traditio et Innovatio

# **MicroSof Project: study of the microplastic contamination in soil samples from 42 different sites in France**

Palazot Maialen, Kedzierski Mikaël, Bruzaud Stéphane.

To date, the majority of the research on microplastics (MP) has focused on the marine environment. The pollution of continental ecosystems is comparatively little studied, even though the majority of all plastics is used and disposed on land. Recent studies on soil contamination by plastics and microplastics have revealed the presence of plastic particles in various terrestrial ecosystems, from the most urbanized and industrialized areas to the most remote areas. The main sources of plastic in soils include land application of contaminated sewage sludge and compost, plastic mulching, wastewater irrigation, atmospheric deposition, littering and surface runoff [1]. However, estimates of MP concentrations and chemical natures are still limited and differ between studies. In France, only a few studies have tackled the issue of MP contamination in soils. The MicroSof projects aims to establish first national references on the contamination of French soils by microplastics. To do so, soil samples from 42 different sites under different land uses will be analyzed. A protocol for the extraction and characterization of MP is currently being set up. After pre-treatment and subsampling, the samples will be digested with Fenton's reagent following the methodology outlined by Hurley et al. (2018) [2] and MP will be extracted by density separation. The study will then allow to quantify (mass, number) and to qualify (chemical nature) the accumulated plastic fragments, as well as the size and the shape of these fragments. [1] M. Bläsing et W. Amelung, « Plastics in soil: Analytical methods and possible sources », Science of The Total Environment, vol. 612, p. 422-435, janv. 2018, doi: 10.1016/j.scitotenv.2017.08.086. [2] R. R. Hurley, A. L. Lusher, M. Olsen, et L. Nizzetto, « Validation of a Method for Extracting Microplastics from Complex, Organic-Rich, Environmental Matrices », Environ. Sci. Technol., vol. 52, no 13, p. 7409-7417, juill. 2018, doi: 10.1021/acs.est.8b01517.

Keywords : Fenton's reagent , infrared spectroscopy , microplastic extraction , microplastics , soil

334202

# THE MICROSOF PROJECT: STUDY OF THE MICROPLASTIC CONTAMINATION IN SOIL SAMPLES FROM 42 DIFFERENT SITES IN FRANCE.

### Maialen Palazot<sup>\*1</sup>, Mikaël Kedzierski<sup>1</sup>, Stéphane Bruzaud<sup>1</sup>

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- **ISSUES** Recent studies have revealed the **presence of microplastics (MP) in various terrestrial ecosystems**, from the most urbanized and industrialized areas to the most remote areas.
  - Main sources of plastic in soils include: land application of contaminated sewage sludge and compost, plastic mulching, wastewater irrigation, atmospheric deposition, littering and surface runoff [1].
  - Estimates of MP concentrations and chemical natures are still limited and differ between studies. In France, only a few studies have tackled the issue of MP contamination in soils.

# THE MICROSOF PROJECT

- The MicroSof project aims to establish first national references on the contamination of French soils by microplastics.
- Soil samples (~1kg) from 42 different sites under different land uses will be analyzed:
  - 30 samples from crop lands
  - 4 samples from forests

Quality controls: Positive and negative controls, use of 100% cotton lab coats and natural fiber clothing, experiments conducted in a laminar flow cabinet,

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samples kept covered, use of glass or stainless steel materials...

- 4 samples from grasslands
- 4 samples from vineyards and orchards



### **OBJECTIVES**

References

 Develop a protocol for the extraction and Density: 1,8 g.cm<sup>-3</sup> characterization of microplastics trapped in soil Extract all MP, even samples with different characteristics. those of high density such as PVC and PET. • Quantify (number) and qualify (chemical nature, size, shape) the extracted microplastics. Nal recycling, ImageJ up to 10 cycles [3] Number Size Shape 50 mL Falcons MATERIAL AND METHODS (WORK IN PROGRESS...) Vacuum Pre-Density Wet sieving Analysis Subsampling SOM<sup>\*</sup> digestion filtration of the treatment Remove the separation Pictures + image supernatant ⇒ Triplicates of Fenton's reagent finest fraction processing Drying 40°C Saturated Nal T°C < 40°C 300um and 50g program, FTIR (<50um) Sieving 5 & 2mm solution 50um filters \*Soil Organic Matter Larger agglo-Because of technical limitations, -merates are lightly Catalyst crushed during only the fraction solution H<sub>2</sub>O<sub>2</sub> (30%) drying and sieving [50µm; 5mm[ will be FeSO<sub>4</sub> 7 H<sub>2</sub>O analyzed. 1:1 ratio . 2065 Exothermic reaction [300µm ; 5mm[ → ATR-FTIR Temperature kept 1 below 40 °C using [50µm ; 300µm[ → µFTIR 0 an ice bath intermittently Following Hurley et al. (2018) [2]

CURRENT AND FUTURE WORK

- Currently: validation phase of the first version of the protocol, recovery experiments using aged polymers.
  - Need to adapt the protocol according to the type of soil.
  - Université Bretagne Sud Université Bretagne Sud INRACO

# Session 24.4\_Ma. Chaired by Sascha Müller, Copenhagen

# Surface properties of microplastic particles matter for particle-cell-interactions

Ramsperger Anja, Rudolph Julia, Völkl Matthias, Witzmann Thomas, Freitag Ruth, Jérôme Valérie, Fery Andreas, Scheibel Thomas, Kress Holger, Laforsch Christian. Paper number 332951

# Investigation of microplastic particle exchange at the air-water-interface via the process of bursting bubbles

*Oehlschlägel Lisa Marie, Georgi Christoph, Held Andreas Balthasar.* Paper number 333936 \_MICRO 2020. Fate and Impact of Microplastics: Knowledge and Responsibilities. 23-27 November 2020\_\_\_\_

Surface properties of microplastic particles matter for particle-cell-interactions

Ramsperger Anja, Rudolph Julia, Völkl Matthias, Witzmann Thomas, Freitag Ruth, Jérôme Valérie, Fery Andreas, Scheibel Thomas, Kress Holger, Laforsch Christian.

Plastic pollution has been shown in all environmental compartments ranging from the marine and limnetic environment (Imhof et al., 2013), terrestrial ecosystems, like agricultural soils right up to the atmosphere. To date, plastic particles have mainly been categorized by polymer type, shape, and size (Cole et al., 2011). But there is another important issue arising when investigating microplastic and its interaction with cells. With decreasing size, the surface-volume ratio increases which makes surface properties more important to take into account. It is generally believed that the surface properties of the particles influence the cell interaction. Therefore, we investigated un-functionalized polystyrene particles with the size of 3 µm with different surface properties of two different manufactures. We found out that the cellular interaction and uptake of microplastic particles (polystyrene) differs for the two particle types. Using Zeta-Potential measurements and Colloidal Probe-Atomic Force Microscopy (CP-AFM) we could show a significant difference in the electric surface properties: homogeneously charged particles vs. heterogeneously charged particles. The heterogeneous surface charge manifests itself in an electrostatic interaction of the particles that depends on the mutual orientation of the particles. CP-AFM is therefore a magnificent tool to obtain additional informations about surface charge and it's distribution on microplastic particles.

Keywords : Atomic Force Microscopy , Colloidal Probe , microplastic , particles , surface charge





# Surface Charge Matters for Particle-Cell Interactions

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### Motivation

Microplastic particles and there effect on and interaction with living cells have been studied a lot in the recent years. Though results are often contradictory even when particles with same polymer type, size and shape are used <sup>[1]</sup>. Therefore we investigated the surface charge of non-functionalised, spherical polystyrene (PS) particles with the same size (3  $\mu$ m) from two **different manufacturers (P-MPP & M-MPP**). P-MPP shows **higher** particle-cell interactions & internalisation.



- · Microplastic particles can have same polymer type, size, shape but different surface charge
- Particle-cell-interactions depent on surface charge

### Acknowledgements

Günter Auernhammer

#### References

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# Investigation of microplastic particle exchange at the air-water-interface via the process of bursting bubbles

Oehlschlägel Lisa Marie, Georgi Christoph, Held Andreas Balthasar.

In this study, which is part of the SFB 1357 Microplastics, the transition of MP particles from water bodies into the air is examined. Investigating the water-air transition path is important because MP particles are abundant in water bodies. A key question is whether or not MP particles are transferred from water bodies into the atmosphere, and at what rate? One transfer process is the ejection of water drops into the air. For droplet generation, several mechanisms are known. The bursting of gas bubbles at the water-air interface is an important mechanism. In order to quantify MP particle transition rates due to bubble bursting and the influence of MP properties on the transition process, we set up an experiment in a glass flask containing 1 l of desalted water, a stainless-steel frit for generating bubbles and 1 l of initially particle-free air. For generating bubbles, a stainless-steel frit with a volume flow rate of 20 ml min-1 of particle-free air is used. For the first experiments, spherical polystyrene (PS) particles with diameters of  $0.5 - 3 \,\mu\text{m}$  are available. An optical particle counter measures the number of airborne particles after diffusion drying in 31 size bins from 0.25 µm to 32 µm. In these first experiments, we find a strong enhancement of airborne particle number concentrations in the size range of the PS particles suspended in the water. Comparing the ratio of suspended PS particles in water and the measured airborne particle concentration we find a decreasing transition rate with increasing particle size in our preliminary experiments. Further experiments are ongoing to validate these findings, and we are also planning experiments with varying particle sizes, concentrations and different polymer types as well as different bubble sizes and bubble concentrations. This poster is part of the SFB1357 Local Node Event.

Keywords : air , airborne particle number concentrations , bubble bursting , SFB1357 , transition rate , water interface



### Bursting bubbles experiment for the understanding of microplastic particle exchange at the air-water-interface [333936]

Lisa Marie Oehlschlägel<sup>1</sup>, Christoph Georgi<sup>1</sup>, Andreas Held<sup>1,2</sup> <sup>1</sup> Environmental Chemistry and Air Research, TU Berlin, Berlin, Germany <sup>2</sup> Bayreuth Center of Ecology and Environmental Research, University of Bayreuth, Bayreuth, Germany



CRC 1357 Microplastics

#### Objectives and relevance



We expect that water drops that are ejected into the air will transport Microplastic Particles (MPP) to the atmosphere. This is a well-known transition process for other components in the water such as salt.

Droplets can be generated by several mechanisms. One of the main processes is bursting of gas bubbles. Due to buoyant forces, gas bubbles formed in the water rise to the water surface and burst.

Fig. 1: Scheme of film- (black) and jetdrops (blue) after bubble bursting

# Bubble bursting: Droplet generation

- Small film droplets develop 1.
- 2. Bubble cavity collapses & a water jet arises vertically
- 3. Jet drops can develop

We investigate if film and/or jet drops lead to the transport of MPP into the air above.

#### **Research questions**

- Can airborne MPP be observed after bubble bursting?
- Does the MPP concentration in the water influence the transition rate?
- What fraction of MPP is transported into the air?
- Does the size of the particles influence the transition rate?

### Experimental setup



Fig. 2: Experimental setup for the bubble bursting experiments with MPP. The numbers are explained in the description above.

- Glass flask containing 1 liter of de-ionized water (1)
- Particle-free headspace (volume of 1 liter) (2)
- . Peristaltic pump pumping (3) filtered air through a stainless-steel frit (2  $\mu$ m/ 5  $\mu$ m/ 10  $\mu$ m diameter) (4) for bubble generation and film drop production
- Diffusion dryer (5) and optical particle counter (Grimm Mini-LAS 11-R) (6) for particle sizing and detection
- Spherical polystyrene particles (diameter 0.5 µm, 0.75 µm, 1 µm, 2 µm) (7)

#### Methods

We prepare MP suspensions of different particle number concentrations and sizes in multiple experiments. The optical particle counter measures the number of particles ejected into the air after diffusion-drying in different size ranges. Subsequently, we compare the results of different particle sizes measured by the OPC (air environment) and particle concentrations in suspension (water). The volume flow rate of filtered air for bubble generation is 20 ml min<sup>-1</sup>.



Fig. 3: Particle size distribution with different amounts of 0.5 µm PS particles suspended in 1 l water (experiment duration: 1 day)



Fig. 4: Ratio of particle concentrations in the air headspace to PS concentrations suspended in water for different particle sizes (experiment duration: 1 day). The concentration in the headspace is a mean value of the displayed range for the equivalent particle diameter.

#### Conclusions and future tasks

- Strong enhancement of airborne particle concentrations in the diameter range of the suspended particles
- Concentration of the airborne particles depends on the MPP concentration suspended in water.
- The fraction of airborne MPP ranges between 0.3 1.5 per one million particles suspended in water using a volume flow rate of 20 ml min<sup>-1</sup> for bubble generation.
- The transition efficiency seems to decrease with particle size
- Further experiments are ongoing to validate these preliminary results. Ongoing and future tasks are:
  - Experiments with different particle concentrations, sizes, polymer types, and bubbles with varying properties
  - > Characterizing bubble size spectra and concentrations





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# Session 24.5\_O. Chaired by Arnaud Huvet, Plouzané

### **Realism in microplastics research? Learnings from multigenerational studies with Daphnia** *magna Schür Christoph, Wagner Martin.* Paper number 334479

### **Microplastic characteristics across environmental compartments** *Kooi Merel, Primpke Sebastian, Mintenig Svenja, Gerdts Gunnar, Koelmans Albert.* Paper number 334480

# **Realism in microplastics research? Learnings from multigenerational studies** with *Daphnia magna*

Schür Christoph, Wagner Martin.

The call for environmental realism in ecotoxicology studies with microplastics has largely been focused on moving away from spherical beads and emulating concentrations we are able to detect in the environment. But the interactions of microplastics and biota in the aquatic environment are shaped by a plethora of factors, both biotic and abiotic. We therefore expanded the standardized test protocols with Daphnia magna to include food limitation as additional stressor and to cover four consecutive generations of animals. The setup was used throughout two large-scale studies to compare effects of virgin and aged irregular microplastics as well as natural reference particles on Daphnia magna. Food limitation proved an effective secondary stressor, affecting all endpoints except mortality. Exposure to microplastics increased mortality and affected reproduction and growth endpoints, while the natural reference particles had no such effects. Exposure to aged microplastics produced lower mortality than virgin microplastics of the same batch but did not differently affect reproduction, adult and neonate size, and length development. The multigenerational approach under food limitation elicited insights otherwise not visible in shorter experimental periods, since most effects only manifested after several generations. We present the key learnings from both studies, compare our findings to those of similar studies, and translate them into recommendations towards future microplastics studies.

Keywords : aged microplastics , chronic toxicity , Daphnia , ecotoxicology , freshwater , growth , kaolin , mortality , multigeneration , polystyrene , reproduction , wastewater

### **Microplastic characteristics across environmental compartments**

Kooi Merel, Primpke Sebastian, Mintenig Svenja, Gerdts Gunnar, Koelmans Albert.

Environmental microplastic (EMP) consists of a mixture of particles with different sizes, shapes and polymer types. These characteristics drive transport, fate, exposure, effects and risks, so understanding the variability of these features is of paramount importance. Here, we combine EMP datasets obtained from samples from different environmental compartments, including marine and freshwater surface waters and sediments, waste water effluents, and freshwater organisms. All data have been produced through µFTIR mapping or imaging using state-of-the-art standardized automated analysis software (AAP/MPAPP/siMPle). A total of over 100 samples, containing information on individual particle properties including size, shape and polymer ID for over 40,000 particles, has been analyzed for patterns. We parameterized continuous distributions that describe the particle properties, on the sample as well as on the compartment level, and identify the statistical significance of inter- and intra-compartment differences. This includes testing if the distributions for size and shape differ per polymer type. Lastly, we check for correlations between the different properties. Aside from using these distributions as input and guidance for models and experiments, we explore for the existence of general scaling laws, which can be used for extrapolating (sub-)samples to the full microplastic spectrum.

Keywords : effluent , environment , freshwater , marine , microplastic , properties , sediment , water

# Session 24.5\_Me. Chaired by Eva Cardona, Menorca

Quality criteria for microplastic effect studies in the context of risk assessment: A critical review

De Ruijter Vera, Redondo-Hasselerharm Paula, Gouin Todd, Koelmans Bart. Paper number 332939

# The fate of microplastics when making sludge into crude oil – the impact of a hydrothermal liquefaction process on microplastics in wastewater treatment plant sludge

*Chand Rupa, Kohansal Komeil, Helmer Thomas, Toor Saqib, Vollertsen Jes.* Paper number 333974

**Table salt and coarse salt: a source of contamination by microplastic** *Campos de Lelis, Diego Caetano, Dias da Cunha Rafaela Luiza, De Brito-Gitirana Lycia.* Paper number 331571

# Quality criteria for microplastic effect studies in the context of risk assessment: A critical review

De Ruijter Vera, Redondo-Hasselerharm Paula, Gouin Todd, Koelmans Bart.

In the literature, there is widespread consensus that methods in plastic research need improvement. Current limitations in quality assurance and harmonization prevent progress in our understanding of what the true effects of microplastic in the environment are. Following the recent development of quality assessment methods for studies reporting concentrations in biota and water samples, we propose a method to assess the quality of microplastic effect studies. We reviewed 105 microplastic effect studies with aquatic biota, provided a systematic overview of their characteristics, developed 20 guality criteria in four main criteria categories (particle characterization, experimental design, applicability in risk assessment, and ecological relevance), propose a protocol for future effect studies with particles, and, finally, used all the information to define the weight of evidence with respect to demonstrated effect mechanisms. On average, studies scored 44.6% (range 20-77.5%) of the maximum score. No study scored positively on all criteria, reconfirming the urgent need for better quality assurance. Most urgent recommendations for improvement relate to avoiding and verifying background contamination, and to improving the environmental relevance of exposure conditions. In far too many instances, studies suggest and speculate mechanisms that are poorly supported by the design and reporting of data in the study. This represents a problem for decisionmakers and needs to be minimized in future research. In their papers, authors frame 10 effects mechanisms as 'suggested', whereas 7 of them are framed as 'demonstrated'. When accounting for the quality of the studies according to our assessment, three of these mechanisms remained. These are inhibition of food assimilation and/or decreased nutritional value of food, internal physical damage and external physical damage. We recommend that risk assessment addresses these mechanisms with higher priority.

Keywords : effect mechanism , microplastic , article toxicity , quality criteria , risk assessment

# The fate of microplastics when making sludge into crude oil – the impact of a hydrothermal liquefaction process on microplastics in wastewater treatment plant sludge

Chand Rupa, Kohansal Komeil, Helmer Thomas, Toor Saqib, Vollertsen Jes.

Microplastics (MPs) are entering wastewater treatment plants (WWTPs) from various sources such as primary MPs in consumer products and secondary MPs from wear of plastic items and textiles. There is a scientific consensus that the efficiency of WWTPs in retaining MPs is high, with up to 99% being retained and ultimately ending up in the sludge. Within a totally different research diciplin, scientist are also working on producing bio-crude oil from the sludge by processing it at pressures around 30 Mpa and temperatures around 400°C known as hydrothermal liquefaction (HTL). In an interdisciplinary study, we have studied the fate of MPs in WWTP sludge fed into a continuous HTL reactor. The HTL products as a mixture of bio-crude, aqueous, and solid residues have been investigated. Duplicate samples of each matrix were analyzed for MP. Wet-oxidation (H2O2), sodium dodecyl sulfate, and enzymatic treatment were performed to remove natural organic materials such as cellulose, protein, and lipid compounds. Inorganic particles were removed by density separation using sodium polytungstate solution. The bigger particles (¿ 500 µm) were separated by sieving and potential MP candidates handpicked and analyzed by ATR-FTIR (Agilent Cary 630, diamond ATR). The smaller particles (10-500 µm) were analyzed by FPA-µFTIR (Agilent Cary 620/670) and the particles identified using the software siMPle for automated analysis of the generated spectral image. 13 polymer types, comprising particles of 11-3207 µm, were observed in the sludge with Polyurethane (PU), Polyethylene (PE), Polypropylene (PP), Polyester (PEsT), alkvd, and cellulose acetate as the dominant ones. In the HTL products, only a few olefins such as PP and PE with sizes of 13.3-81.6 µm were observed. Furthermore, the MP removal efficiency of the HTL process was between 99.65% and 99.99% by particle numbers and mass respectively.

Keywords : aqueous and solid residue , bio , crude , hydrothermal liquefaction , Primary MPs , secondary MPs , sludge

### Table salt and coarse salt: a source of contamination by microplastic

Campos De Lelis Diego Caetano, Dias Da Cunha Rafaela Luiza, De Brito-Gitirana Lycia.

Pollution from plastic waste has attracted attention due to the long-term durability of plastic waste in the environment and its adverse effects on marine fauna as well as humans. Despite the occurrence of microplastics in sea salt consumed by humans in several countries, there is no available information on the presence of microplastic in commercial salts produced in Brazil. This study analyzed five different brands of table salts and three brands of coarse salts from two main salt producing regions in Brazil (sea salts from Areia Branca City of Rio Grande do Norte State and lake salts from the Araruama lagoon, in Cabo Frio City of Rio de Janeiro State January). Samples were analyzed under a light microscope to verify the grain size and under a stereo microscope to visualize the plastic fragments. In addition, ATR-FTIR analysis was used to identify the type of microplastic. The results indicated that Brazilian commercial salts contained polyethylene and polypropylene. As table salts are used as condiment and food preservative, people are continually exposed to plastics. Therefore, more effective rules for regulating the salt refinement process are needed.

Keywords : atr , brazil , coarse , contamination , food , ftir , microplastic , salt , table
#### Session 24.5\_Ma. Chaired by Salud Deudero, Mallorca

**Low microplastic loads in North Sea, Mediterranean and Atlantic topshells** *Ehlers Sonja, Ellrich Julius, Koop Jochen.* Paper number 329516

# Two sides of the same coin: A coupled modelling description of the dynamics of microplastics and associated contaminants in the Mediterranean Sea

*Guerrini Federica, Mari Lorenzo, Casagrandi Renato.* Paper number 334423

## Aqua-lit project: how can the aquaculture sector contribute to reducing marine litter in the Mediterranean Sea?

*Vidal Maria, Alomar Carmen, Deudero Salud.* Paper number 334547

# How to detect the impact of microplastics in Mediterranean MPAs: the Plastic Busters MPAs approach

Fossi Maria Cristina, Baini Matteo, Casini Silvia, Galli Matteo, Caliani Ilaria, Campani Tommaso, Giani Dario, Rosso Massimiliano, Tepsich Paola, Scotti Gianfranco, Giannini Francesca, Galgani Francois, Sebastian Leccia, Romeo Teresa, Panti Cristina. Paper number 334847

#### **Low microplastic loads in North Sea, Mediterranean and Atlantic topshells** Ehlers Sonja, Ellrich Julius, Koop Jochen.

Topshells are common marine snails inhabiting shallow subtidal habitats along shorelines worldwide. These snails feed by browsing particles off the substrate. Ecotoxicologists use topshells as bioindicators for harmful substances such as heavy metals. In this study, we investigate whether topshells can serve as bioindicators for microplastic pollution. For that, we are examining microplastic (particles ; 5 mm) loads in topshell (Phorcus turbinatus, Phorcus sauciatus, Steromphala cineraria) and water samples collected at wave-exposed and wave-sheltered locations on the French and Italian Mediterranean coast (in 2007/2008 and 2019), on Helgoland (in 2009/2020) and Madeira island (in 2020). We consistently found relatively low topshell microplastic loads, composed of fragments, fibres, films and spheres of various colours and polymer types such as polyester, polyethylene, polyamide, polystyrene and polypropylene. Interestingly, we found that many microplastics consisted of paint chips. Currently, we are analysing the water samples to evaluate whether the topshell plastic loads reflect environmental microplastic concentrations and compositions. We verified all microplastics using micro-Fourier-transform infrared spectroscopy (µFTIR). This is exceptional, since the few studies that examined microplastics in marine snails verified only small microplastic subsets using µFTIR or relied entirely on visual microplastic identification. However, as microplastic validation studies indicated that up to 70 % of the visually examined particles are actually misclassified as microplastics, our study is the first to report verified microplastic loads in marine snails.

Keywords : exposed shores , gastropods , paint chip , sheltered bays ,  $\mu FTIR$ 

# Two sides of the same coin: A coupled modelling description of the dynamics of microplastics and associated contaminants in the Mediterranean Sea

Guerrini Federica, Mari Lorenzo, Casagrandi Renato.

Floating plastic litter in the marine environment is hardly found as clean fragments, rather it is often colonized by marine organisms and contaminated with organic pollutants, as mounting evidence highlights. Those interactions between plastics and both the biotic and the abiotic components of the seascape are even more relevant for microplastics, due to its high surface-to-volume ratio: they affect the dynamics of microplastics at sea (as biofouled particles tend to sink) and may exacerbate its toxicity to marine biota. A thorough assessment of the risk caused by microplastics and other pollutants of concern to marine ecosystems thus demands for a joint description of these two kinds of pollution. Here we illustrate a novel modelling framework to simultaneously describe in a simple, yet comprehensive, way the transport of microplastics in the sea surface, the advection-diffusion of selected organic contaminants, and the gradient-driven chemical exchanges between microplastic particles and the marine environment. Focusing on the Mediterranean Sea, we simulated two years of microplastic-related pollution (our coin) by coupling the dispersal of a target pollutant (through Eulerian advection-diffusion, one side of the coin) and of microplastic particles (using a Lagrangian framework, the other side of the coin). The two sides make the coin, because each particle can chemically interact with the surrounding environment while being subject to current-driven movement before eventually sinking. Over the simulated period, we found that the patterns of organic pollution at sea may significantly be affected by the presence of plastic particles acting as vectors, and consequently that particle sinking plays a significant role in regulating the balance on the sea surface layer of both microplastics and the pollutants they carry. Although simplified, the modelling framework described here moves a first, yet useful step towards a comprehensive quantification of the multifaceted threat posed by plastic-related marine pollution.

Keywords : Advection , diffusion , Eulerian modelling , Lagrangian modelling , marine pollution , microplastics , organic pollutants

# Aqua-lit project: how can the aquaculture sector contribute to reducing marine litter in the Mediterranean Sea?

Vidal Maria, Alomar Carmen, Deudero Salud.

Marine litter has multiple sources based both at sea and land. Amongst these sources, aquaculture can contribute to the generation of marine litter. From a bibliographic research on scientific papers, it has been observed that between 4.08% and 12.33% of the items found in beaches and 14.75% of seafloor marine litter is related to aquaculture practices (Sandra et al., 2020). Given that aquaculture is expected to increase substantially during the next 10 years, it is important to gather information in the three main waste management components related to the non-organic residues: (1) prevention and reduction, (2) monitoring and quantification, (3) removal and recycling. Consequently, the aim of this study is to present lessons learnt, innovative solutions and currently applied best practices on the mentioned waste management components, gathered during an interactive Learning Lab, which involved 16 Spanish representatives of multiple types of aquaculture stakeholders of the Mediterranean Sea basin. Preliminary results show that, for most of the aquaculture stakeholders, needs regarding non-organic litter management at a short-term are related to creating synergies among all the stakeholders for a common understanding of aquaculture marine litter quantification. At a mid-term, some of the recommendations included making efforts in eco-design of the gear and to enforce the current policies, among others. Finally, innovation in automated waste management collection systems and fostering incentives in following environmental certification including marine litter in the criteria, were also highlighted from a long-term point of view. All this information has contributed to assess and evaluate the potential future impacts of the aquaculture sector in the marine ecosystems regarding the non-organic marine litter. The outcomes will be transferred to the sector by creating a toolbox available through different platforms. Acknowledgments: This project has received funding from the European Union's under grant agreement EASME/EMFF/2017/1.2.1.12/S2/04/S12.789391.

Keywords : Aquaculture , marine ecosystems , marine litter , monitoring , prevention , recycling

#### How to detect the impact of microplastics in Mediterranean MPAs: the Plastic Busters MPAs approach

Fossi Maria Cristina, Baini Matteo, Casini Silvia, Galli Matteo, Caliani Ilaria, Campani Tommaso, Giani Dario, Rosso Massimiliano, Tepsich Paola, Scotti Gianfranco, Giannini Francesca, Galgani Francois, Sebastian Leccia, Romeo Teresa, Panti Cristina.

Plastic Busters MPAs is a 4-year-long Interreg Med-project aiming to contribute to maintaining biodiversity and preserving natural ecosystems in pelagic and coastal Mediterranean marine protected areas (MPAs), by defining and implementing a harmonized approach against marine litter. The project entails actions that address the whole management cycle of marine litter, from monitoring and assessment to prevention and mitigation, as well as actions to strengthen networking between and among pelagic and coastal MPAs. The overarching aim of this presentation is to describe the Plastic Busters MPAs harmonized monitoring approach to detect the impact of marine litter (particularly microplastics) on Mediterranean ecosystems and marine biodiversity, including endangered species (cetaceans, sea turtles and birds) inhabiting pelagic and coastal MPAs in the largest SPAMI of the Mediterranean sea. The implementation of the monitoring strategy and the preliminary results obtained from the monitoring carried out in the Pelagic SPAMI Pelagos Sanctuary and in the Tuscan Archipelago National Park (PNAT) will be presented. In spring-summer 2019, 34 researchers of 8 European institutions, monitored more than 2230 nautical miles, collecting 140 samples of superficial microplastics in the study areas and carrying out 280 monitoring of surface macrolitter simultaneously monitoring biota. During these campaign, in order to assess the ecotoxicological impact on biodiversity, cetacean species skin biopsies, neustonic invertebrates, lantern fish, mussels and several edible fish species were collected. The entire sampling design was guided by the development of a marine litter distribution model in order to identify the possible microplastic hot spot areas and of the potential impact on biota. Furthermore, seasonal monitoring of the marine litter were regularly carried out on the beaches of the PNAT islands and the coasts of the Pelagos Sanctuary in order to identify which are the most abundant waste and the sources and then guide the mitigation actions.

Keywords : Biodiversity , Hot Spots , Marine Litter , Marine Protected Areas , Mediterranean Sea , Microplastics , Monitoring , Pelagos Sanctuary

#### Session 24.6\_O. Chaired by Christophe Maes, Plouzané

# Towards a plastic pollution free Galapagos: Beaching variability of marine debris near the archipelago

*Ypma Stefanie, Van Sebille Erik, Jones Jen, Donnelly Andy.* Paper number 333876

### Closing the mediterranean marine floating plastic mass budget: Inverse modelling of sources and sinks

*Kaandorp Mikael, Dijkstra Henk, Van Sebille Erik.* Paper number 334120

## Global modelling of plastic beaching indicates coastlines and coastal waters as significant plastic reservoirs

*Onink Victor, Jongedijk Cleo, Hoffman Matthew, Van Sebille Erik, Laufkötter Charlotte.* Paper number 334255

#### Mapping the connectivity of the major rivers in Indonesia

*Maes Christophe, Dobler Delphine, Martinez Elodie.* Paper number 334381

# Towards a plastic pollution free Galapagos: Beaching variability of marine debris near the archipelago

Ypma Stefanie, Van Sebille Erik, Jones Jen, Donnelly Andy.

The Galapagos Archipelago and the Galapagos Marine Reserve host one of the world's most unique ecosystems. Although being a UNESCO world heritage site and being isolated from any dense population, over 8 tonnes of plastic are collected on the islands each year. To decrease the impact of plastic waste in the region, scientific evidence is needed on the sources and fate of the marine plastic. In order to do so, we combine hydrodynamic fields from ocean-, wave-, wind- and tide-models using the OceanParcels particle tracking framework to track virtual particles through the marine reserve. First results using this method indicate that non-local sources of plastics that reach the Galapagos Islands are likely specific areas along the mainland of South America and international fishing operations outside the Ecuadorian border of the Galapagos Marine Reserve. In addition, a beaching parameterization has been developed to quantify where and when virtual particles wash ashore. We will present composite analyses of pathways of virtual particles, beaching events and the connectivity between the different islands. These results show that the particle pathways and beaching strongly depend on the dry and wet seasons characteristic for the Galapagos Islands. This work, in combination with a growing observational data set, will form the basis of a predictive model that will support the Galapagos National Park in their efforts to free the Galapagos Archipelago from marine debris.

Keywords : archipelago , beaching , marine debris

# Closing the Mediterranean marine floating plastic mass budget: Inverse modelling of sources and sinks

Kaandorp Mikael, Dijkstra Henk, Van Sebille Erik.

Plastics entering the ocean face a fate which is still relatively unknown: how much of it will stay afloat, how quickly will it degrade, and how much of it will end up on the beach? Estimates of plastic inputs are orders of magnitude larger than the quantities found in the surface waters. In order to get a better understanding of the fate of these plastics, an inverse modelling methodology is presented here, where parameters in a Lagrangian ocean model are calibrated by comparing with field measurements of plastic concentrations. A case study is presented for the Mediterranean. Parameters for beaching, sinking, and for various sources of microplastics are estimated using surface water measurements. Results suggest that coastal population accounts for most plastic waste, about twice as much as river emissions. It is estimated that 49—63% of plastic waste ends up on coastlines, and 37—51% sinks down. The estimated input for 2015 of 2,100—3,400 metric tonnes per year is much less than previous estimates for the Mediterranean. This work is part of ongoing investigations into the marine plastic mass budget. The methodology is coupled to a fragmentation model, estimating size-classes of plastics in the environment. This enables us to estimate the magnitude of fragmentation as a sink, and to account for size-dependent transport processes. Adding more types of measurements, such as samples from beaches, should allow us to constrain the different parameters even more accurately in the future.

Keywords : beaching , inverse modelling , Lagrangian , Lagrangian modelling , mass budget , modelling , sinking , sources

# Global modelling of plastic beaching indicates coastlines and coastal waters as significant plastic reservoirs

Onink Victor, Jongedijk Cleo, Hoffman Matthew, Van Sebille Erik, Laufkötter Charlotte.

The distribution of plastic in the ocean is poorly constrained, with the mass of floating plastic at the ocean surface being orders of magnitude smaller than estimated plastic inputs. Coastlines likely contain significant amounts of plastic, but inconsistent methodologies between beached plastic observations prevent determining the mass and distribution of globally beached plastic. We present Lagrangian model sensitivity experiments to estimate the beached fraction of marine plastic and to investigate the global distribution of beached plastic on coastlines. We perform simulations where particles, representing masses of floating plastic, are inserted at the ocean coasts. The particles are then advected by surface currents (HYCOM/NCODA global reanalysis and surface Stokes drift from the WaveWatch III global reanalysis) for 5 years. Beaching is parametrized stochastically using exponentional probability. Here, we test the sensitivity to e-folding time scales between 1 and 100 days, applied when plastic is within the coastal zone, within 10km of the nearest coastline. Resuspension of beached plastic is parameterised exponentially with an e-folding timescale between 69 and 273 days. No other loss processes are implemented. Between 39-95% of floating plastic mass is beached after 5 years, with the beached fraction depending on the ratio between the beaching and resuspension timescales. In all simulations, at least 77% of floating plastic mass is found either beached or within the coastal zone, indicating coastal regions are a significant reservoir of mismanaged terrestrial plastic. The distribution of beached plastic is closely related to the input distribution, with the highest concentrations found in Southeast Asia and the Mediterranean. Our results highlight coastlines and coastal waters as important reservoirs of marine plastic debris and indicate a need for greater understanding of plastic transport near and at the coastlines.

Keywords : Beaches , Coastal , Lagrangian modelling , Plastic budget

#### Mapping the connectivity of the major rivers in Indonesia

Maes Christophe, Dobler Delphine, Martinez Elodie.

Under the auspices of AFD and IRD, a collaborative research project aims to support the monitoring and modeling of marine debris in the Indonesian Seas and in the South East region. The project goals at strengthening Indonesian institutions awareness and knowledge about marine pollution by plastics, in order to implement mitigation strategies. Due to their high durability and relative low cost, the plastic production keeps increasing every year, and a substantial quantity of it eventually enters and accumulates in the oceans. Emissions and transport of such litter materials are complex and depend on waste management, population density and hydrology, but the majority of these wastes enters into the oceans through rivers. The first priority is to understand their source and beaching locations and to identify the potential connectivity between them at the regional and basin scale of the oceans. A simple Lagrangian assessment of the dispersion originating from 21 major rivers in Indonesia is addressed with the specific goal of identifying the regional and temporal distribution of "beaching particles" along the coastlines. The present study is based on the state-of-art ocean modeling and should be viewed as a first effort to establish the river connectivity implied in the ocean dispersion of marine debris and litter in Indonesia.

Keywords : coastal impact , Indonesian seas , Lagrangian dispersion , river sources

#### Session 24.6\_Me. Chaired by Christian Laforsch, Bayreuth

**LIMNOPLAST microplastics in Europe's freshwater ecosystems: from sources to solutions** *Laforsch Christian.* Paper number 334554

**Interactions of microplastics with xenobiotics in the environment, influence of particle aging on sorption and desorption** *Hüffer Thorsten.* Paper number 334239

Limits of quantification – a (hopefully) provocative discussion on our ability to quantify small microplastics in complex matrices Vollertsen Jes. Paper number 334541

# LIMNOPLAST microplastics in europe's freshwater ecosystems: from sources to solutions

Laforsch Christian.

Microscopic plastic debris, so-called microplastics (MP), pose a global challenge. To meet this challenge, the EU-funded ITN-project LimnoPlast will for the first time bring together environmental, technical, and social sciences with the vision to transform a new understanding of freshwater MP to innovative solutions with experts from all over Europe. Exactly this trans- and interdisciplinary approach is also addressed within the planned program of this special session, with a heterogenous speaker composition regarding the expertise. As this is an ITN project with a special focus on early stage researchers (ESR), fortunately some ESRs are part of this submitted program and can present their research at this international conference. This special session will be closed with a panel discussion with 4 experts of the LimnoPlast consortium with different scientific background.

Keywords : behavioural approaches to the microplastics problem , freshwater ecosystems , mental models , microplastics , panel discussion , particle aging , pollution , quantification of small microplastics , risk perception , sorption and desorption

# Interactions of microplastics with xenobiotics in the environment - influence of particle aging on sorption and desorption

Hüffer Thorsten.

The pervasive use of plastics together with inadequate waste management has led to a global contamination of ecosystems with end-of-life plastic products. The global plastic pollution has been shown to pose a planetary threat because it is causing planetary-scale exposure that is not readily reversible. Thus, the presence of (micro-scale) plastics has received increasing attention over recent years, not only within the scientific community but also among regulators, policy makers and the general public. Originally, being a topic of interest in marine research, the concern about these materials has expanded to freshwater and most recently also to terrestrial and atmospheric systems. One of the major proposed knowledge gaps is the understanding of relevant environmental transformation and transport processes. Plastic particles released in the environment are subjected to various aging process including biotic (biofilm formation) and abiotic aging (UV radiation, oxidation, and physical stress). These processes lead to an alteration in the physical and chemical properties of the polymeric particle and may ultimately result in an embrittlement and fragmentation. Understanding the interactions between microplastics and those contaminants remains important for evaluating the materials' impact on the environment. Despite the number of sorption studies available for microplastics, there is little information on how ageing of the particles influences their properties and ultimately the sorption and leaching behavior.

Keywords : Organic Compounds , Plastics

#### Limits of quantification – a (hopefully) provocative discussion on our ability to quantify small microplastics in complex matrices

Vollertsen Jes.

The group of small particles we tend to call 'microplastics' covers a diverse group of manmade polymers. It also covers particles of natural polymers used by man, composite materials, particles of materials containing polymers, etc. Its chemical quantification hence becomes quite diverse. In addition we want to quantify polymer type, mass, particle numbers, sizes, shapes, and more. This further complicates its chemical quantification. At today's state-of-the-art, the scientific community is quite good at quantifying microplastics even in complex matrices, at least this statement holds true down to a certain size. The smaller the particles, the more difficult it is to sample them, extract them from the sampled material, and finally analyze them. But what is the size limit at which we are able to not just detect some particles in a matrix, but to arrive at a valid quantification? For sure this depends on the applied techniques for sampling, extraction, and analysis. For sure, an analytical technique like visual sorting is less able to quantify small particles than e.g. µRaman or µFTIR imaging. But how good is each method, and how honestly are these issues addressed when presenting scientific findings? Is it not so, that there is a strong tendency to state that the (size) limit of detection is the (size) limit of quantification? For example: I found a 20 µm microplastics in my matrix, hence I am able to quantify the amount of microplastics down to this size. And is it not true, that very few researchers do recovery tests, and those who do, typically use particles not necessarily representative of the ones in the analyzed matrix. This presentation discusses some issues about (size) limit of quantification and how these affect our measurements of microplastics in the environment.

Keywords : Microplastics , particles , polymers , quantification , sampling

#### Session 24.6\_Ma. Chaired by Bruno Tassin, Marne la Vallée

**Microplasticos in the Great Lakes** *Rios Mendoza Lorena*. Paper number 332439

**Why analysing microplastics in floodplain sediments is beneficial** *Lechthaler Simone.* Paper number 332825

**Drivers of microplastic toxicity on Daphnia magna** Zimmermann Lisa, Wagner Martin, Völker Carolin. Paper number 333105

#### **Microplasticos in the Great Lakes**

#### Rios Mendoza Lorena.

Microplastics (MPs) are becoming one of the most cited emergent contaminants in the last decade. These tiny synthetic polymers are associated with human activity. The inadequate disposal of plastics has made this material a ubiquitous pollutant on beaches, rivers, lakes, and oceans. MP particles are a new type of pollution reported in the Great Lakes with unknown impacts in the ecosystem and human health. Little information is currently available on the composition, distribution, or fate of MPs debris in the western end of Lake Superior and St. Louis River Estuary. This research aims to identify possible sources, abundance, and MPs' potential to adsorb toxic compounds. We collected 22 samples from nine main tributaries rivers to western Lake Superior and 12 Creeks, and one inland lake in Wisconsin during summer and fall of 2017 and 27 during summer 2016. To determine the type of synthetic polymer, we used an FTIR-ATR Micro Spectrophotometer. The MPs were classified by color, size, and morphology. Results showed that fibers and fragments were the main MPs. St Louis and French Rivers presented a high concentration of cotton fibers. The PAHs fingerprinting was related to pyrogenic sources.

Keywords : Freshwaters , Great Lakes , Microplastics , toxic compounds

#### Why analysing microplastics in floodplain sediments is beneficial

Lechthaler Simone.

Microplastics has already been detected in every environmental area and were thus defined as omnipresent anthropogenic contaminant. The current research focuses on understanding transport and sedimentation processes and detection of sinks. Thereby, fluvial systems are the predominant transport path for microplastic from land-based sources. Due to numerous sources along a river course, a continuous entry of microplastics is possible. If microplastics are transported in the river, floods can lead to sedimentation on the floodplains. Thus, they act as storage for microplastics which is why the analysis of floodplain sediments offers many information on flood related deposition and indicates a chronological context. So far, there are hardly any studies on microplastics in floodplains. Therefore, this study sampled at nine river sites (Inde River, Germany) depth profiles in floodplain areas which were examined for microplastics, grain size and heavy metals. In addition, composite samples from the riverbed and surface samples outside the flooding area were taken. An increase of microplastics [n/kg] from source to mouth was recorded and slip-off slopes showed higher microplastic abundance than cut-off slopes. Microplastics accumulated predominantly in fine grain sediments and the distribution along the river was similar to lead, which was exemplarily considered as one of the analysed heavy metals. As a new method, sedimentation rates of floodplains were determined based on microplastic detection. Furthermore, the temporal deposition of sediments was linked to the identified polymers where older polymers, such as polyethylene, were found in all layers and younger polymers, such as polyethylene terephthalate, only in the upper layers. The maximum time of deposition can be identified with the year of polymer development and therefore be used as a new dating method in the future. Overall, this study provides not only information on concentration of microplastics in floodplains but also on time related sediment deposition.

Keywords : Floodplain Sediments , Fluvial Systems , Microplastic Marker , Sedimentation Rates

#### Drivers of microplastic toxicity on Daphnia magna

Zimmermann Lisa, Wagner Martin, Völker Carolin.

Microplastics are a heterogeneous set of materials that differ not only in particle properties, like size and shape, but also in chemical composition, including polymers, additives and side products. Thus, we were interested in the driving factor of microplastic toxicity: Is it the chemicals or the particle itself? To address this question, we chronically exposed Daphnia magna to three types of irregular microplastics, polyvinyl chloride (PVC), polyurethane (PUR) and polylactic acid (PLA) microplastics as well as to natural kaolin particles, ; 59 µm. First, we compared the toxicity of the different materials by exposing the daphnids to high concentrations of particles (10, 50, 100, 500) mg/L). To investigate the drivers of toxicity, we used four exposure scenarios, including the original microplastics and microplastics from which we removed the extractable chemicals as well as the compounds extracted or migrating from the plastics to differentiate between chemical and particle toxicity. All tested microplastics negatively affected the life-history of D. magna. Exposure to PVC had the strongest effect on daphnid reproduction, PLA significantly reduced the survival. Thus, the toxicity of microplastics depends on the endpoint and the material. In addition, the results indicate that bio-based and biodegradable plastics can be as toxic as their conventional counterparts. Microplastics were more toxic than the natural particle kaolin when comparing numerical concentrations. Importantly, the contribution of plastic chemicals to the toxicity was also plastic type-specific. Plastic chemicals were the main driver for toxicity in case of PVC but not PUR and PLA microplastics. Our findings highlight that microplastics are not created equal: Multiple plastic types as well as their chemical composition need to be taken into account when assessing their environmental hazards. In addition, ecotoxicological research needs to move forward to investigating the impacts of so far understudied polymers, such as PVC, PUR and "bioplastics".

Keywords : bioplastic , chemical toxicity , ecotoxicology , effect , freswater invertebrate , leachates , particle toxicity

#### Session 24.7\_O. Chaired by Thierry Huck, Plouzané

#### A comparison of floating marine debris across three ocean basins.

Jones Lowenna, Duncan Emily, Penn Emily, Godley Brendan, Santillo David, Rendell-Bhatti Flora, Rowan Henthorn, Jucker Meret, Jimenez-Guri Eva. Paper number 334103

#### A spatially variable scarcity of floating microplastics in the eastern North Pacific Ocean

Egger Matthias, Nijhof Rein, Quiros Lauren, Leone Giulia, Royer Sarah-Jeanne, Mcwhirter Andrew, Kantakov Gennady, Radchenko Vladimir, Pakhomov Evgeny, Hunt Brian, Lebreton Laurent. Paper number 334131

#### The physical oceanography of the transport of floating marine debris

Van Sebille Erik, Aliani Stefano, Law Kara, Maximenko Nikolai, Alsina Jose M, Bagaev Victor, Bergmann Melanie, Chapron Bertrand, Chubarenko Irina, Cózar Andrés, Delandmeter Philippe, Egger Matthias, Fox-Kemper Baylor, Garaba Shungudzemwoyo, Goddijn-Murphy Lonneke, Hardesty Britta, Hoffman Matthew, Isobe Atsuhiko, Jongedijk Cleo, Kaandorp Mikael, Khatmullina Lilia, Koelmans Albert, Kukulka Tobias, Laufkötter Charlotte, Lebreton Laurent, Lobelle Delphine, Maes Christophe, Martinez-Vicente Victor, Morales Maqueda Miguel Angel, Poulain-Zarcos Marie, Rodriguez Ernesto, Ryan Peter, Shanks Alan, Shim Won Joon, Suaria Giuseppe, Thiel Martin, Van Den Bremer Ton, Wichmann David. Paper number 334133

**The global modeled sinking characteristics of biofouled microplastic** *Lobelle Delphine, Kooi Merel, Koelmans Albert A., Van Sebille Erik.* Paper number 334265 \_MICRO 2020. Fate and Impact of Microplastics: Knowledge and Responsibilities. 23-27 November 2020\_\_\_\_

#### A comparison of floating marine debris across three ocean basins

Jones Lowenna, Duncan Emily, Penn Emily, Godley Brendan, Santillo David, Rendell-Bhatti Flora, Rowan Henthorn, Jucker Meret, Jimenez-Guri Eva.

Synthetic debris, more specifically plastic pollution in ocean ecosystems, is now a global concern. Five major ocean gyres have been identified as holding vast quantities of floating debris, with concerns the Arctic is fast becoming a sixth accumulation zone. Disparities in methodologies and reported results make accurate and reliable comparisons among studies difficult. Here we present a comparable assessment of sea-surface debris concentrations across three ocean basins. Sea- surface trawl samples (n=44) were obtained from locations within the Arctic (ARC n=13), Atlantic (ATL n=24) and Pacific (PAC n=7) Oceans, reporting a 100% incidence of synthetic material. Primarily particles were: suspected plastic particles (SPP's; n=484 ARC: 65; ATL: 184; PAC: 299). The methodologies utilised here could be utilised by citizen science projects across the globe, generating essential, comparable baseline data of plastic debris whilst furthering awareness and education of the subject.

Keywords : FT , IR , Methodologies , Ocean , Polymer , Sample , Trawl

# A spatially variable scarcity of floating microplastics in the eastern North Pacific Ocean

Egger Matthias, Nijhof Rein, Quiros Lauren, Leone Giulia, Royer Sarah-Jeanne, Mcwhirter Andrew, Kantakov Gennady, Radchenko Vladimir, Pakhomov Evgeny, Hunt Brian, Lebreton Laurent.

Plastic waste accumulating in the global ocean is an increasingly threatening environmental issue. To date, the floating and thus most visible fraction of ocean plastic pollution has been mapped at global scale. Yet, large knowledge gaps exist in our current understanding of the transport and transformation processes of positively buoyant plastic debris at the sea surface. Observations at sea typically report an apparent scarcity of microplastics (; 5 mm) relative to the expected abundancesize distribution based on fragmentation of larger plastic objects. Here, we provide a comprehensive study on the relative abundance of microplastics (¿ 500 µm) and mesoplastics (0.5-5 cm) in the surface waters of the eastern North Pacific Ocean using data from 1'136'040 plastic fragments collected by 679 neuston trawl deployments between 2015 and 2019. Our results reveal that the apparent microplastic scarcity is not uniformly distributed across the region. Instead, we show that the relative abundance of floating microplastics increases from the outside to the inside of the North Pacific Garbage Patch. We hypothesize that this observation could be explained by (i) a spatially variable microplastic removal due to spatial differences in ocean productivity, (ii) a differential dispersal of micro- vs. mesoplastics with a preferential accumulation of microplastics in the subtropical gyre, and/or (iii) the timescales associated with transport and fragmentation of plastic objects at the ocean surface with older, more degraded, floating plastic accumulation in subtropical gyres. The results presented here highlight that global estimates of the accumulation and removal of positively buoyant microplastics need to consider spatial aspects such as variations in ocean productivity, the dominant physical transport processes in a given area, as well as the time needed for a plastic object to reach the specific offshore location.

Keywords : floating microplastics , North Pacific Garbage Patch , Ocean plastic pollution , ocean surface

#### The physical oceanography of the transport of floating marine debris

Van Sebille Erik, Aliani Stefano, Law Kara, Maximenko Nikolai, Alsina Jose M, Bagaev Victor, Bergmann Melanie, Chapron Bertrand, Chubarenko Irina, Cózar Andrés, Delandmeter Philippe, Egger Matthias, Fox-Kemper Baylor, Garaba Shungudzemwoyo, Goddijn-Murphy Lonneke, Hardesty Britta, Hoffman Matthew, Isobe Atsuhiko, Jongedijk Cleo, Kaandorp Mikael, Khatmullina Lilia, Koelmans Albert, Kukulka Tobias, Laufkötter Charlotte, Lebreton Laurent, Lobelle Delphine, Maes Christophe, Martinez-Vicente Victor, Morales Maqueda Miguel Angel, Poulain-Zarcos Marie, Rodriguez Ernesto, Ryan Peter, Shanks Alan, Shim Won Joon, Suaria Giuseppe, Thiel Martin, Van Den Bremer Ton, Wichmann David.

Marine plastic debris floating on the ocean surface is a major environmental problem. However, its distribution in the ocean is poorly mapped, and most of the plastic waste estimated to have entered the ocean from land is unaccounted for. Better understanding of how plastic debris is transported from coastal and marine sources is crucial to quantify and close the global inventory of marine plastics, which in turn represents critical information for mitigation or policy strategies. At the same time, plastic is a unique tracer that provides an opportunity to learn more about the physics and dynamics of our ocean across multiple scales, from the Ekman convergence in basin-scale gyres to individual waves in the surfzone. In this review presentation, we comprehensively discuss what is known about the different processes that govern the transport of floating marine plastic debris in both the open ocean and the coastal zones, based on the published literature and referring to insights from neighbouring fields such as oil spill dispersion, marine safety recovery, plankton connectivity, and others. We discuss how measurements of marine plastics (both in situ and in the laboratory), remote sensing, and numerical simulations can elucidate these processes and their interactions across spatio-temporal scales. We provide ideas on how to further our understanding of the transport of plastic litter across various scales.

Keywords : fluid dynamics , marine debris , ocean circulation , physical oceanography , remote sensing

#### The global modeled sinking characteristics of biofouled microplastic

Lobelle Delphine, Kooi Merel, Koelmans Albert A., Van Sebille Erik.

Microplastic debris ending up at the sea surface has become a ubiquitous issue in the modern day and the characterisation of the sinking of floating microplastic on a global scale still remains largely unknown. Here, we map the modeled effects of biofouling (algal growth on the substrate) on the removal of microplastic from the sea surface to deeper depths. Using NEMO-MEDUSA output to simulate the hydrodynamic, biological and physical properties of the seawater combined with a particle-tracking framework, we estimate the sinking onset timescale of each virtual particle and the time required to reach its first sinking depth (i.e. the depth it reaches when its vertical velocity becomes zero for the first time). Different sizes and types of plastic (with different densities) are simulated, where our results show that sinking characteristics are largely size-dependent, whereas all plastic densities produce a very similar global distribution of the sinking onset time. Generally, oligotrophic regions with low algal concentrations result in particles not sinking within the 90-day simulation time for particles between 10 mm and 10  $\mu$ m. On a global scale, the smallest particles simulated here (0.1  $\mu$ m) sink almost immediately and their trajectories produce the longest time to reach their first sinking depth. The impact of advection and seasonality on the sinking characteristics of the virtual microplastic is small.

Keywords : Biofouling , Lagrangian transport , Sinking

#### Session 24.7\_Me. Chaired by Fan Liu, Aalborg

**Elements of a risk/benefit governance framework for microplastics** Kai Purnhagen

**Risk perception and mental models regarding microplastics in Norway** *Felipe Rodriguez Marcos, Böhm Gisela, Doran Rouven.* Paper number 334401

**Behavioural approaches to the microplastics problem and solutions** *Grünzner Maja, Pahl Sabine, White Mathew, Thompson Richard.* Paper number 334512

#### Risk perception and mental models regarding microplastics in Norway

Felipe Rodriguez Marcos, Böhm Gisela, Doran Rouven.

Plastics are indispensable in daily life but have become a planetary threat. They can be produced as or degrade into microplastics (MP), which have been detected in marine and freshwater environments across the globe. Previous literature points towards the need for an interdisciplinary approach combining natural and social sciences in addressing the problem. This includes an understanding of the role of the public in aggravating and /or mitigating the problem, particularly regarding risk perception and associated behavioral responses. The current presentation reports an empirical study that aims to investigate lavpeople's intuitive understanding of MP. For this purpose, we asked an online sample (¿N=1000) from the Norwegian public to state what they think when they read or hear the word 'microplastics.' They were instructed that they should write down the first thing that came to their minds, and that their answer would ideally include a few sentences, although just some few words were fine, too, if they did prefer this themselves. All obtained answers were coded in accordance with a coding scheme, which was developed in line with prior psychological research on risk perception and mental models in an environmental context. The analyses attended to both the content and structure of the responses attempting to identify shared mental models, especially when it comes to beliefs about causes and impacts of MP. Understanding these mental models addresses some existing knowledge gaps in the social science literature on MP, which may assist in finding a societal response to an eminent environmental problem.

Keywords : mental models , microplastics , risk perception

#### Behavioural approaches to the microplastics problem and solutions

Grünzner Maja, Pahl Sabine, White Mathew, Thompson Richard.

We humans created the plastics age (Thompson et al., 2009; Yarsley & Couzens, 1945). While European citizens are concerned about plastic pollution and its effects on the environment (Eurobarometer, 2020), microplastics, specifically in freshwater systems, have been highlighted as a major pollutant with an urgent need for solutions (Wagner & Lambert, 2018; Zeng, 2018). Change of consumption behaviour (33%) was selected most frequently, when Europeans were asked about choosing the most effective actions to tackle environmental problems (Eurobarometer, 2020). Citizens are aware of the problem and as they believe that their behaviour matters, the present work, under the Horizon2020 LimnoPlast project, aims to provide a novel approach, in complementing the findings of environmental and technical research. This is achieved by contributing a social science perspective, focusing on identifying behaviours leading to microplastics pollution while simultaneously looking into behavioural aspects of innovations tackling microplastics. Our presentation will provide an overview of existing behavioural approaches regarding this "wickedproblem". These insights can help to inform future empirical studies on how to tackle microplastics pollution from a social and behavioural science perspective. After all, we humans, are the cause and the solution (Pahl et al., 2017).

Keywords : Behaviour , Microplastics Solutions , Social Science

#### Session 24.7\_Ma. Chaired by Miriam Weber, Bühl

**Biodegradable plastics opportunities and challenges** *Weber Miriam, Schlegel Katharina, Battagliarin Glauco.* Paper number 331682

# Reliable test methods to evaluate the biodegradation of plastic materials in the marine environment for LCA

Lott Christian, Eich Andreas, Schlegel Katharina, Biedermann Eynat, Becker Chong, Lasut Markus T., Weber Miriam. Paper number 334516

# State of the art and gaps of standard test methods and specifications for biodegradation of plastics in the open environment

Kuhfuß Hanna, Eich Andreas, Lott Christian, Weber Miriam. Paper number 334553

**Options for a feasible risk assessment of biodegradation of plastics in the open environment** *Weber Miriam, Eich Andreas, Kuhfuß Hanna, Lott Christian.* Paper number 334523

#### **Biodegradable plastics - opportunities and challenges**

Weber Miriam, Schlegel Katharina, Battagliarin Glauco.

Biodegradable plastics (BDPs) are a growing market and are discussed as options to mitigate environmental plastic pollution. In some cases, BDPs are applied as substitute materials and are exempt from regulations. One of the most prominent examples is the current discussion concerning the restrictions on intentionally added microplastics from ECHA. In addition, BDPs are also perceived as a more sustainable alternative for applications in the natural environment (e.g. agriculture, fisheries) and where loss is intrinsic to the use (abrasion, wear & tear). However, biodegradability, as an inherent material property, also brings along (new) risks for the environment such as carbon enrichment, release of greenhouse gasses, and for society such as dis-/misinformation, greenwashing and the possibility of favoring littering. Additionally, the work on methods to characterize the biodegradation of these materials and their degradation products in different compartments are sometimes questioned and are still objects of research. For this session we invite contributions on the performance of BDPs in the natural environment, their environmental effects both on ecosystem and organism levels and the methods related to the above-mentioned aspects. Societal effects, life-cycle assessment and substitution potential might also be addressed.

Keywords : a communication maze , Bioplastics , certification and regulation , Standardization , State of the art of biodegradable plastics , Sustainability value

# **Reliable test methods to evaluate the biodegradation of plastic materials in the marine environment for LCA**

Lott Christian, Eich Andreas, Schlegel Katharina, Biedermann Eynat, Becker Chong, Lasut Markus T., Weber Miriam.

Current life cycle analysis (LCA) methodologies lack indicators to assess the biodegradation of plastic materials introduced to the natural environment. If ending up in the sea, biodegradable plastics are assumed to be less persistent than conventional ones. However, reliable methods and data on the biodegradation of any plastic material in the ocean are not systematically available. We applied a 3-tier test scheme of reliable and environmentally relevant methods in laboratory, mesocosm and field tests to assess the biodegradation as well as the disintegration performance of several plastics including certified compostable materials under real environmental conditions in coastal habitats of Southern Europe and SE Asia. The mathematical modeling of the experimental results and the calculation of a specific half-life of a plastic material under specific environmental conditions show that the persistence is highly depending on where a certain plastic item is ending up. The specific half-life as a material property thus allows for the numerical comparison of different environmental scenarios and can be fed into LCA models.

Keywords : biodegradable plastic , half , LCA , life , standard test

# State of the art and gaps of standard test methods and specifications for biodegradation of plastics in the open environment

Kuhfuß Hanna, Eich Andreas, Lott Christian, Weber Miriam.

Standardization is used to achieve comparability, by defining details of procedures and test conditions, helping stakeholders such as consumers and regulators. Standards containing comprehensive information are also called 'specification'. They are used as guidelines and are an explicit set of requirements to be satisfied by a material, product, system or service, as for example for the claim 'industrial compostable' or 'soil biodegradable'. Such standards can also be set up as 'over-arching or comprehensive standards' and are used as a baseline for certification programs. Standard test methods mostly provide detailed directions on performing specific tests, which produces a test result and the basis for accreditation programs. That way, results from different institutions are better comparable and assure a certain quality level. For assessing the biodegradation in the open environment standard test methods and specifications are available at different stages. Most advanced are the tests for seawater, then tests for soil and least advanced are the tests for freshwater. In general, it can be said that there are rather few standard tests available at all. This is due to the fact that they were developed quite specifically and rather for individual applications. For the open environment, the diversity of conditions that are crucial for degradation cannot be ignored. The assessment of biodegradation under different environmental conditions such as soil type, water content, sediment grain sizes, nutrients, oxygen concentration, etc. are not yet included in the available tests. Such data collection would allow to take into account the diversity of the receiving environment, i.e. soils, freshwater and seawater habitats and to indicate the biodegradation rate as a range. In summary, if needed the existing standards should be adapted according to relevant criteria, and if missing be set up for the relevant environmental conditions.

Keywords : Biodegradable , Certification Baseline , Compostable , Freshwater , Guidelines , Open Environment , Seawater , Soil , Standard Test Methods , Standardization

# Options for a feasible risk assessment of biodegradation of plastics in the open environment

Weber Miriam, Eich Andreas, Kuhfuß Hanna, Lott Christian.

Biodegradable plastic materials are increasingly being discussed as an alternative for conventional non-biodegradable plastic and as a mitigation strategy against plastic pollution, especially for items with an intentional input (e.g. mulch films, etc.), with a high risk of loss (e.g. fishing devices, etc.) and where loss is intrinsic to use (e.g. abrasion of aquaculture nets, etc.). Regardless of the source of the waste, all this contributes to the accumulation of plastic in the open environment. The question for society is how to deal with biodegradable plastic known to end up in the open environment. We compiled options for biodegradation testing schemes showing several scenarios. They are based on the delicate balance of either limited informative value or increased costs. The testing schemes include the proof of biodegradability in a laboratory test. If this test is successfully completed, the proof for environmental safety shall be provided by at least performing one ecotoxicity test. The advantage of such an approach is its low costs however, the information usable for an environmental risk assessment (ERA) remains minimal. Increased information could come through a proof for environmental safety provided by ecotoxicity testing on several trophic levels and impact on ecosystem level. To assess the range of lifetimes in the environments of interest field and tank tests should be performed and data fueled into LCA and LCIA. The disadvantages are increased costs and that the risk of waste accumulation is not yet categorizeable. In order to increase the usability of such information for risk assessment mathematical modelling for scenarios of 'no accumulation' is suggested if data quality is sufficient. The higher costs for testing, should be reduced by the development of an economically feasible evaluation scheme with a set of harmonized standard test methods and specifications.

Keywords : ecotox tests , impact tests , life cycle assessment , modelling , testing scheme

Poster sessions 24.8, 24.9 and 24.10

#### Session 24.8\_O. Chaired by Manuel Monge-Ganuzas, Urdaibai

**Is wind a good proxy for floating microplastics? A case study in Galway Bay, Ireland** *Frias João, Lyashevska Olga, Haleigh Joyce, Pagter Elena, Nash Roisin.* Paper number 331745

Microplastic pollution in beaches around Iskenderun Bay, located in the Northeastern Levantine coast of Turkey *Çevik Cem, Gundogdu Sedat.* Paper number 333122

Microplastics in the Maldonado stream basin (Maldonado, Uruguay): assessment and analysis of this new vector of pollution De Feo Bárbara, Krojmal Evelyn, Lozoya Juan Pablo, Gonzáles Magda, SuÁrez BÁrbara, Teixeira De Mello Franco. Paper number 334598

# Is wind a good proxy for floating microplastics? A case study in Galway Bay, Ireland

Frias João, Lyashevska Olga, Haleigh Joyce, Pagter Elena, Nash Roisin.

According to a recent report, floating microplastic debris at the ocean's surface represent approximately 1% of all plastics found in the environment, while 99% are thought to either be deposited at along coastlines or at the bottom of the ocean. This exploratory research work conducted on Galway Bay, a coastal bay located in the Northeast Atlantic Ocean, assesses the density of floating microplastics and the potential influence of wind in their dispersal. Twenty manta trawl samples retrieved a total of 1182 microplastics from a total surface seawater volume of 2039.86 m3. The average microplastic density ( $0.56 \pm 0.33$  MP m-3), which mainly comprised fibres (n=1017), followed by fragments (n=148), fishing line (n=9), beads (n=3), films (n=2) and foams (n=2). This study reports for the first time primary microplastics (microbeads) floating in Irish coastal waters, which could probably result from personal care items, although source it is not obvious. Black was the predominant microplastic colour, followed by blue and red. This exploratory study provides a snapshot of microplastics density in Galway Bay where the results show no obvious pattern of microplastic distribution based solely on wind speed and direction. Therefore, oceanic currents, like other authors suggest, seem to play a more influential role on surface microplastic distribution. Although the abundance of microplastic densities is slightly higher than similar bays around the world, they are in the same order of magnitude. This work highlights that microplastics in surface waters are a multifaceted issue which requires multiple sample collection points for coastal monitoring in order to reflect the area being sampled and ensuring a more holistic approach.

Keywords : microbeads , microplastics , monitoring , MSFD , water surface

# Is wind a good proxy for floating microplastics? A case study in Galway Bay, Ireland

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# INTRODUCTION

accounts for 5% and most plastics are thought to be deposited at the bottom of the plastic distribution in the environment. Deposition in beaches and/or coastal areas webs. It is estimated that floating microplastics at the ocean's surface represent ~1% of marine wildlife, particularly planktonic organisms', who are the basis of aquatic food exponentially<sup>4</sup>. Among marine litter, microplastics (MPs)<sup>5,6</sup> represent higher risks for environment at unprecedented rates<sup>1,2,3</sup>, while global production continues to rise ocean (94%)° Plastic items of all shapes, colours and sizes are stockpiling globally in the marine

of floating microplastics main goals of the study was to assess whether wind is a good proxy to assess hotspots Bay, as well as to find a representative baseline for this emerging pollutant. One of the This exploratory work intended to assess the density of floating microplastics in Calway

# MATERIALS AND METHODS

30 × 10 cm<sup>2</sup> cod end was attached. and 61 cm wide, to which a 2 m long rectangular aperture of 15 cm high made of aluminium and has a (Fig. 3) collected on board of the RV Celtic Seawater surface samples were 300 µm mesh size nylon net with a Voyager (Fig. 2), using a manta trawl The manta trawl frame is



igure 3. Manta trawl used for sampling

coordinates each tow, as well as the initial and final GPS revolutions were registered prior to and atter flowmeter with back-run stop, equipped with a Hydro-Bios mechanical whose



diameter of 11 cm. The trawl frame was also The cod end was made of 3-mm thick grey PVC tubes with a total length of 23 cm and a

The manta net was deployed from the starboard side of the vessel, at an average

added to the jar. Samples were immediately frozen -20 °C, without adding fixing larger >5 mm were carefully removed using metal tweezers, and not accounted for decontaminated stainless-steel sieves (100 and 300  $\mu$ m). All natural and artificial litter solutions, until further analysis. here. The sieve was rinsed with filtered seawater three times and the contents were water from the outside, and the sample washed through a set of previously from Viršek et al. (2016)<sup>9</sup>, briefly the cod end was then thoroughly rinsed with ultrapure speed of 2 knots for approximately 20 min, following an adapted methodology

details on the method used can be found in Frias et al. (2020)  $^{10}$ potassium hydroxide (KOH) solution. **Fig. 4** illustrates the laboratory procedure. Full In the laboratory samples were defrosted and alkaline digested, using a 10%



Figure 4. Simplified laboratory procedure

# SAMPLING SITE

Galway Bay is a large semi-enclosed bay, located in the West of Islands, as seen in left side of Fig. 1. a. Ireland. It is protected by the strong Atlantic Ocean swells by the Aran

bay. All trawls are independent and were dependent on weather of microplastics into the bay (e.g. the River Corrib). Sampling stations and December 2017 (Fig. 1. b.). Stations were selected taking into A total of 20 manta trawl samples were collected between October conditions several transects were made tollowing circulation patterns within the Galway City towards Blackhead, Co. Clare; and in the Outer Bay followed transect lines: in the Inner Bay the transect went from consideration proximity to Galway City and potential inflow sources



Figure 1. a. Calway Bay and inner and outer bay boundary. b. Starting (purple) and ending point (blue) for eac trawl.

CONCLUSIONS

A total of **1182 MPs** were retrieved from **2039.86 m<sup>3</sup>** of seawater

# RESULTS

as seen in Fig. 5. microplastic colours were black, blue and red, the total and **fragments 16%**. The predominant colour, size and shape, as described in Fig. 4. In Bay surface seawaters. These were diverse in A total of 1182 MPs were retrieved from Galway relation to MPs types, fibres represented 81% of



fibre fragment others ■black ■blue ■other ■red

Figure 5. Predominant MPs types and colours

be found in Fig. 6.



hydrodynamic currents<sup>11</sup> seem to play a influence by the dominant wind. Strong SW winds and five stations do not exhibit the dominant wind speed and direction while sampling (B). Most stations are influenced by

stronger in role in surface distribution compared to winds in the Bay speed and direction 20 manta trawls and  ${\bf B}.$  stacked wind rose with dominant wind speed and direction (m  $s^{-1}).$ Figure7. A. Micro Microplastic density (MP m<sup>-3</sup>) azimuths for the



Monitoring of microplastics in seawater surface, particularly in bays,

No obvious density distribution pattern was observed

surface currents speed and direction

Wind by itself cannot serve as a proxy for oceanographic data on Water circulation in this bay seems to be more influential than wind The average MP density in Galway Bay is 0.56 ± 0.33 items m<sup>-3</sup>

C. film, D. fragments, E. fibres F. paint chip (potential vessel) retrieved. A.&B. microbeads Figure 4. Microplastics

contamination)

The authors recommend a holistic approach to sample collection

conditions) that might play an important role in MP distribution influential factors (e.g. river input, population density, atmospheric variability among samples & sites, as well as to have an overview of should rely on at least 6-10 different sampling sites to account for

environmental variables (wind speed & direction, precipitation

CONTACTS

joaotrias.com frias@gmit.ie (surface water, sediment, biota, etc.), and recording as many i.e. collection of samples from different matrices in the same sites

than in the Inner Bay (0.62  $\pm$  0.40 items m<sup>-3</sup>). Variability in MPs densities can Microplastic density was higher in the Outer Bay (0.46  $\pm$  0.16 items m<sup>-3</sup>)



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An Chomhairle um Thaighde in Éirinn CAROLINE

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# Microplastic pollution in beaches around Iskenderun Bay, located in the Northeastern Levantine coast of Turkey

Çevik Cem, Gundogdu Sedat.

The determination of the microplastic distribution is beneficial as a measure of the potential effects on the environment. Particularly in semi-closed ecosystems such as the Mediterranean Sea, this is getting more important. The Mediterranean Sea has a high risk of pollution as it was enclosed by highly populated and industrialized countries. The most important type of pollution that threatens the Mediterranean Sea is the pollution caused by plastics. The most important type of plastics that threatens the Mediterranean ecosystem is the microplastic pollution. Here, for the first time, we determined the level of microplastic pollution in beaches around Iskenderun Bay, located in the Northeastern Levantine coast of Turkey. For this purpose, microplastic pollution was determined in May 2018 at 13 locations. Sampling was carried out from each station during May 2018, especially during periods of minimum wave and wind conditions. Each station was assumed to have a homogeneous distribution of microplastics. Sampling was carried out at the points determined by random sampling method with the help of 1x1 m quadrates to obtain the first 5 cm depth of the sand. Microplastic concentration were found 100.7±35.7 pcs/kg (1044±219.2 pcs/m2). The highest microplastic concentration was observed in Dörtvol location with  $658.3 \pm 42 \text{ pcs/kg}$ . In all stations, 5 different types of microplastics (Fibre/Filament, Film, Fragment, Foam, Pellet/Granular) were found and the most frequent microplastics were found to be fragment type microplastics. Microplastics in the 1-5 mm length group were the most common size group. The results of this project show that the coasts of Iskenderun Bay are threatened by high levels of microplastic pollution. This study was supported by the Turkish Scientific and Technological Research Council (TUBITAK) as part of project number 117Y212 entitled "Determining the State of Microplastic Pollution in the Sandy Beaches around Iskenderun Baÿ.

Keywords : beach , Microplastics , northeastern mediterranean sea , plastic pollution
# COAST OF TURKEY microplastic pollution in beaches around iskenderun bay, located in the northeastern levantine

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### Abstract

by random sampling method with the help of 1x1 m quadrates to obtain and wind conditions. Sampling was carried out at the points determined locations. Sampling was carried out during periods of minimum wave determined the level of microplastic pollution in beaches around pollution caused by plastics and microplastics. Here, for the first time, we important type of pollution that threatens the Mediterranean Sea is the enclosed by highly populated and industrialized countries. The most important. The Mediterranean Sea has a high risk of pollution as it was closed ecosystems such as the Mediterranean Sea, this is getting more measure of the potential effects on the environment. Particularly in semithe first 5 cm depth of the sand. Iskenderun Bay, located in the Northeastern Levantine coast of lurkey -or this purpose, microplastic pollution was determined in May 2018 at 13 The determination of the microplastic distribution is beneficial as a







# Results and Discussion

microplastic pollution. the coasts of Iskenderun Bay are threatened by high levels of microplastics (Fibre/Filament, Film, Fragment, Foam, Pellet/Granular) were the most common size group. The results of this project show that ragment type microplastics. Microplastics in the 1-5 mm length group vere tound and the most trequent microplastics were tound to be förtyol location with 658.3  $\pm$  42 pcs/kg. In all stations, 5 different types of cs/m2). The highest microplastic concentration was observed in icroplastic concentration were found 100.7±35.7 pcs/kg (1044±219.2

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### Microplastics in the Maldonado stream basin (Maldonado, Uruguay): assessment and analysis of this new vector of pollution

De Feo Bárbara, Krojmal Evelyn, Lozoya Juan Pablo, Gonzáles Magda, SuÁrez BÁrbara, Teixeira De Mello Franco.

Plastics are one of the most common and persistent pollutants in aquatic ecosystems worldwide. They have significant harmful effects on both marine and freshwater ecosystems. Numerous investigations were made in marine ecosystems but too little is known of the freshwater ecosystems, and for this, the generation of basal information is a priority. From the continent, plastic waste is transported through river systems (e.g. rivers and streams) and urban drains, eventually reaching coastal areas and oceans. When analyzing these contributions, it is essential to consider the characteristics of the lotic system analyzed at a basin scale, including human activities in the area of influence. The Maldonado stream is the main fluvial system in Maldonado, Uruguay, a direct tributary of the Atlantic Ocean. In its basin, this stream crosses agricultural and livestock areas, and borders the cities of San Carlos and Maldonado. These different activities, and especially urban areas, can contribute with various pollutants, among which plastic waste stands out. The aim of this study is to carry out the first detailed analysis of presence, distribution, and transport of microplastics along the Maldonado stream basin. Covering from the upper of the basin to the mouth of the Maldonado river in the Atlantic Ocean, surface water was sampled using a motor pump and a skimmer (aprox. 1500liters) in 5 sites throughout 2019 (seasonal frequency). Densities were determined considering fibers larger than 0.5mm and fragments larger than 0.1 mm. All these items were classified and quantified according to their type, size and color. The first results show a total density of 323 items.m-3 in May and 236 items.m-3 in July, with fibers as the most represented type of MP in both samples (93% and 7%, 90% and 10%, respectively). Our results provide the first baseline information of microplastics on this coastal stream basin.

Keywords : Maldonado , microplastics , pollution , stream basin



### Microplastics in the Maldonado stream basin (Maldonado, Uruguay): evaluation and analysis of this new vector of pollution

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### 1. INTRODUCTION

Plastics and microplastics (MPs) are one of the most common and persistent pollutants in aquatic ecosystems worldwide. They have significant harmful effects on both marine and freshwater ecosystems. Numerous investigations were made in marine ecosystems but too little is known about the situation in freshwater ecosystems, and for this, the generation of basal information is a priority.

From the continent, plastic waste is transported through fluvial systems and urban drains to the oceans. When analyzing these contributions, it is essential to consider the characteristics of these dynamic systems from a watershed scale, including human activities and their influence areas.

The Maldonado stream, with a basin of about 1,400 km<sup>2</sup>, is the main stream in Maldonado, Uruguay, and is a direct tributary of the Atlantic Ocean. In its basin, this stream crosses agricultural and livestock areas, and borders the cities of San Carlos and Maldonado. These different activities, and especially urban areas, contribute various pollutants, among which plastic waste stands out. This study provide the first assessment of MPs on the Maldonado stream basin.



Figure 1. Location of the Maldonado stream basin. Sites 1 and 2 are located upstream San Carlos city. Sites 3, 4 and 5 are located downstream San Carlos city.

### 2. OBJECTIVES

### General

 Carry out the first detailed analysis of the presence, distribution, and transport of microplastics in the Maldonado stream basin.

### **Specific**

- Classify and quantify MPs according to type, size and colour.
- Determine the MPs density (items/m<sup>3</sup>) in the different sampled sites.
- Evaluate the relationship between the main urban centers of the Maldonado stream basin with the abundance and composition of MPs.

### 3. METHODS

- Water samples were taken with a pump that filtered on average 1,25 m<sup>3</sup> of superficial water in five sites of the basin, throughout 2019 and with a seasonal frequency.
- Samples were filtered with a 100 µm mesh and the retained material was deposited in a glass petri dish for direct observation with a stereo microscope.
- The MPs found were measured, quantified and classified according to type (i.e. fiber, fragment, foam, pellets, films), size and colour.
- Just fibers greater than 1.0 mm and fragments greater than 0.5 mm were taken into account.
- A laboratory contamination control was placed near the stereo microscope and MPs found were verify with a microscope with polarized light.
- Statistic analysis were made with 2 ways ANOVA test after evaluating normality and homogeneity of variances.



Figure 2. Pump sampling of surface waters, and filter column.

### 4. RESULTS

- MPs average total densities in site 1 and 2 were 47,6, in 3 and 4 were 29,5 and in 5 were 50,7 items/m<sup>3</sup>. On average, site 1 and 2 had the highest densities of MPs, while site 3 and 4 had the lowest.
- Fibers were the most represented type.
- Fibers densities had no significant difference between samples (f= 1,5; p= 0,2) or sites (f= 2,3 ; p= 0,08).
- Fragments densities had significant difference between samples, with October having the highest densities (f= 7,0; p= 0,003).
- Significant differences were found in total densities of MPs in sites (f= 3,1; p= 0,028) and month sampled (f= 3,3; p= 0,0049).

### 4. RESULTS



Figure 3. Average total density of MPs (A), fibers (B) and fragments (C) in each site, for the three sample periods.



Figure 4. Percentage of fibers and fragments found in each site, for the three sample periods.

### **5. CONCLUSIONS**

- The presences of MPs was ubiquitous in each site in March, July and October.
- The proximity to urban areas is not the only factor that determine the presence of MPs in freshwater systems.
- Other factors, such as wind may be influencing the dispersion of MPs (Gonzalez-Pleiter, et al., 2020).
- In site 5, the intrusion of the ocean to the Maldonado stream may be contributing with MPs.

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### Session 24.8\_Me. Chaired by Maria Murcia, Menorca

Microplastics in marine species within the Wadden Sea along the coastline of Lower Saxony, Germany

Schwarz Matthias, Polt Laura, Dau Kirsten, Fischer Elke. Paper number 329233

### Occurrence of microplastics in the gastrointestinal tracts of *Chelon saliens* along the Turkish coast

*Gundogdu Sedat, Çevik Cem, Temiz Ataş Nihan.* Paper number 333108

### Microplastics in marine species within the Wadden Sea along the coastline of Lower Saxony, Germany

Schwarz Matthias, Polt Laura, Dau Kirsten, Fischer Elke.

The aim of the study is to evaluate the occurrence and distribution of potential microplastic contamination in species of the Wadden Sea of Lower Saxony, Germany and to develop prerequisites for future monitoring approaches. Furthermore, potential correlated species- and individual-related factors such as size/weight, trophic level, feeding strategy are assessed and spatial gradients and further influencing factors are evaluated. Fish samples were taken in July 2018, invertebrate samples in September 2019 and June/July 2020 at six locations along the North Sea coastline of Lower Saxony, Germany. The species studied included the benthic species blue mussel (Mytilus edulis), common periwinkle (Littorina littorea), lugworm (Arenicola marina), mud shrimp (Corophium spp.) and the demersal fish species flounder (Platichthys flesus). Subsequently to sampling, the whole organisms and digestive tracts of Platichthys flesus were investigated at the laboratory of MRC. Laboratory analysis comprised dissection and digestion of biogenic organic material with a solution of potassium hydroxide and sodium hypochlorite. Samples were transferred to cellulose filters (613, VWR International, 5-13 µm retention) and stained with the lipophilic dve Nile Red (1 mg/ml in chloroform) prior to counting and scaling of microplastic particles implementing fluorescence microscopy (Axioscope 5/7 KMAT, Zeiss). A subset of particles identified as synthetic polymers was additionally analyzed with µRaman imaging (DXR2xi Raman Imaging Microscope, Thermo Fisher Scientific). Results on microplastic findings both for the investigated species, potential spatial patterns and influencing parameters will be presented.

Keywords : fish , invertebrates , microplastics , North Sea , Wadden Sea





### Microplastics in marine species within the Wadden Sea along the coastline of Lower Saxony, Germany

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### Introduction

Studies on selected benthic and demersal species in the Schleswig-Holstein Wadden Sea have shown that large proportions of the animals studied in almost all of the species were contaminated by microplastics (Fischer 2019). In the species investigated, the studies resulted in significant differences and a spatial gradient tendency with increasing contamination from north to south. This study covered the northern coastline of Schleswig-Holstein only, an investigation along the coastline of Lower Saxony has not been carried out and is targeted within the present study. Thus, the project will contribute to implementing requirements according to MSFD by: recording and evaluating the nature and extent of microplastics in biota based on five selected species at six investigation sites; major objectives are:

- determining differences in microplastic contamination of the species studied and major influencing factors;
   showing potential spatial gradients and
- based on the results, making recommendations for a future monitoring strategy to detect microplastics in biota in the coastal waters of Lower Saxony, Germany.

### **Material and Methods**

### Study Area



Fig. 1: Sampling sites along the Lower Saxony coastline: west (left) and east (right) invertebrates (1 – Dollart, 2 – Leybucht, 3 – Neuharlingersiel, 4 – Tettens, 5 – Jadebusen, 6 – Cappel/Neufeld) and pisces (P1 - Borkum, P2 - Baltrum, P3 - Jadebusen, P4 – Weser estuary (AuWe)).

### Sampling campaigns

Sampling of invertebrates took place in August 2019 and June/July 2020. Four selected species (Mytilus edulis, Corophium spp., Arenicola marina and Litorina littorea) were sampled according to their prevalent habitat at six investigation sites along the coastline of Lower Saxony (see. Fig. 1). A total number of >3,200 individuals were taken and pooled from three (Arenicola marina) to twenty (Corophium spp.). Fish samples were taken in parallel with the regular contaminant monitoring within the region, the gastrointestinal tract was dissected and all samples were stored frozen (-18°C) in aluminium foil until further treatment.

### Sample treatment

After dissection and recording of basic parameters such as dimensions and weight (Fig. 2), samples were digested in a solution consisting of potassium hydroxide (KOH) and sodium hypochlorite (NaClO) (300 ml KOH 10M + 150 ml NaClO 6-14 % filled to 1000 ml with MilliQ water) and left to stand for 48 h under a fumehood. Samples were transferred onto filters (613, VWR International 5-13 µm retention), stained with 1 ml nile red solution (1 mg nile red/ml chloroform) and left to dry.

### QA/QC management

In order to reduce background contamination as much as possible, precautions have been taken, such as the use of glass and stainless steel materials, the filtration of all chemical solutions applied and the integration of procedural blanks.



Fig. 2: Recording of basic parameters (dimensions and weight) – Mytilus edulis, Arenicola marina, Corophium spp., Littorina littorea, Plattichthys flexus

### Analysis of synthetic polymers

Synthetic polymers were detected via nile red staining and fluorescent microscopy (AxioScope7 – FTIC filter, Zeiss). All particles larger 20  $\mu$ m were recorded, measured and classified according to their shape (fragments and fibers). A subset of identified particles are analyzed for polymer composition implementing  $\mu$ Raman spectroscopy (DXRxi2, Thermofisher Scientific) (Fig. 3).



Fig. 3: Detection of synthetic particles (Nile red staining/ fluorescent microscopy) and polymer composition via μRaman spectroscopy

### **Preliminary results**

Preliminary results on synthetic fragments from a subset of all species (except for Littorina littorea) and individuals covering 143 samples representing 850 individuals are shown in Fig. 4 and 5.

Results on concentrations show that all species investigated are affected by microplastic pollution. Fragments are clearly dominating (>90 %) compared to fibers within all samples. Thus, within the preliminary results only fragments are considered.

Highest abundance of microplastic particles per individual according to invertebrate species are found in Arenicola marina with median concentrations of 15.4 fragments per individual (mean 22.2) followed by Mytilus edulis (median 17.1 median 18.7) and Corophium spp. (median 0.70, mean 0.75). The low abundance in Corophium above all results in the small size and weight of the species. Fig. 4B shows concentrations per fresh weight where relations between species differ accordingly. Considering different investigation sites the preliminary results clearly reflect the tendency of highest values at the site Neuharlingersiel being affected by ferry and shipping activities. Results on the fish species Platichthys flesus overall range from 3.0 to 63.0 particles per individual (median 18.0, mean 21.9) and show lowest concentration at the site P1\_Baltrum and highest at P4\_Jadebusen (Fig. SA and B). Please note that results are not yet corrected for blank values ranging from 0.0 to 5.0 particles per individual (except for Coropohium ranging from 0.0 to 0.7 particles per individual).



Fig. 5: Microplastic concentrations in Platichthys flesus according to investigation sites (A – per individual, B – per g tissue weight)

### **Conclusions / Outlook**

The preliminary results already provide a good overview but needs to be backed with a larger data set that will be generated in the following months. Furthermore, statistics concerning correlation to influencing factors such as individual characteristics, spatial gradients and local factors will be calculated and data will be compared with other findings from the North Sea. Additionally, sediment cores are investigated and the specific correlation of sediment-species interaction will be investigated focussing on Arenicola marina.

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### **Occurrence of microplastics in the gastrointestinal tracts of** *Chelon saliens* **along the Turkish coast**

Gundogdu Sedat, Çevik Cem, Temiz Ataş Nihan.

The manufacturing of plastics has increased rapidly since 1950, with annual plastic production reaching 359 million tons in 2018. It is estimated that 4.8–12.7 million tons of plastic end up in marine ecosystems every year. A total of 611 studies conducted at 4358 locations across the globe, as of September 2019, have revealed that 76.9% of marine litter consists of plastics. It is further estimated that 92% of marine plastic pollution is in the form of microplastics (MPs). Generally, plastic particles that are smaller than 5 mm are considered MPs. This high quantity of MPs in the sea poses a significant threat to marine life. In this study, the presence of microplastics (MPs) in the stomachs and digestive tracts of 62 individuals of leaping mullet (Chelon saliens (Risso, 1810)), collected along the Marmara, Aegean, and Mediterranean coasts of Turkey was examined microscopically and through µ-Raman analysis. A total of 159 MP particles were extracted. Among the examined fishes, the average MP concentration was 2.5 MP per fish (MPs fish–1). The size of the MPs ranged from 0.028 to 4.89 mm. To determine the polymer types of the MPs, a µ-Raman analysis was conducted. The most frequently detected polymers were polyethylene (41.67%), polyethylene terephthalate/polyester (12.5%) and polypropylene (8.3%). The results of this study showed that MP pollution represents an emerging threat to the fish of Turkish marine waters. This study was supported by Greenpeace Mediterranean as part of the "Türkiye'deki deniz canlılarında mikroplastik kirliliği: Plastikten kurtul oltaya gelme" project.

Keywords : Chelon saliens , Microplastics , plastic pollution , Turkish marine waters

# OCCURRENCE OF MICROPLASTICS IN THE GASTROINTESTINAL TRACTS OF Chelon saliens ALONG THE TURKISH COAST

Sedat Gündoğdu<sup>1\*</sup>, Cem Çevik<sup>1</sup> Nihan Temiz Ataş<sup>2</sup> <sup>1</sup>Cukurova University, Faculty of Fisheries, Department of Basic Sciences, <sup>2</sup> Greenpeace Mediterranean – Istanbul, Turkey Microplastic Researcg Group









# **Results and Discussion**

examined microscopically and through µ-Raman analysis.

along the Marmara, Aegean, and Mediterranean coasts of Turkey was 62 individuals of leaping mullet (Chelon saliens (Risso, 1810)), collected presence of microplastics (MPs) in the stomachs and digestive tracts of in the sea poses a significant threat to marine life. In this study, the

of the MPs ranged from 0.028 to 4.89 mm. To determine the polymer this study showed that MP pollution represents an emerging threat to terephthalate/polyester (12.5%) and polypropylene (8.3%). The results of frequently detected polymers were polyethylene (41.67%), polyethylene types of the MPs, a µ-Raman analysis was conducted. The most the average MP concentration was 2.5 MP per fish (MPs fish-1). The size A total of 159 MP particles were extracted. Among the examined fishes,

the fish of Turkish marine waters.

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### Session 24.8\_Ma. Chaired by Guillaume Duflos, Boulogne sur mer

Exposure of microplastic at levels relevant for human health: cytotoxicity and cellular localization of polystyrene microparticles in four human cell lines

Peng Miao, Grootaert Charlotte, Rajkovic Andreja, Janssen Colin, Asselman Jana. Paper number 334460

### Effect of polyester microfibres released from a domestic dryer machine on human lung organoids and *Xenopus laevis*

Winkler Anna, Santo Nadia, Madaschi Laura, Cherubini Alessandro, Rusconi Francesco, Rosso Lorenzo, Tremolada Paolo, Lazzari Lorenza, Bacchetta Renato. Paper number 334495

### **Inorganic-doped nano and micro plastics as model materials for multifunctional detection** *La Spina Rita, Cassano Domenico, Facchetti Samantha, Ponti Jessica, Fumagalli Francesco Sirio, Ricciardi Nicoletta, Gilliland Douglas.* Paper number 334493

### Exposure of microplastic at levels relevant for human health: cytotoxicity and cellular localization of polystyrene microparticles in four human cell lines

Peng Miao, Grootaert Charlotte, Rajkovic Andreja, Janssen Colin, Asselman Jana.

Microplastics (MPs), which are ubiquitous in our living environment, can enter into human body via diverse pathways such as food packaging, contaminated food and bottled mineral water. Therefore, it is essential to assess the risk of MPs daily human intake. Up to date, almost all of related publications used concentrations that are much higher than likely present in these sources. Thus, investigation at levels of MPs relevant for human health exposure can help us rationally understand the threats of MPs. This study is aimed to evaluate cytotoxicity and quantify the cellular uptake and localization of MPs within the concentration range reported in bottled mineral water in human cell lines. To this aim, four types of human cell lines derived from colon (Caco-2), liver (HepG2) and lung (A549 and BEAS-2B) were exposed to 2-µm fluorescent PS microspheres (1E+3-1E+7 particles/L). A series of cellular and biochemical assays (intracellular reactive oxygen species, mitochondrial membrane potential, sulforhodamine B and MTT assay) were conducted. To confirm the cellular uptake, the fluorescent cells containing PS were counted by flow cytometry to evaluate the probability of cells embedded PS under different concentrations. Furthermore, laser confocal scanning microscopy was used to observe the distribution and count the number of PS microspheres in four cell lines.

Keywords : cellular intake , cytotoxicity , human cell lines , human health , microplastics

# Exposure of microplastics at levels relevant for human health: cytotoxicity and cellular localization of

# polystyrene microparticles in four human cell lines

Miao Peng<sup>1,2</sup>

Charlotte Grootaert<sup>3</sup>

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Colin Janssen<sup>1,2</sup>

Jana Asselman<sup>2</sup>









of microplastics (MPs) on human health? understand the threats How to rationally



Environ. Sci. Technol. 2019, 53, 1748–1765

Microplas Main diar <i>Water Re</i>	
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able 1 Four cell lines exp	osed to 2-µm fluo	rescent PS at levels re	elevant for human health	$10^{3}-10^{7}$ particles
Acute cellular assays Human cell lines	Mitochondrial membrane potential	Reactive oxygen species	Sulforhodamine B assay	MTT assay
A549	No effect	Adverse effect	No effect	No effect
BEAS-2B	No effect	No effect	No effect	No effect
Caco-2	No effect	Adverse effect	No effect	No effect
HepG2	No effect	No effect	No effect	No effect





adverse ettects cause certain cell lines and translocate into relevant for MPs at levels uman health can

<sup>3</sup>Laboratory of Food Microbiology and Food Preservation, Faculty of Bioscience Engineering, Ghent University, 9000 Ghent, Belgium <sup>2</sup>Blue Growth Research Lab, Ghent University, Greenbridge, Wetenschapspark 1, 8400 Ostend, Belgium <sup>1</sup>Laboratory of Environmental Toxicology and Aquatic Ecology, Faculty of Bioscience Engineering, Ghent University, 9000 Ghent, Belgium microscopy.

flow cytometry.

exposed to fluorescent MPs taken with confocal Fig. 2 Three-dimensional images of human cells

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### Effect of polyester microfibres released from a domestic dryer machine on human lung organoids and *Xenopus laevis*

Winkler Anna, Santo Nadia, Madaschi Laura, Cherubini Alessandro, Rusconi Francesco, Rosso Lorenzo, Tremolada Paolo, Lazzari Lorenza, Bacchetta Renato.

Washing synthetic clothes and textiles releases microplastics into the environment with the discharged water. However, microfibre emissions from dryer machines into air and waterways are still poorly investigated. This study aims to characterise and quantify the release of polyester fibres from a domestic dryer machine and to analyse their effects on two test models representing potential targets of freshwater and airborne contamination; the animal model Xenopus laevis (early development stage of a frog) and an innovative 3D model consisting of human lung organoids. Organoids are the most advanced in vitro models but are not vet applied to evaluate the biological effect associated with microplastic exposure. For this study, a variety of polyester clothes and tissues was washed and dried. Characterisation of morphology and hydrodynamic behaviour of released fibres into the air filter (0.46 g/kg dry weight) and into discharged water was performed by optical microscopy, SEM-EDS and DLS. The models were exposed to the environmentally relevant polyester fibres in various concentrations (1, 10 and 50 mg/L) and incorporation phenomena and effects were analysed by SEM and confocal microscopy. Preliminary results show ingestion of large amount of fibres by the animal model and an unexpected interaction between lung organoids and microfibers. More information on the effects will be presented at the conference. In addition to the implications of the results, the applicability of the used human organoids as test models for microplastic exposure will be discussed.

Keywords : Confocal microscopy , Dryer , Human health , Microplastic fibres , Organoids , SEM

ONDAZIONE IRCCS CA' GRANDA Defedate Maggiore Policlinico

Regione



### Effects of polyester microfibers released from a domestic dryer machine on human lung organoids and Xenopus laevis

Anna Winkler<sup>a</sup>, Nadia Santo<sup>b</sup>, Laura Madaschi<sup>b</sup>, Alessandro Cherubini<sup>c</sup>, Francesco Rusconi<sup>c</sup>, Lorenzo Rosso <sup>d</sup>, Paolo Tremolada <sup>a</sup>, Lorenza Lazzari \*<sup>c</sup>, Renato Bacchetta \*



Microplastic fiber (MPF) emissions from dryer machines into air and waterways are poorly investigated. Thus, the objective was to characterise and quantify the release of polyester fibres from a domestic dryer machine and to analyse their effects on two test models representing potential targets of airborne and freshwater contamination; in-vitro human lung organoids and the in-vivo animal model Xenopus laevis. Since human organoids were not yet applied to evaluate the biological effect associated with MP/NP exposure, this study seeks to evaluate their applicability and suitability in microplastic exposure tests.



animal model Frog Embryo Teratogenesis Assay-Xenopus,

The drying process emitted a total weight of 0.4 g/kg dry weight of MPFs into the exhaust air (1) and 1.24 mg/kg dry weight of MPFs

> Interestingly, the shape of these MPFs differed at the transversal

with a minimum height of 1-3  $\mu$ m along the thinnest dimension.

surface; exhibiting a varying profile from flat and twisted to tattered

The hydrodynamic diameter ranged from 100 nm to 340 nm

(referring to the diameter of a spherical particle of the same

FFTAX

MPF release of a dryer machine

volume as the detected MPF).

Mean Fibre length was 663  $\pm$  333  $\mu$ m.

into the wastewater (2).

### Human organoids

- Self-organizing 3D structure grown from stem cells Recapitulate essential aspects of the *in-vivo* organ structure and function while being more

experimentally tractable than model organisms <sup>1</sup>





(2)

**RESULTS** (preliminary)

Confocal microscopy

Human lung organoids; adult stem cell derived, obtained from healthy human lung tissues

> Optical microscopy and SEM analysis. Analysis in progress.

### Immunofluorescence staining

Scanning Electron Microscopy (SEM) was also

### Cultivation of lung organoids



(a+b) Confocal microscopy images of the 3D structure of immunolabelled lung organoid (control sample) and surface section of lung organoid showing markers for cilia (green, Alexa Fluor 488) and nucleus (blue, Hoechst 33342). Counterstained is the actin cytoskeleton (red, F-actin). > Mature organoids are 400-600 μm in diameter.

(c) SEM image of organoid surface demonstrating cell diversity.



Confocal microscope images and counter phase contrast (b/w) (left) and SEM images (lower right) of lung organoids after exposure to MPFs from a dryer ntrations. MPFs are indicated with a white ar e at diffe

### **CONCLUSION AND OUTLOOK**

- Lung organoids were affected by MPFs at all concentration steps.
  - deformation of their 3D structure, such as openings (a) and teared off cells (c).
  - Interestingly, we observed an internalisation of the MPFs in organoids at the second and third exposure concentration (e-g). While some MPFs seem to be embedded in the surface of organoids (h+i), we can also observe a full internalisation of MPF sections (g and its magnification).
  - Image g gives the impression that a MPF entered the organoid by pushing through the surface and exiting it again, while pulling along cell tissues creating a dip at the entrance and a bulge at the exit. This effect might be resulted by the dynamical processes of the MPFs being suspended in the solution and are free to move around.



Xenopus exposed to MPFs

**O** No mortality **O** No growth inhibition ⊘ Malformation: No difference between Xenopus of CTRL and MPF-I, while Xenopus of MPF-II and MPF-III exhibit intestinal malformations, probably due to the presence of the fibres in the intestine that prevent normal intestinal development.

- The drying of synthetic clothes releases MPFs and contributes thus to the contamination of MP/NP in our environment. We were able to quantify MPF release into the wastewater. MPFs in the exhaust air were filtered by the dryer, therefore, the next step is quantifying the MPF release into the ambient air, as studied only once before <sup>2</sup>.
- We cultivated human lung organoids and were able to display their cytoarchitectural organization by immunofluorescence staining. Organoids (3D model) have the potential to replace animal models and primary human tissues (cell lines, 2D models) in MP/NP research to fully demonstrate human Þ physiological responses. However, the lung organoid model requires further advancement in the heterogeneity of its cellular composition in order to better mimic the lung
- tissue physiological functions. Other shortcomings remain; their accessibility and higher costs than conventional 2D models. To detect inflammatory cytokine in organoids (triggered eventually by tissue damage) and to further validate the pseudostratified airway composition of organoids (basal cells, functional multi-ciliated cells, mucus-producing secretory cells, and club cells), gene expression analysis by qRT-PCR will be performed to complete this study.

### Further tests on lung organoids will be performed with different shaped and sized synthetic polymers (i.e. nanospheres).

### AFFILIATIONS

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Organoids exposed to MPFs Organoids exposed to 1 mg/L MPFs exhibited

### Inorganic-doped nano and micro plastics as model materials for multifunctional detection

La Spina Rita, Cassano Domenico, Facchetti Samantha, Ponti Jessica, Fumagalli Francesco Sirio, Ricciardi Nicoletta, Gilliland Douglas.

The large amount of plastic produced, and therefore its degradation into nano and micro plastics and its accumulation in water, soil and air has opened a debate on possible harmful effects on the environment and humans. To date, both representative materials and effective methods for the extraction, identification and quantification of nano and micro plastics from complex matrices are still lacking. In this study, we present the synthesis of polypropylene (PP) nanoparticles (NPs), being PP amongst the most commonly detected plastic debris throughout the environment. In addition, the traceability of the PP has been achieved by introducing selected markers into PP NPs such as inorganic and/or fluorescent dyes. A similar approach has been used for Polyvinyl chloride (PVC) microplastics, which is the third most widely produced synthetic polymer (ca. 40 million tons/year) and widespread for its various applications. The synthesis of fluorescent and platinum-labelled PVC microparticles was optimized and the as produced microplastics were administered to freshwater mussels. The latter are widely used in absorption and bioaccumulation studies of microplastics.

Keywords : micro and nano plastics , mussels , synthesis



European Commission

### Inorganic-doped nano and micro plastics as model materials for multifunctional detection (abstract n° 334493).

Rita La Spina<sup>a</sup>, Domenico Cassano<sup>a</sup>, Samantha Facchetti<sup>a</sup>, Jessica Ponti<sup>a</sup>, Francesco Fumagalli<sup>a</sup>, Nicoletta Riccardi<sup>b</sup> and Douglas Gilliland<sup>a</sup> <sup>a</sup>European Commission, Joint Research Centre (JRC), Ispra (VA), Italy <sup>b</sup>Consiglio Nazionale delle Ricerche (CNR), Institute of Ecosystem Study, Pallanza (VB), Italy

ABSTRACT: The large amount of plastic produced, and therefore its degradation into nano and micro plastics and its accumulation in water, soil and air has opened a debate on possible harmful effects on the environment and humans. To date, both representative materials and effective methods for the extraction, identification and quantification of nano and micro plastics from complex matrices are still lacking. In this study, we present the synthesis of polypropylene (PP) nanoparticles (NPs), being PP amongst the most commonly detected plastic debris throughout the environment. In addition, the traceability of the PP has been achieved by introducing selected markers into PP NPs such as inorganic and/or fluorescent dyes. A similar approach has been used for Polyvinyl chloride (PVC) microplastics, which is the third most widely produced synthetic polymer (ca. 40 million tons/ year) and widespread for its various applications. The synthesis of fluorescent and platinum-labelled PVC microparticles was optimized and the as produced microplastics were administered to freshwater mussels. The latter are widely used in absorption and bioaccumulation studies of microplastics.





gradient. Scale bar 200 um

Conclusions: The synthesis of doped PP nanoplastics and doped PVC microplastics have been demonstrated. The traceability of the plastics have been achieved by introducing selective markers which enable the detection using spectroscopic techniques but also following the presence of metal. The detection of PVC doped microplastics have been demostrated after exposure to mussels.

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PVC-PtOEP MPs ingested by mussels and extracted with sucrose-ZnCl<sub>2</sub> density

### Session 24.9\_O. Chaired by Manuel Monge-Ganuzas, Urdaibai

### Monitoring of meso and microplastic debris in Playa Grande beach (Tenerife, Canary Islands, Spain) during a moon cycle

González-Hernández María, Hernández-Sánchez Cintia, González-Sálamo Javier, López-Darias Jessica, Hernández Borges Javier. Paper number 333774

### Microplastic debris in six beaches of Tenerife (Canary Islands, Spain)

Álvarez-Hernández Clara, Cairós Carlos, López-Darias Jessica, Mazzetti Elisa, Hernández-Sánchez Cintia, González-Sálamo Javier, Hernández-Borges Javier. Paper number 333795

### **Determination of microfibres in marine sediments of La Palma Island (Canary Islands, Spain)** *Villanova Solano Cristina, Díaz Peña J. Francisco, Hernández-Sánchez Cintia, González-Sálamo Javier, Hernández-Borges Javier.* Paper number 333885

### Monitoring of meso and microplastic debris in Playa Grande beach (Tenerife, Canary Islands, Spain) during a moon cycle

González-Hernández María, Hernández-Sánchez Cintia, González-Sálamo Javier, López-Darias Jessica, Hernández Borges Javier.

Microplastics abundance in the coastal zones varies not only over space but also over time. Concerning the periodicity of the sampling, many studies frequently develop a single sampling while others develop a long-term evaluation, in many cases once a month. In order to accomplish this, it is important to know how the abundance of microplastics varies on small temporal scales; of particular interest is also the monitoring during a moon cycle, in order to study the possible influence of the tides on microplastics arrival. To the best of our knowledge, microplastics monitoring during a complete moon cycle has only been developed in very few occasions [1,2]. In this work, the occurrence and composition of meso- (5-25 mm) and microplastics (1-5 mm) in Playa Grande beach was monitored during a complete moon cycle on the different moon phases between 17th June and 16th July 2019 [3]. A total of 10 points were sampled each day finding an average content of mesoplastics of 18 g/m2 (0.36 g/L) and of microplastics of 78 g/m2 (1277 items/m2 or 1.6 g/L). Polypropylene and polyethylene were the most abundant types of plastics. Among the analysed particles, fragments accounted for 83 % of the total. The obtained results revealed that microplastics presence could not be related in this case with the tides but with the orientation and strength of the wind. References 1 A. R. A. Lima, M. Barletta, M. F. Costa, J. A. A. Ramos, D. V Dantas, P. A. M. C. Melo, A. K. S. Justino and G. V. B. Ferreira, J. Fish Biol., 2016, 89, 619–640. 2 J. A. A. Ramos and W. V. N. Pessoa, Mar. Pollut. Bull., 2019, 142, 428-432. 3 M. González-Hernández, C. Hernández-Sánchez, J. González-Sálamo, J. López-Darias, J. Hernández-Borges, Mar. Pollut. Bull., 2020, 150, 110757.

Keywords : beaches , Canary Islands , microplastics , Moon cycle

### MONITORING OF MESO AND MICROPLASTIC DEBRIS IN PLAYA GRANDE BEACH (TENERIFE, CANARY ISLANDS, SPAIN) DURING A MOON CYCLE

**Universidad** de La Laguna

María González-Hernández<sup>1</sup>, Cintia Hernández-Sánchez<sup>2,3</sup>, Javier González-Sálamo<sup>1,3</sup>, Jessica López-Darias<sup>1</sup>, and Javier Hernández-Borges

idad de La Laguna (ULL). Avda. Astrofísico Fco. Sánchez, s/nº. 38206 5 ogia, Medicina Forense y Legal y Parasitologia, Área de Medicina Prev Inversidad de La Laguna (ULL), Via Auxillar Paso Atto, nº, 2 3001 Sant o La Laguna (ULL), Avda. Astrofísico Fco. Sánchez, s/nº. 38206 San Cri ias, Uni<sup>.</sup> lica, To:





Microplastics abundance in the coastal zones varies not only over space but also over time. As a result, the sampling period and the spatial distribution are of special concern. Concerning the periodicity of the sampling, many studies frequently develop a single sampling while others develop a long-term evaluation, in many cases once a month. In order to accomplish this, it is important to know how the abundance of microplastics varies on small temporal scales, which can also differ from one place to another, as a result of the local variation in hydrological processes; of particular interest is also the monitoring during a moon cycle, in order to study the possible influence of the tides on microplastics arrival. To the best of our knowledge, microplastics monitoring during a complete moon cycle has only been developed in two occasions<sup>1,2</sup>, in the first case in different parts of an estuary and, in the second, in different parts of a sandy beach (water sampling)

In this work, the occurrence and composition of meso- (5-25 mm) and microolastics (1-5 mm) in Plava Grande beach (Tenerife, Canary Islands, Spain) was monitored during a complete moon cycle on the different moon phases between 17th June and 16<sup>th</sup> July 2019. A total of 10 points were sampled each day finding an average content of mesoplastics of 18 g/m<sup>2</sup> (0.36 g/L) and of microplastics of 78 g/m<sup>2</sup> (1277 items/m<sup>2</sup> or 1.6 g/L). Polypropylene (PP) and polyethylene (PE) were the most abundant types of plastics. Among the analysed particles, fragments accounted for 83 % of the total. The obtained results revealed that microplastics presence could not be related in this case with the tides but with the orientation and strength of the wind



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CajaCanarias

### A. R. A. Lima, M. Barletta, M. F. Costa, J. A. A. Ramos, D. V Dantas, P. A. M. C. Melo, A. K. S. Justino and G. V. B. Ferreira, J. Fish Biol., 2016, 89, 619-640 J. A. A. Ramos and W. V. N. Pessoa, Mar. Pollut. Bull., 2019, 142, 428-432

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### Microplastic debris in six beaches of Tenerife (Canary Islands, Spain)

Álvarez-Hernández Clara, Cairós Carlos, López-Darias Jessica, Mazzetti Elisa, Hernández-Sánchez Cintia, González-Sálamo Javier, Hernández-Borges Javier.

Relative recent evidence has shown that the southward flowing Canary Current is leaving on the Canary Islands important amounts of marine debris, specially plastic, from the open North Atlantic Ocean, mainly on North and Northeast exposed beaches [1-3]. However, up to the development of this study by the end of 2018, no works had focused on the study of the presence of microplastics in beaches of the occidental islands (Tenerife, La Palma, La Gomera and El Hierro), which is highly necessary in order to fully evaluate their content, distribution and also the possible detection of "hot spots" or "black points" with an extremely high content of microplastics. In this work, the occurrence and composition of microplastics (1-5 mm) was evaluated in six beaches of the island of Tenerife for the first time [4]. Two of them were located at the North coast (El Socorro and San Marcos) and the rest in the South littoral (Leocadio Machado, El Porís, Los Abriguitos and Plava Grande). Sampling was developed during the months of October, November and December 2018 (depending on the beach). Isolated microplastics were identified by attenuated total reflection Fourier transform infrared spectroscopy. All beaches showed a relatively low content of microplastics, except Playa Grande, which showed an average content of 98.97 g/m2. The major polymers found were polyethylene, polypropylene and polystyrene, accounting for 69 %, 18 % and 4 %, respectively. References [1] A. Herrera et al., Mar. Pollut. Bull., 2018, 129, 494–502. [2] C. Edo et al., Mar. Pollut. Bull., 2019, 143, 220–227. [3] J. Baztan et al., Mar. Pollut. Bull., 2014, 80, 302–311. [4] C. Álvarez-Hernández et al., Mar. Pollut. Bull., 2019, 146, 26–32.

Keywords : beaches , Canary Islands , Microplastics



### MICROPLASTIC DEBRIS IN SIX BEACHES OF TENERIFE (CANARY ISLANDS, SPAIN)

Clara Álvarez-Hernández<sup>1</sup>, Carlos Cairós<sup>1</sup>, Jessica López-Darias<sup>1,2</sup>, Elisa Mazzetti<sup>1</sup>, Cintia Hernández-Sánchez<sup>3,4</sup>, Javier González-Sálamo<sup>1,4</sup>, and Javier Hernández-Borges<sup>1,4,\*</sup>

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### INTRODUCTION

Relative recent evidence has shown that the southward flowing Canary Current is leaving on the Canary Islands important amounts of marine debris, especially plastic, from the open North Atlantic Ocean, mainly on North and Northeast exposed beaches<sup>1-3</sup>. However, up to the development of this study by the end of 2018, no works had focused on the study of the presence of microplastics in beaches of the occidental islands (Tenerife, La Palma, La Gomera and El Hierro), which is highly necessary in order to fully evaluate their content, distribution and also the possible detection of "hot spots" or "black points" with an extremely high content of microplastics

In this work, the occurrence and composition of microplastics (1-5 mm) was evaluated in six beaches of the island of Tenerife for the first time<sup>4</sup>. Two of them were located at the North coast (El Socorro and San Marcos) and the rest in the South littoral (Leocadio Machado, El Porís, Los Abriguitos and Playa Grande). Sampling was developed during the months of October, November and December 2018 (depending on the beach). Isolated microplastics were identified by attenuated total reflection Fourier transform infrared spectroscopy. All beaches showed a relatively low content of microplastics, except Plava Grande, which showed an average content of 98.97 g/m<sup>2</sup>. The major polymers found were polyethylene (PE), polypropylene (PP) and polystyrene (PS), accounting for 69 %, 18 % and 4 %, respectively

### **EXPERIMENTAL**



### **RESULTS AND DISCUSSION**





CONCLUSIONS

- > The sampling of 6 beaches of the island of Tenerife revealed a low microplastic content (below 115.5 items/m<sup>2</sup>, 3.5 g/m<sup>2</sup> and 0.069 g/L) above the high tide line, except for Playa Grande beach, located on the South of the islands and with a NE orientation, which is not periodically cleaned, and which suffers from strong microplastic debris episodes as a result of the Canary Current.
- > Microplastic content of Playa Grande (2971.5 items/m<sup>2</sup>, 99 g/m<sup>2</sup> and 2.0 g/L) is comparable, or even higher, to those previously found in some beaches of the oriental islands of the archipelago and clearly suggests the need of developing periodical monitoring programs for further studies.

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> Most microplastics obtained were fragments of PP, PS and PE (ATR-IR was used for microplastic characterization) which is also in accordance with global plastic production.

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(%) 25 percentage 20

15 Weight 1

1-2

2-4

Particle size (mm)

4-5

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J.L.D. and J.G.S. would like to thank "Cabildo de Tenerife" for the Agustín de Betancourt contract at the Universidad de La Laguna (ULL). C.C. would like to Las would me to thank caunua de interner of the Agosant led Estantiant Contrast at the Oniversional de la Lagonal (ULE). Co support research family algone) of the Conomy, industry, Tade and Knowledge (ACIB) of the Canary Islands Government for the contrast to support research he international Research Campus (CEI) of the Universidad de La Laguna (ULL). Authors thank the Research Support General Service (SGA) of the International Research Campus (CEI) of the Universidad de La Laguna (ULL). Authors thank the Research Support General Service (SGA) of the International Research Campus (CEI) of the Universidad de La Laguna (ULL). Authors thank the Research Support General Service (SGA) of the International Research Campus (CEI) of the Universidad de La Laguna (ULL). Authors thank the Research Support Security (SGA) of the Campus (SGA) o the Canary Agency of Econo activities of the Inte the Uni

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### **Determination of microfibres in marine sediments of La Palma Island (Canary Islands, Spain)**

Villanova Solano Cristina, Díaz Peña J. Francisco, Hernández-Sánchez Cintia, González-Sálamo Javier, Hernández-Borges Javier.

Microplastics are widely recognised as contaminants of emerging concern. In the marine environment, seabed sediments have been identified as a major sink with a high potential to accumulate them [1]. Among microplastic forms, microfibres appear among the most frequently reported in the environmental compartment [2]. The aim of this study was to quantify plastic microfibres in marine sediments of La Palma island (Canary Islands, Spain). Sediment samples were collected during July 2020 at four sampling locations with uniform sandy seabed and a water depth between 5 and 10 m. At each sampling point, three samples were taken parallel to the coast and separated 10 m from each other. Stainless steel cores, 10 cm depth, were used for sample collection, and microfibres content was evaluated at the laboratory every 2.5 cm (total samples = 48). First, each sediment sample (10 g) was digested with 33 % H2O2 in order to remove the organic matter. After that, several sequential flotations developed by the addition of a saturated solution of sodium chloride were carried out, and each supernatant was filtered through a 50 µm stainless steel filter. Filters were covered to prevent airborne contamination and stored for subsequent visual sorting using a binocular light microscope. Results revealed that all assessed sediment samples contained microfibers. Isolated microfibres were mainly translucent and, to a lesser extent, coloured (e.g. blue, red and black). This study has confirmed the widespread distribution of microplastics in sediments from an oceanic island, providing the first report on microplastics in marine sediments of the Canary Islands. References 1 T. Martellini, C. Guerranti, C. Scopetani, A. Ugolini, D. Chelazzi and A. Cincinelli, TrAC Trends Anal. Chem., 2018, 109, 173–179. 2 S. L. Wright, R. C. Thompson and T. S. Galloway, Environ. Pollut., 2013, 178, 483–492.

Keywords : Canary Islands , marine sediments , microfibres , Microplastics

### DETERMINATION OF MICROFIBERS IN MARINE SEDIMENTS OF LA PALMA ISLAND (CANARY ISLANDS, SPAIN) Universidad

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### INTRODUCTION

Microplastics are widely recognised as contaminants of emerging concern. In the marine environment, seabed sediments have been identified as a major sink with a high potential to accumulate them<sup>1</sup>. Among microplastic forms, microfibers appear among the most frequently reported in the environmental compartment<sup>2</sup>

de La Laguna

The aim of this study was to quantify plastic microfibers in marine sediments of La Palma island (Canary Islands, Spain). Sediment samples were collected during July 2020 at four sampling locations with uniform sandy seabed and a water depth between 5 and 10 m. At each sampling point, three samples were taken parallel to the coast and separated 10 m from each other. Stainless steel cores, 10 cm depth, were used for sample collection, and microfibers content was evaluated at the laboratory every 2.5 cm. First, each sediment sample (10 g) was digested with 33 % H<sub>2</sub>O<sub>2</sub> in order to remove the organic matter. After that, several sequential flotations developed by the addition of a saturated solution of sodium chloride were carried out, and each supernatant was filtered through a 50 µm stainless steel filter. Filtrates were visualized under a binocular light microscope. Laboratory blanks were also analyzed with every batch of samples in order to check that no laboratory contamination took place. Results revealed that all assessed sediment samples contained microfibers which were mainly translucent and, to a lesser extent, colored (e.g. blue, red and black).

This first study of Puerto Naos and Tazacorte sampling points has confirmed the widespread distribution of microplastics in sediments from an oceanic island, providing the first report on microplastics in marine sediments of the Canary Islands.

Fig. 1. Sampling points of MICROSED project in La Palm

### **EXPERIMENTAL**



### **RESULTS AND DISCUSSION**



### CONCLUSIONS

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- > In this work, all assessed sediment samples contained microfibers, mainly translucent fibers (88 %), being the rest blue, red, black and green. Blue and red fragments (4 in total) were also found in some of the samples.
- > The vast majority of the fibers had a length between 1 and 2 mm which did not vary as a function of depth in both Puerto Naos and Tazacorte sampling points. Besides, no statistical differences were observed in the length of the fibers between cores in both sampling locations.
- > The number of microfibers found per 10 grams of sediment seems to increase with depth in some cores (i.e. Core 1 Puerto Naos y Core 2 Tazacorte) but this issue cannot be generalized
- > This study has confirmed the presence of microplastics in sediments of an oceanic island such as La Palma, providing the first report on microplastics in marine sediments of the Canary Islands.
- > The two remaining sampling points (i.e. Puerto Espíndola and Santa Cruz de La Palma) are currently being studied. The composition of the microfibers is also being analyzed by Raman spectroscopy

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CV.S. would like to thank "Fundación Canaria Diario de Avisos" for her contract. J.G.S. would like to thank "Cabildo de Tenerife" for the Agustin de Betancourt contract at the Universidad de La Laguna (ULI). Authors would also like to thank "Fundación Canaria Diario de Avisos" for financial support (MICROSED project) as well as "Oceanologico", especially Lias Schwerter, for her help during the sampling. FUNDACIÓN 

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### Session 24.9\_Ma. Chaired by Matthias Völkl, Bayreuth

### Microplastics: a possible risk for the fin whale (*Balaenoptera physalus*) resident population from Gulf of California, México?.

Goñi Godoy Gara, Panti Cristina, Baini Matteo, Galli Matteo, Fossi Cristina, Viloria Gómora Lorena, Urbán Ramírez Jorge. Paper number 333132

### A pilot study to determine the potential impacts of plastics on Aotearoa-New Zealand's marine environment

Pantos Olga, Audrezet Francois, Doake Fraser, Donaldson Lloyd, Dupont Pierre, Gaw Sally, Kingsbury Joanne, Weaver Louise, Lear Gavin, Northcott Grant, Pochon Xavier, Smith Dawn, Theobald Beatrix, Wallbank Jessica, Zaiko Anastasija, Maday Stefan. Paper number 334094

### Investigating microplastic pollution in seasonally stratified waters

Jones Nia, Neill Simon. Paper number 334111

### Microplastics: a possible risk for the fin whale (*Balaenoptera physalus*) resident population from Gulf of California, México?

Goñi Godoy Gara, Panti Cristina, Baini Matteo, Galli Matteo, Fossi Cristina, Viloria Gómora Lorena, Urbán Ramírez Jorge.

Interaction between marine litter and cetaceans has been documented in 47 species. Specifically, filter feeding animals such as whales are susceptible to plastic contamination due to the magnitude of prey intake, which increases the risk of interaction respecting to large accumulations of microplastics (MPs). The longevity of these mammals allows them to be considered sentinel species of ocean pollution, whose exposure to plastic particles will depend mainly on the feeding behavior of the species and the level of pollution in their feeding areas. In this study, we present for the first time, evidence of MPs presence in feces of fin whale resident population of the Gulf of California collected in two feeding areas of the species: La Paz Bay (n=2; 0.103 ± 0.045 items/g) and Northern Gulf (n=1; 0.066 items/g). These results were compared in terms of abundance and composition with particles found in the analysis of surface water samples of the same areas, whose concentrations was lower for the Northern Gulf than for La Paz Bay ( $0.014 \pm 0.006$  and  $0.021 \pm 0.002$  items.m-2, respectively). All the samples analyzed presented plastic particles, mainly fibers as the dominant item. FTIR-spectrometry analysis identified polyester as the majority polymer. The similarity in the composition of MPs between fecal samples of fin whale and surface water samples can be taken as an indication of the fin whale level exposure in different seasons and feeding areas concerning to MPs pollution that affects the region.

Keywords : feces , fin whale , Gulf of California , microplastics



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### A pilot study to determine the potential impacts of plastics on Aotearoa-New Zealand's marine environment

Pantos Olga, Audrezet Francois, Doake Fraser, Donaldson Lloyd, Dupont Pierre, Gaw Sally, Kingsbury Joanne, Weaver Louise, Lear Gavin, Northcott Grant, Pochon Xavier, Smith Dawn, Theobald Beatrix, Wallbank Jessica, Zaiko Anastasija, Maday Stefan.

Once in the ocean, plastics are rapidly colonised by complex communities. Due to the buoyant and resilient nature, ocean plastics pose a significant risk to ecosystems and fishery-based economies through their role in the translocation of invasive species and pathogens or changes in ecosystem function. Factors affecting the development and composition of these communities are still poorly understood, and there is currently no information on the biofilms that form on marine plastics in the southern hemisphere or their potential risks to the environment. This study aims to address this knowledge gap. To do this, two chemically and structurally distinct polymers, which are also common in marine plastic litter, nylon 6 and polyethylene, were deployed for 3-months in the Port of Lyttelton, Christchurch, New Zealand. Biofilm present after 2 weeks was dominated by diatoms and cyanobacteria. Metagenomic analysis showed that the plastisphere was distinct from the communities associated with glass control surfaces and the surrounding water. Polymer-specificity of the bacterial communities seen at 2-weeks was absent in subsequent time points, whereas fungal communities did not change over time. Although mechanical properties of the plastics did not change over time, physical modification to the surface of the plastics was observed, with the most pronounced change seen in nylon, with pitting conforming to the shape of microbial cells. This is the first study to examine the microbial communities associated with marine plastics in New Zealand waters, allowing the improvement of the understanding of the potential risk they pose. A year-long study is now underway to examine the fate of 5 plastic types, the plastisphere that develops, the presence of invasive species and potential pathogens, and plastic degrading microbes which may present bioremediation potential, as well as changes to the mechanical properties and inherent and acquired chemicals associated with the plastics.

Keywords : marine plastics , nylon , plastisphere , polyethylene , Southern Hemisphere



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### A pilot study to determine the potential impacts of plastics on Aotearoa-New Zealand's marine environment

### Introduction

Plastic of all sizes, from nano- to macro-, and of all different polymer types are found in our oceans around the world. They present a range of threats to the environment at all sizes, and may enter the food web at different trophic levels. Impacts from ingestion range from causing physical internal damage to causing a false sense of satiation, resulting in subsequent implications for animal health and fitness. Inherent or acquired chemicals associated with the plastics may also pose a significant risk to the biota at different trophic levels both following direct consumption of the plastics and from bioaccumulation and biomagnification through the food chain. They provide a new substrate for the development of biofilms, and their buoyant and resilient nature means they pose a threat to ecosystems through their role in the translocation of invasive species and pathogens in the oceans.



Figure 1. Plastics enter our oceans through both land-based and sea-based activities. The weathering of plastic through biological nical processes n to fragment into increasingly smaller pieces; microplastics and na

This is the first study to examine the effect of polymer type on the biofilm communities that form on marine plastics in Aotearoa-New Zealand (A-NZ) waters, improving our understanding of their potential impacts. Here we present the results of a 3-month exposure experiment, where the bacterial and fungal communities of 2 polymers were examined.

### **Objectives**

Identify whether polymer type influences the microbial

communities and higher order species that form the plastisphere. Determine changes to plastics during exposure to the marine environment which may influence their fate.



Figure 2. Experimental deployment site within Lyttelton Port of Christchurch, NZ (Lat;Long: 43.6075815; 172.723340). A) Steel structure attached to the pontoon holds vertical rods with experimental plastics in place, allowing movement up and down with the tide and continuous submersion. B) Changes in the tensile properties of the polymers over time were determined using standard shapes. C) Three vertical rods with experimental plastics and glass controls attached.

### Materials & Methods

A system made of marine grade stainless steel (316) was designed to allow the submersion of plastics in surface waters. Bespoke injection moulded plastics (nylon 6 (PA) and linear low-density polyethelyne (LLDPE)) with known additives were affixed to the structure which was attached to the side of a pontoon (Fig. 2) at the Lyttelton Port of Christchurch, Canterbury, A-NZ. Glass supported by stainless steel (316) frames were used as an inert control substrate. Triplicates of each substrate type were distributed evenly over three rods, and three depths (20, 40, 60 cm). Temperature/light loggers placed at each height.

Mechanical properties (modulus, max stress and elongation at

break) of the plastics were carried out (ISO 527-2).

- ⇒ Total genomic DNA extraction (Qiagen DNeasy PowerSoil) ⇒ 16S rRNA gene V3-V4 (341F/805R) and ITS2 rRNA gene (fITS7/ITS4) amplified  $\Rightarrow$  sequenced (MiSeq 2x300 paired end reads).
- Sequence processing: conducted with Dada2 using the Silva v128 database.
- Sequence analysis: conducted with phyloseq and vegan in R v3.5.1.

### **Results:**



Figure 3. A) Macroscale differences are evident between the upper and lower surface biofilms of plastic and glass at 3 months. B) Mechanical testing found no significant change (n = 5) in the stiffness (modulus) of the plastics over 3 months. C) Biofilm bacterial communities showed change over time; fungal communities did not. Ellipses = 95% Cl. D) Diatoms identified by SEM and 18S rRNA sequence analysis identified 7 genera (data not shown) predominated the biofilm matrices (Scale bars = 20 um)

Biofilms rapidly developed on all substrate types (Fig. 3A).

⇒ No light or temperature difference occurred between the 3 depths, and no depth effect on microbial community composition was observed.

⇒ Mechanical properties (Fig.3B) of both plastics did not change significantly over time.



Biofilm microbial communities were independent of those in the water column.

- Bacterial but NOT fungal biofilm communities changed over time (Fig. 3C).
- Material-specificity of bacterial communities at 14 days only.
- ➡ Alpha-diversity of both fungal and bacterial communities did not change with age or material.

Figure 4. Relative abundance of fungal and bacterial genera (top 4 and 5 genera, respectively) present within the biofilm communities at the 3 sampling times.

### **Discussion/Conclusions**

- Although the mechanical properties of PA and LLDPE did not change significantly over 80 days exposure in seawater, microscale changes may occur which may influence their interaction with microbial biofilms. Further SEM analysis is being carried out.
- ⇒ The substrate-type effect seen in bacterial communities at 14 days may be a result of differences in surface texture and colour of the polymers and glass. Disappearance by 44 days, could be due to the dense biofilm present, screening the influence of the substrate.
- ⇒ The nonspecific association of fungi suggests the absence of selective pressure from either the substrate or young biofilm. Loss of conformity at 80 days is unclear.
- ⇒ The absences of an anticipated polymer-type effect due to the hydrophobic/hydrophilic nature of LLDPE/PA, but may have occurred earlier in the biofilm community succession.

Acknowledgements: We are grateful to Lyttelton Port Company for granting continued support, access and permission to use their pontoon for deployment. We thank Evelyn Hollaus and Erin McGill (ESR) for technical assistance. Funding was provided by Ministry of Business, Innovation and Employment (MBIE) Endeavour Fund C03X1802.









Scion

### Investigating microplastic pollution in seasonally stratified waters

Jones Nia, Neill Simon.

Up to 236 thousand tonnes of microplastic are estimated to be in the ocean's surface water yet highresolution data sets from physical observations in shelf seas and oceans are uncommon due to logistics and cost. Macro-scale gyres are known accumulation zones of plastic, however, meso-scale, seasonal gyres are less discussed. In some cases, the proximity of these gyres to the coast as well as the timescales on which they form, could give an interesting insight into extent of microplastic contamination in a particular region as well as the wider hydrodynamics of the gyre itself. Data of microplastic contamination in much of the Irish Sea is vet to be published, the details of which could be of interest to the communities of the highly populated coastlines in the region. Details of an ambitious and novel field campaign through the Western Irish Sea Gyre aiming to resolve spatial variability of microplastic in surface, sub-surface and seabed sediments will be presented. Over 200 surface, sub-surface and seabed samples were collected aboard the Bangor University's School of Ocean Science's RV Prince Madog during July 2020 using trongo nets, niskin bottles and a box corer. Researchers collecting data identified possible microplastic particles in the surface water of the gyre however more detailed analysis is now needed. The samples will be analysed for microplastic extent, mass and composition using LD-IR and sediments dated using radioactive Pb210. Being able to understand and quantify current microplastic contamination within the Western Irish Sea Gyre will lay the foundations for research into microplastic contamination on a larger scale within the Irish Sea and provide much needed data for the validation of regional microplastic dispersal models.

Keywords : Bangor University , field , fieldwork , gyre , irish sea , marine science , methods , microplastic , seasonal stratification , uk , uk coast

### **INVESTIGATING MICROPLASTIC POLLUTION** · IN SEASONALLY STRATIFIED WATERS

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BACKGROUND



The retention of plastic waste in oceanic gyres, more commonly known as "garbage patches", is a phenomenon well-known to researchers and the general public alike (Figure 1). These gyres are permanent circulating currents collecting marine debris over decades (Lebreton et al., 2018).

In contrast, small, seasonal gyres operate on smaller spatiotemporal scales allowing for a higher resolution analysis of microplastic presence and distribution. A cyclonic near surface gyre in the Western Irish Sea is known to collect biological matter however its ability to retain microplastic and the general concentrations of microplastic in most of the Irish Sea is unknown. Due to secondary currents associated with the gyre we hypothesise that microplastic concentrations in the centre of the gyre will be greater than the peripheral concentrations.

54.4

54.2

54

53.6

53.4

53.2

atitud 53.8

This study aims to:

· determine the presence of microplastic in the Irish Sea

• quantify microplastic concentrations throughout the water column through sampling surface waters, sub surface waters and sea bed sediments.

### **STUDY SITE:** The Western Irish Sea Gyre

Large gyre systems are known to accumulate microplastic. however less is known about smaller, seasonal avres.

The Western Irish Sea Gyre forms following the onset of stratification where a cool, dense dome of water is trapped by a well mixed layer during spring and summer where the cooler water warms slowly relative to the well mixed surface layer (Figure 2). Abrupt changes from well mixed to stratified waters occurs at tidal mixing fronts where the frontal density gradients drive a baroclinic flow which is geostrophic to first approximation. This flow presents itself at the surface as a cyclonic circulation of water where the flow is parallel to the isopycnals. This residual circulation of has been known to retain biological matter (e.g larval and juvenile fish) (Robins et al., 2013).

A 40 km transect (northwest to southeast), consisting of 12 sample stations, was chosen chosen to represent the gyre at during July, 2020 (Figure 3).

### SAMPLING METHODS



Surface water was sampled with a 250 µm Trongo Net. being towed for 3 minutes at each of the 12 sample stations. The samples in the three nets were then transferred to three sample bottles using a 250 µm sieve and filtered seawater.

### SUBSURFACE **ATER**

seawater.

Subsurface waters were sampled at 10m depth across the 12 sample stations with a niskin bottle rosette capturing ~36 litres per station. Samples were concentrated with a 250 µm sieve and filtered

### EA BED SEDIMENTS stations Three were sampled for sea bed

sediments using the box corer to maintain sediment structure. Each sample was then subsampled on deck at 5, 10 and 15 cm depth respectively and transferred to individual glass containers.

### CONTAMINATION CONTROL

Contamination control was maintained throughout the cruise by restricting the number of scientists sampling

.....

3

Longitude

Direction of currents



0

(2), wearing cotton/well washed clothes to reduce fibre contamination and using glass and stainless steel equipment where possible.



Analysis: Possible microplastic particles as well as larger plastic litter were observed in the surface water of the gyre while out in the field, however more detailed analysis is now underway. The samples will be analysed for microplastic concentration and composition by using a Laser Direct Infrared (LDIR) which will be able to define the type of plastic as well as the amount and size of particles.

Following this, hydrodynamic models developed by the Met Office will be used in conjunction with particle tracking models to follow particles to see where/if the particles move en masse or if they disperse throughout the Irish Sea and on what timescales they do so.

Application: Being able to quanitfy contamination within the Western Irish Sea Gyre will lay the foundation for research into microplastic contamination on a larger shelf sea scale and provide insight into the hydrodynamics of the gyre. This data set will also be valuable in informing future models of how microplastic moves around the region, and how the specific sources and sinks impact nearby coastlines and beyond.

This study is a part of a wider PhD studentship investigating the dispersal of microplastic in the marine environment which aims to determine the spatiotemporal variability of microplastic and the pathways from estuaries to shelf seas.







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Figure 2: Schematic representation of a baroclinic gyre. Dotted circles denotes flow out of the page and crossed circles denotes flow into the page. Figure adapted from Horsburgh (1999).



### Session 24.10\_O. Chaired by Manuel Monge-Ganuzas, Urdaibai

**A "sediment transport" perspective on microplastics movement: incipient motion** *Varrani Arianna*. Paper number 334298

### Accelerated hydrolysis method for producing partially degraded polyester microplastic fiber reference materials

Sarno Antonio, Olafsen Kjell, Kubowicz Stephan, Sørensen Lisbet, Booth Andy M.. Paper number 334355

### Characterization of microplastics in sediments from the submarine outfall of the wastewater treatment plant, Mar del Plata City (Argentina)

*Diaz-Jaramillo Mauricio, Rodriguez Florencia, Islas Maria Soledad, Gonzalez Mariana.* Paper number 334447

**Introducing a novel approach for microplastic analysis in river sediments** *Kurzweg Lucas, Schirrmeister Sven, Adomat Yasmin, Harre Kathrin, Socher Martin.* Paper number 334564

### **A "sediment transport" perspective on microplastics movement: incipient motion** Varrani Arianna.

Sediment transport in freshwater environments is generally divided into two modes: suspended load and bedload. In our anthropized rivers, both modes can include natural sediments (like sand and gravel) and artificial particles (such as macro- and microplastics-MP). The understanding of how these particles behave in water needs experimental evidence of their transport dynamics, which may take the form of bedload, especially for those MP which have a specific density higher than water. For this reason, mobile-bed laboratory experiments have been performed in a recirculating flume to evaluate the hydrodynamics conditions, namely the discharge, leading to initiation of motion (i.e. incipient motion) of single bed grains. The bed layer was composed by MP grains with a diameter around 3 mm. In sediment transport studies, the definition of incipient motion conditions is a knotty point since the fundamental studies made by Shields in 1936, which identified the threshold conditions for the transport of natural materials. The preliminary results show that, for PBT ellipsoidal particles and PVC cylindrical grains, the threshold for motion is comparable with lighter materials investigated by Shields. To confirm these initial findings, more laboratory investigations are needed, varying the flow conditions, the MP material and the bed composition.

Keywords : bedload , incipient motion , laboratory experiments , sediment transport

<ul> <li>Introduction         <ul> <li>Introduction             <ul> <li>Introduction             <ul> <li>Introduction                  <ul> <li>Introduction</li></ul></li></ul></li></ul></li></ul></li></ul>	Institute of Geophysics Polish Academy of Sciences	Arianna Varrani of Geophysics, Polish Academy of Sciences, Warsaw, P avarrani@igf.edu.pl	oland
Introduction Introduction Introduction Introduction Introduction Interview Inter	Abstract Plastics entering rivers are transported by the c experimental evidence lack, hindering a deeper under experimental study is here presented. The aim was to in esults show that, for transitional to turbulent flows, bediver	urrent. The transport mechanisms for this anthropic debris standing of the transport process. Borrowing the meth dentify the conditions for the onset of motion of plastic p ad motion occurs for mean bed shear stresses	, is supposedly similar to that of natural sediments, but ods from sediment transport research, a preliminary articles lying on a uniformly composed plastic bed. The
<ul> <li>Flow depth</li> <li>Flow velocity</li> <li>Invall resistance</li> <l< td=""><td>Introduction Introduction livers and streams act as vectors of plastic debris to heir final sink. Transport mechanisms for lightweight naterials are assumed to follow the same principle as or natural heavier sediments. The theory developed or determining the threshold conditions - in natural ediments (started with the pioneering work by A. hields, 1936) can be therefore used for a first ssessment of erosion behaviour. Methods The experimental setting sees a 5 m-long flume, bedded with a 5 cm-thick layer of plastic particles. A series of runs with steady flow conditions and ncreasing discharges allowed to see the onset of bed particles movement. Incipient motion conditions were related to the presence of bedload transport in the nterrogation area. From the flow variables, the average bed roughness was computed, therefrom the mean bed shear stress was estimated. This was compared with a theoretical ange of values derived by Shields' data for transitional</td><td><math display="block">\mathbf{Figure 1: Investigated range for incipient motion threshold (modified from Komar, 2007) These values were used to define the upper and lower threshold for incipient motion conditions. PBT ellypsoids <math display="block">\mathbf{PT ellypsoids}</math></math></td><td><ul> <li>The present analysis of threshold of plastic particles is in line with previous results for laminar flows by Waldschläger &amp; Schüttrumpf (2019) and transitional to turbulent flow by Ballio &amp; Radice (2007).</li> <li><b>Conclusions</b></li> <li>Clearly, more detailed information about the near-bed flow field are needed, to estimate the near-bed velocity which relates to triggering of particle movement. Still in most cases the only available data regard discharge and water depth, and therefore the simple methods briefly outlined can be of use in case of lack of detailed information on the near-bed the flow field.</li> <li><b>Ballio</b>, F., &amp; Radice, A. (2007). Grain kinematics in weak linear transport. <i>Archives of Hydro-Engineering and Environmental Mechanics</i>, 54(3), 223-242.</li> <li>Komar, P. D. (2007). The entrainment, transport and sorting of heavy minerals by waves and currents. <i>Developments in Sedimentology</i>, 58, 3-48.</li> <li>Shields, A. (1936). Anwendung der Aehnlichkeitsmechanik und der Turbulenzforschung auf die Geschiebebewegung. <i>PhD Thesis Technical University Berlin</i>.</li> <li>Waldschläger, K., &amp; Schüttrumpf, H. (2019). Erosion Behavior of Different Microplastic Particles in Comparison to Natural Sediments. <i>Environmental Science &amp; Technology</i>, 53(22), 13219-</li> </ul></td></l<></ul>	Introduction Introduction livers and streams act as vectors of plastic debris to heir final sink. Transport mechanisms for lightweight naterials are assumed to follow the same principle as or natural heavier sediments. The theory developed or determining the threshold conditions - in natural ediments (started with the pioneering work by A. hields, 1936) can be therefore used for a first ssessment of erosion behaviour. Methods The experimental setting sees a 5 m-long flume, bedded with a 5 cm-thick layer of plastic particles. A series of runs with steady flow conditions and ncreasing discharges allowed to see the onset of bed particles movement. Incipient motion conditions were related to the presence of bedload transport in the nterrogation area. From the flow variables, the average bed roughness was computed, therefrom the mean bed shear stress was estimated. This was compared with a theoretical ange of values derived by Shields' data for transitional	$\mathbf{Figure 1: Investigated range for incipient motion threshold (modified from Komar, 2007) These values were used to define the upper and lower threshold for incipient motion conditions. PBT ellypsoids \mathbf{PT ellypsoids}$	<ul> <li>The present analysis of threshold of plastic particles is in line with previous results for laminar flows by Waldschläger &amp; Schüttrumpf (2019) and transitional to turbulent flow by Ballio &amp; Radice (2007).</li> <li><b>Conclusions</b></li> <li>Clearly, more detailed information about the near-bed flow field are needed, to estimate the near-bed velocity which relates to triggering of particle movement. Still in most cases the only available data regard discharge and water depth, and therefore the simple methods briefly outlined can be of use in case of lack of detailed information on the near-bed the flow field.</li> <li><b>Ballio</b>, F., &amp; Radice, A. (2007). Grain kinematics in weak linear transport. <i>Archives of Hydro-Engineering and Environmental Mechanics</i>, 54(3), 223-242.</li> <li>Komar, P. D. (2007). The entrainment, transport and sorting of heavy minerals by waves and currents. <i>Developments in Sedimentology</i>, 58, 3-48.</li> <li>Shields, A. (1936). Anwendung der Aehnlichkeitsmechanik und der Turbulenzforschung auf die Geschiebebewegung. <i>PhD Thesis Technical University Berlin</i>.</li> <li>Waldschläger, K., &amp; Schüttrumpf, H. (2019). Erosion Behavior of Different Microplastic Particles in Comparison to Natural Sediments. <i>Environmental Science &amp; Technology</i>, 53(22), 13219-</li> </ul>
	<ul> <li>flow depth</li> <li>grain diameter</li> <li>wall resistance</li> <li>energy losses</li> <li>flow velocity</li> <li>flow depth</li> <li>flow depth</li> <li>flow depth</li> <li>shear stress</li> </ul>	Figure 2: Values of mean bed shear stresses exceeds the threshold range for incipient motion for both materials.	<b>Acknowledgements</b> Sincere thanks to prof. Radice, for hosting me at his laboratory; dr.ing. Mrokowska for setting up a new lab at IG-PAS; and dr.ing Nones for countless support and inspiration. The work was supported within statutory activities No. 3841/E-232 41/S/2020, Ministry of Science and Higher Education of Poland.

A "sediment transport" perspective on microplastics movement: incipient motion

### Accelerated hydrolysis method for producing partially degraded polyester microplastic fiber reference materials

Sarno Antonio, Olafsen Kjell, Kubowicz Stephan, Sørensen Lisbet, Booth Andy M..

Microplastic fibers (MPFs) from textiles contribute significantly to the microplastic (MP) load in many environmental matrices. MPFs have been shown to be readily ingested by a wide range of organisms representing multiple trophic levels and have been shown to negatively impact organisms. Most fate and effect studies to date rely on pristine reference MP materials that have limited relevance compared to the partially degraded MP particles and fibers typically present in the natural environment. The current study aimed to develop and validate a rapid method for generating environmentally relevant polyester (PET) MPF reference materials with controllable levels of degradation. Importantly, the method produced the same degradation products, terephthalic acid (TA) and ethylene glycol (EG), as those generated during natural UV (sunlight) exposure of PET. Alkaline hydrolysis provided linearly increasing degrees of degraded PET MPFs over just a few hours, with full decomposition into molecular fragments occurring after 3 hours. The extent of physical degradation was determined by scanning electron microscopy, while chemical degradation was quantified by measuring the production of TA and EG degradation products. The proposed accelerated hydrolysis degradation method is relevant for producing partially degraded PET MPF reference materials for use in fate and effect studies and has the potential for application to other polymer susceptible to hydrolysis.

Keywords : ethylene glycol , microfiber , polyester , terephthalic acid , UV degradation

### Accelerated hydrolysis method for producing partially degraded polyester microplastic fiber reference materials

SINTEF

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### Introduction

Microplastic fibers (MPFs) derived from synthetic textiles and other sources represent a significant proportion of the microplastic (MP) load in many environmental matrices<sup>1-3</sup> and have been shown to have negative impacts on aquatic organisms  $^{\rm 4,\,5}.$  However, the majority of studies investigating the fate and effects of MP particles have employed commercially available pristine spherical reference materials, limiting the relevance of the data produced<sup>6, 7</sup>. Recent studies have explored ways to produce more environmentally relevant MP reference materials<sup>1-3,8</sup>, but they do not reflect the partially degraded nature of MPs and MPFs in the natural environment.

UV-induced oxidation (photodegradation) is the most effective degradation mechanism for many plastic materials released into the natural environment<sup>9,10</sup>. These physicochemical changes are important when trying to assess the risks associated with MP pollution, as they may influence MPF environment fate, bioavailability and effects on organisms<sup>6,11-13</sup>. However, photodegradation of polymers is still very slow (weeks to months) under typical environmental conditions<sup>9,14-16</sup>. There is, therefore, a need for accelerated degradation methods that allow rapid and controlled simulation of environmental degradation mechanisms for the production of partially degraded MP and MPF reference materials6,17

Here we demonstrate a fast and cheap hydrolytic degradation method for producing environmentally relevant, partially degraded polyester (PET) MPFs reference materials for use in fate and effects studies. The degree of degradation was quantitatively determined by measuring the PET degradation products terephthalic acid (TA) and ethylene glycol (EG), allowing specific levels of degradation to be achieved. Scanning electron microscopy (SEM) imaging was used to visualize the degree of physical modification resulting from degradation process. Finally, the environmental relevance of the accelerated hydrolysis method was verified by comparing the hydrolysed PET and degradation products to PET MPFs exposed to UV irradiation in seawater

### Take-home message

- · developed a cheap and fast method to degrade PET microfibers to use as reference material
- the extent of hydrolysis can be monitored by LC-UV and/or LC-MS analysis of PET degradation products (ethylene glycol and terephthalic acid)
- the same degradation products generated by photodegeneration
- · our method generates reference material that resembles weathered PET microfibres, although there are slight differences



### **Methods**

### Accelerated hydrolysis

200 mg of PET MPFs were introduced in glass bottles containing 25 mL aqueous solution of NaOH (10 % NaOH; pH 14) and hydrolysis was conducted at 90 °C using an oil bath.

### UV degradation

UV degradation of PET fibers was performed using a Suntest CPS+ (Atlas Material Testing Solutions) equipped with a xenon UV lamp (1500 W) and fitted with a natural daylight filter. PET fibers were mixed into MilliQ or seawater at a concentration of 8 mg mL  $^{\rm 1}$  and placed into 35 mL quartz tubes with glass stoppers. Irradiation was conducted at 65 W/m<sup>2</sup> and the temperature in the exposure chamber was maintained at 24 ± 3 °C.

### LC-UV-MS/MS analysis of degradation products

EG was derivatized by adding 50  $\mu$ L sample, 100  $\mu$ L 4 M NaOH and 50  $\mu$ L benzoyl chloride to 250 µL with deionized water. The reaction was vortexed and incubated at room temperature for 5 min. Next, the reaction was quenched by adding 50  $\mu L$  10% glycine and incubating for 3 min at room temperature. The dibenzoyl derivate was then extracted with 1 mL pentane. Phase separation was accomplished by centrifugation at 10 000 xg for 5 min, and the organic phase was evaporated under nitrogen at 50 °C. The dried extract was dissolved in 800  $\mu$ L 10 mM ammonium formate (pH 2.8) in 50% acetonitrile. Derivatized samples were diluted 500-fold prior to analysis. TA was analysed without derivatization by diluting samples 100-fold prior to analysis. Samples were analysed on an Agilent 1260 LC coupled to a 4670 mass spectrometer using a ZORBAX Eclipse Plus C18 column (2.1x50 mm, 1.7  $\mu\text{m})$  with 10 mM ammonium formate and acetonitrile as the mobile phase

### Scanning electron microscopy

MPFs were mounted on double sided tape and coated in a thin layer of evaporated gold to make them conductive. MPF imaging was performed (1000x magnification) using a FEI Nova Nano SEM 650 scanning electron microscope (SEM), operated in high vacuum mode at accelerating voltages of 5 kV, with an emission current of 90 pA and at a working distance of 8 mm.

### **Results and discussion**

### TA and EG assav

Accelerated hydrolytic degradation method development

PET fibers were subjected to alkaline hydrolysis over time to determine whether they are suitable for use as partially degraded MPF reference materials. Physical and chemical degradation was studied using a combination of SEM, gravimetry and LC-UV-MS/MS. SEM analysis showed superficial fiber degradation occurs after 30 min, with significant structural damage observable after

1 h. After 6 h, substantial structural degradation had occurred, and no material was detected after 24 h (Figure 2A). LC-MS/MS analysis of PET degradation products similarly showed a linear time-dependent increase in both TA and EG degradation products that plateaued after 2.5 to 3 h, increasing slightly at 24 h (Figure 2B). This was inversely mirrored when measuring undissolved PET gravimetrically (Figure 2B).





Figure 2: Accelerated hydrolysis fully degrades PET. A) SEM images of PET subjected to accelerated hydrolysis (1000x magnification). B) TA and EG measured by LC-UV-MS/MS plotted with undissolved PET determined gravimetrically during accelerated hydrolysis

We suggest that by 6 h, the fibers are degraded into a mixture of monomers (i.e. EG and TA) and short water-soluble polymers. At this point, the PET fibers are almost undetectable by SEM. Full hydrolysis of the water-soluble polymers into the final degradation products occurs by 24 h. Thus, alkaline hydrolysis of PET fibers can be used to generate reference material of partially degraded MPF by attenuating hydrolysis time.

### Comparison of hydrolysis and UV degradation

We confirmed whether PET degradation by alkaline hydrolysis resembles UV-degradation that occurs in the environment. To this end, we exposed PET fibers in seawater to simulated sunlight and measured the generation of EG and TA as described above. Small holes were observed in the fibers exposed to UV after ~33 days (Figure 3A), suggesting the onset of physical changes to



the PET MPFs. Both TA and EG were measured after only 6 days and steadily increased throughout the experiment (Figure 3B). Controls incubated in the dark at room temperature over the same amount of time contained no measurable EG or TA.

We conclude that PET degradation can be measured by TA and EG, and by extension that the degree of fiber degradation in our reference material is a valid benchmark for environmental fate studies. As hydrolysis occurs mainly in polymers that have water-sensitive groups in the polymer backbone, such as polyesters (including polyethylene terephthalate), polyanhydrides, polyamides, polyethers and polycarbonates, the method described above has potential application for producing a broad range of partially degraded plastic reference materials.

re 3: UV-irradiated PET re cid and ethylene glycol. A) SEM images of UV irradiated PET fibers. B) TA nd EG red by LC-UV-MS/MS

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### Characterization of microplastics in sediments from the submarine outfall of the wastewater treatment plant, Mar del Plata City (Argentina)

Diaz-Jaramillo Mauricio, Rodriguez Florencia, Islas Maria Soledad, Gonzalez Mariana.

Despite widespread detection of microplastic pollution in the marine environment, data describing microplastic inputs to the coast of Argentina are scarce. Particularly, the discharge of municipal wastewater treatment plant is considered an important pathway for microplastic to aquatic environments, but only a few studies are available. Mar del Plata city (38° S, 57° W), is the main touristic resort of the Atlantic coast of Argentina. Wastewater plant from Mar del Plata city (Argentina) discharges into marine environment (4 km offshore) by a submarine outfall. Although this outfall is working since 2014, diminishing the biological and chemical pollution of nearest coastal areas, its role as microplastics source to the marine environment was not evaluated. This work, showed a preliminary study of microplastic occurrence and characteristics in subtidal sediments (11-21 mt deep) from two sites located close to the submarine outfall of wastewater plant. The microplastic content (n: 3 samples per site) was determined by floating method using a zinc chloride (ZnCl2) solution. No significant differences were found between sites (p¿0.05). Mean total microplastic abundance in sediments between both sampling sites ranged from 411 to 547 items per kg d.w. The presence of fibers (40-72%), fragments (12-31.5%), films (12.1-29%) and foam (0-4%) were detected as probable microplastic particles. Transparent (34.7%), white (24.2%), black (13.8%) and red (10.8%) type particles were observed as the main color contribution of microplastic. The results obtained represent a first approach to determine the presence of microplastics in sediments under the influence of the submarine outfall from wastewater treatment plant in Argentinian coastal cities. This work is part of UE-CONICET project Coastal ecosystems: Structure, functioning, dynamics and management strategies.

Keywords : Argentina., Microplastic, submarine outfalls, subtidal sediments, wastewater treatment

### CHARACTERIZATION OF MICROPLASTICS IN SEDIMENTS FROM THE SUBMARINE OUTFALL OF THE WASTEWATER TREATMENT PLANT, MAR DEL PLATA CITY (ARGENTINA).



### Mauricio Díaz-Jaramillo<sup>1</sup>, Florencia Rodriguez<sup>\*1</sup>, Maria Soledad Islas<sup>1,</sup> and Mariana Gonzalez<sup>1</sup>

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Despite widespread detection of microplastic (MPs) pollution in the marine environment, data describing point sources into to the South Atlantic coast are scarce. Particularly, the discharge of municipal wastewater treatment plants (WWTP) is considered an important pathway for MPs entering to aquatic environments. Mar del Plata city (38° S, 57° W) with a population of 750.000 people, is the main touristic resort of the Argentinian coast and its WWTP discharges their effluents into the marine environment by a submarine outfall (4 km offshore). Although this outfall is working since 2014, diminishing the biological and chemical pollution of nearest coastal areas, but their role as MPs source to the marine environment was not yet evaluated. This work represents the first approach in the study of MPs occurrence/characteristics in subtidal sediments (11-21 mt deep) from different sites differing in their proximity to the WWTP submarine outfall.


### Introducing a novel approach for microplastic analysis in river sediments

Kurzweg Lucas, Schirrmeister Sven, Adomat Yasmin, Harre Kathrin, Socher Martin.

Microplastics (MP) in environmental compartments are a topic of growing concern. The understanding of fate and impact of microplastic is still hampered by a lack of comparability between studies and time-consuming processing and analysis methods. Therefore, the establishment of a reliable and cost-efficient method for environmental monitoring is urgently required. Most of the common used methods do not fit all requirements. They may not give chemical confirmation of the polymer or are highly time-consuming. Here we want to present a novel approach for MP analysis in river sediments combining electro separation (corona roller separator, Hamos, Prenzberg, Germany) and differential scanning calorimetry (DSC, Netzsch, Germany). Microplastic particles of ultra-high molecular weight polyethylene (PE-UHMW, size range: 100 to 200 µm) were separated from particulate matrices such as river sediment or sand. Samples with MP concentrations from 100 to 1 ppm (parts per million) PE-UHMW were investigated. Recovery rates of the polymer in the enriched fractions were determined using the thermodynamic fingerprints measured by DSC. The pattern of the DSC curve with phase transition temperatures and enthalpies allows the identification and quantification of the polymer type. Electro separation removed  $99.91 \pm 0.03$  percent of the initial matrix from an artificial sediment. In environmental samples,  $93.0 \pm 1.0$  percent to  $99.52 \pm 0.06$ percent of the initial matrix were removed, depending on the particle size distribution, bulk density and total organic carbon. The recovery rate for the PE-UHMW was determined as 91 ± 5 percent in the artificial sediment. We determined the limit of quantification to 40 ppm and the limit of identification to 1 ppm. Recovery rates are lower for environmental samples then for artificial matrices. Moreover, 5 samples could be processed in 8 hours after drying. In conclusion, our approach has potential as a reliable and fast method for monitoring of highly polluted environmental compartments.

Keywords : differential scanning calorimetry , DSC , electro separation , microplastic , monitoring , river sediment



HOCHSCHULE FÜR TECHNIK UND WIRTSCHAFT DRESDEN UNIVERSITY OF APPLIED SCIENCES

Faculty of Civil Engineering Faculty of Electrical Engineering Faculty of Agriculture/Environment/Chemistry

### Introducing a Novel Technique for Microplastic Analysis in River Sediments

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### Motivation and Goals:

- 1) Microplastic (MP, particles < 5 mm) is of growing concern, however no standardized method for MP monitoring is available.
- 2) Development of a fast, low-cost and reliable method ensuring a high sample load is challenging.
- 3) A novel method combining electrostatic separation (ES) and differential scanning calorimetry (DSC) is presented to address these challenges.

### **Combination of Electrostatic Separation and DSC**





### **Application to Environmental Samples** sampling electrostatic separation **DSC** measurement no MP in sample 1-14 and 16-20 20 locations, sampled in spring 2020 separation parameters: mass flow: 1.0 kg/h. drum rotation: 66 rpm. Peak: 206.5 °C. 6.36 mV sample 15: melting signal MP determination in environmental samples using high voltage: 20 kV Area: 5.461 mJ electrostatic separation and DSC identification by DSC: PA vast deviation of confirmation of PA in DSC crucible by mass reduction in FTIR-microscopy Fig. 5: thermogram of sample 15 environmental samples (Fig. 4) sample 15 POLYAMIDE 6 + POLYAM fit: 74.48 % optimization of ES needed to fit: 73.73 % [2] increase removal fit: 64.17 9 fit: 54.97 % 4: removal of sediment from fit: 53.53 9 Fig. environmental samples Fig. 3: sampling locations in Saxony, overall 20 sites Fig. 6: IR-spectrum of the particle (red) and spectra with best fit from the device were sampled library

### **Density Separation to Optimize Mass Reduction**

Mass reduction with a combination of electrostatic separation and density separation was tested

### Comparison of the thermograms





Spike Sample "Sand" area: 147.5 //g after DS after DS

Fig. 8: thermograms of samples from sand and river sediment 2 after ES and after DS

- 2 different sediments show a mass reduction above 99.9 %
- DSC measurements show increased peaks after density separation
- The DS in combination with ES shows a great potential for an application in environmental monitoring

### Conclusion

- combination of electrostatic separation and differential scanning calorimetry is promising for MP monitoring
- fast, cost-efficient and high throughput of samples
- subsequent MP enrichment improves the reliability for DSC measurement
- properties of different sediments are not negligible as shown in experiments

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### Session 24.10\_Me. Chaired by Patricia Ostiategui, Gran Canaria

**Microplastics (MP) in gills and gastrointestinal tract (GIT) of Antarctic fish** *Masserey Lea, Erni-Cassola Gabriel, Burkhardt-Holm Patricia.* Paper number 334199

**Microplastic vector effects: are fish at risk when exposed via the trophic chain?** *Agathe Bour, Sturve Joachim, Höjesjö Johan, Carney Almroth Bethanie.* Paper number 334201

### Looking at the macro is no longer enough: a protocol to address the study of microplastic intake in stranded cetaceans

Montoto-Martínez Tania, De La Fuente Jesús, Puig-Lozano Raquel, Marques Nuno, Fernández Antonio, Gelado-Caballero M<sup>a</sup> Dolores. Paper number 334269

**The influence of microplastics on vertical water flow in sediment columns** *Ehl Ramona, Laermanns Hannes, Horn Julia, Steininger Florian, Bogner Christina.* Paper number 334293 **Microplastics (MP) in gills and gastrointestinal tract (GIT) of Antarctic fish** Masserey Lea, Erni-Cassola Gabriel, Burkhardt-Holm Patricia.

Microplastic pollution has been recognised worldwide and occurs at economic, social and environmental costs, with potentially high ecological impacts on marine ecosystems (Andrady 2017). In the Southern Ocean, Microplastics (MP) have been found in surface water and sediments, but little is known about the interaction with Antarctic fish (Waller et al 2017). With the contamination of remote marine areas by fragmented debris and increasing human presence in Antarctica (Tin et al 2009), the levels of MP pollution are likely to rise accordingly, a reason of concern for members of CCAMLR (Waller et al 2017). Hence, it is crucial to better understand the scale and fate of MP pollution in Antarctica, especially its potential uptake by living organisms, affecting other trophic levels. The master thesis aims at supporting the understanding of MP pollution in the Antarctic ecosystem through its potential occurrence in fish. Antarctic silverfish (Pleuragramma antarcticum) from the Antarctic Peninsula and the Eastern Weddell Sea are investigated. The objectives are to determine (i) whether MP are present in Antarctic fish (ii) whether the abundance of MP differ according to the geographic area (iii) whether the occurrence of MP differ between GIT and gills and (iv) the type of MP that are ingested by Antarctic fish. The gastrointestinal tract (GIT) and gills of 94 fish caught in 2014 and 2018 are analysed. Samples are treated according to the enzymatic digestion protocol of Cole et al. (2015) to dissolve the organic matter. Potential plastic particles ¿300µm are analysed with ATR-FTIR and particles between 10-300µm with µ-FTIR. To limit airborne contamination, preventing measures, such as working under a mosquito net, wearing cotton clothes, filtrating all liquids, carefully cleaning utensils, and negative blanks, are taken for each methodological step.

Keywords : Antarctic fish , Enzymatic digestion , FTIR , Microplastic pollution , Sustainability







**Microplastic vector effects: are fish at risk when exposed via the trophic chain?** Agathe Bour, Sturve Joachim, Höjesjö Johan, Carney Almroth Bethanie.

Trophic transfer has been identified as an important exposure route to microplastics (MPs) in predator species. Many chemicals are sorbed on MPs and can potentially be transferred to organisms in contact with MPs, including via the trophic route ("vector effect" of MPs). However, there is currently no consensus on whether MPs represent a significant exposure pathway to chemicals in contaminated habitats, where other contamination pathways could be predominant. Despite their relevance, effect studies, including vector effect studies, on fish exposed to MPs via trophic chains are currently very scarce. To fill that gap, we exposed three-spined stickleback (Gasterosteus aculeatus) to MPs via prey ingestion, in a long-term experiment. MPs were either pristine or spiked with chlorpyrifos (CPF), an organophosphate pesticide. To understand the specific role of MPs as a vector of contamination, a condition were preys were directly contaminated with CPF in solution was also included. CPF accumulation was observed in fish exposed to CPF-spiked MPs (MP-CPF), confirming the vector potential of MPs. However, CPF accumulation was more important in fish exposed to CPF via prey. Interestingly, CPF organ distribution differed between groups, suggesting that chemical exposure via MPs could alter organ distribution of chemicals. This can result in a change in the organs most at risk, likely increasing intestine exposure. We also assessed the effects of such exposure on fish, including neurotoxicity (AChE inhibition) and organism performance (feeding, locomotion and environment exploration behavior). Different patterns were observed depending on the exposure conditions. Especially, we observed significant AChE inhibition and hyperactivity in fish exposed to MP-CPF, which could result in increased vulnerability to predation.

Keywords : behavior , chlorpyrifos , ecotoxicity , uptake



### Microplastic vector effect: are fish at risk when exposed via the trophic chain?<sup>1</sup>

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### Looking at the macro is no longer enough: a protocol to address the study of microplastic intake in stranded cetaceans

Montoto-Martínez Tania, De La Fuente Jesús, Puig-Lozano Raquel, Marques Nuno, Fernández Antonio, Gelado-Caballero M<sup>a</sup> Dolores.

Marine debris can impact biodiversity in a number of ways, and its effects may vary depending on the type and size of the debris and the organisms that encounter it [1]. Since the first evidence of a marine mammal's interaction with plastic intake, there have been a number of studies on this subject, together with alarming images of stomachs full of marine debris and a growing concern about it. However, very little is known about the presence of microplastics in higher trophic level species such as cetaceans [2]. Up to more recently, they were primarly focused on the study of particles larger than 2.5 cm, and therefore failing to assess the microlitter presence, which remains a challenging task due to large gut content volumes and the difficulties of sampling following careful airborne contamination prevention protocols. Working with stranded cetaceans (n=12), which represent a significant opportunity to study the interaction of marine fauna with plastic debris, we have validated a protocol for microplastic ingestion studies that serves to obtain samples from different multidisciplinary teams (i.e. veterinary and marine sciences schools), without interfering in the work of any of the parties [3]. The successful table set up used for the extraction of microplastic particles from the gastrointestinal contents was proofed advantageous and applicable by any research group that already counts with the necessary facilities to perform cetaceans autopsy analysis, fulfilling the harmonisation needs as explicated by Panti et al. [4]. This approach is fully compatible with necropsy protocol in cetaceans [5], and at the same time complies with the recommendations for reporting ingested plastics in marine megafauna [6]. The proposed workflow allows the collection of valuable data for different interdisciplinary research teams, aiming to harmonize data, facilitate largescale comparisons of plastic ingestion and also give scientific basis to future conservation policies.

Keywords : megafauna , microplastics , necropsy , protocol , stranded cetaceans

### LOOKING AT THE MACRO IS NO LONGER ENOUGH: A PROTOCOL TO ADDRESS THE STUDY OF MICROPLASTIC INTAKE IN STRANDED CETACEANS

**MICR0**2020 INTERNATIONAL CONFERENCE I LANZARIOTE AND BEYOND<sup>\*</sup> FATE AND IMPACTS OF MECROPLASTICS: KNOWLEDGE AND RESPONSIBILITIES

Tania Montoto-Martínez<sup>1\*</sup>, Jesús De la Fuente<sup>2</sup>, Raquel Puig-Lozano<sup>2</sup>, Nuno Marques<sup>3</sup>, Antonio Fernández<sup>2</sup> and M<sup>a</sup> Dolores Gelado-Caballero<sup>1</sup> <sup>1</sup> Research Group on Environmental Technologies, Management and Biogeochemistry. University of Las Palmas de Gran Canaria, Canary Islands, SPAIN; <sup>2</sup> Atlantic Centre of Cetaceans Research. University Institute for Animal Health and Food Safety, University of Las Palmas de Gran Canaria, Canary Islands, SPAIN. <sup>3</sup> Museu da Baleia, Canical , Madeira, PORTUGAL. \* Correspondence author: tania.montoto@ulpgc.es

Which were the knowledge gaps found?

prevention protocols.

What were the keys to success?

Table design: A custom-made

adaptation to the necropsy table was

made connecting its drainage to a set

of three stacked metal sieves (1000,

500 and 200  $\mu$ m) where the washed

stomach contents were retained

after thorough gastrointestinal

(GIT) rinses.

of particles larger than 2.5 cm, and therefore fa

Keywords: microplastics, protocol, stranded cetaceans, necropsy, megafauna.

### INTRODUCTION

Marine debris can impact biodiversity in a number of ways, and its effects may vary depending on the type and size of the debris and the organisms that encounter it [1]. Since the first evidence of a marine mammal's interaction with plastic intake, there have been a number of studies on this subject, together with alarming images of stomachs full of marine debris and a growing concern about it.



### WHAT DID WE DO?

Working with stranded cetaceans, which represent a significant opportunity to study the interaction of marine fauna with plastic debris, we have validated a pro debris, we have validated a protocol for microplastic ingestion studies that serves to obtain samples from different multidisciplinary teams (i.e. veterinary and marine sciences schools), without interfering in the work of any of the parties [3].

### THE PROTOCOL WORKFLOW



### Before you start:

(i) Wear cotton clothes while manipulating the samples,

- (ii) Clean all containers using distilled water prior to its reuse,
- (iii) Perform blank controls filtering MilliQ water,
- (iv) Place a clean petri dish with a filter paper close to the manipulation area to register possible airborne contamination,
- (v) Cover all samples with aluminium foil after each single step and during the procedure.

Once the extraction of the digestion tract is performed and the necessary samples for anatomopathological, microbiological, parasitological and dietary studies are collected:



One by one, empty the digestive content of each GIT section on the table and rub the mucous membranes thoroughly until all the digestive remains have been washed away.

Carry out a macroscopic examination and take samples for histopathology and microbiology.

Rinse carefully so all the digestive content is retained in each of the sieves described above.

The filtered remains will be introduced into different containers for each organ (oesophagus, stomach and duodenal ampulla, and intestine) and will be stored for their subsequent laboratory study.

Check out the complete protocol at:

### References

 $( \bigcirc )$ 

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😹 protocols.io







However, very little is known about the presence of microplastics in higher trophic level species such as cetaceans [2].

And moreover, up to more recently, they were primarly focused on the study

and the difficulties of sampling following careful airborne contamination

ice, which remains a challenging task due to large gut content volumes

Team work: The protocol is simple and cost-effective and allows research teams to collect and analyse samples of the presence of microlitter in a comparable way, therefore reaching a more thorough understanding of the risk microplastics pose to marine mammals.

### CONCLUSIONS

The successful table set up used for the extraction of microplastic particles from the gastrointestinal contents was proofed advantageous and applicable by any research group that already counts with the necessary facilities to perform cetaceans autopsy analysis, fulfilling the harmonisation needs as explicated by Panti et al. [4].

This approach is fully compatible with necropsy protocol in cetaceans [5], and at the same time complies with the recommendations for reporting ingested plastics in marine megafauna [6]. The proposed workflow allows the collection of valuable data for different interdisciplinary research teams, aiming to harmonize data, facilitate large-scale comparisons of plastic ingestion and also give scientific basis to future conservation policies.

### The influence of microplastics on vertical water flow in sediment columns

Ehl Ramona, Laermanns Hannes, Horn Julia, Steininger Florian, Bogner Christina.

The need for research regarding the occurrence and effects of microplastic (MP) in the environment has become apparent by numerous studies. However, even though release rates into terrestrial environments are suggested to be 4-23 times higher than those into oceans (Horton et al., 2017, doi: 10.1016/j.scitotenv.2017.01.190), research about MP in soils is only just starting. The first studies reported that MP influenced soil physical properties. For example, it can be incorporated into soil aggregates (de Souza Machado et al., 2018, doi: 10.1021/acs.est.8b02212), or clog soil pores (Zhang et al., 2019. doi.org/10.1016/i.scitoteny.2019.03.149) possibly affecting soil hydraulic properties. In this study we aim at increasing the mechanistic understanding of the influence of MP particles on vertical water flow. We irrigated soil columns filled with simple substrates with and without the addition of MP particles (polystyrene, 0.1% or 0.5% of dry weight, homogeneously mixed). To compare the arrival times of the irrigation water, NaCl was used as tracer. The arrival times of the drainage and tracer fronts were characterized by the viscous flow approach (e.g. Bogner & Germann, 2019, doi:10.2136/vzj2018.09.0168). Furthermore, we analysed the drainage water for possible MP leaching out of the soil columns by fluorescence microscopy after staining with Nile Red (Konde et al., 2020, 10.1016/j.marpolbul.2020.111475). Preliminary results indicate that MP changed the infiltration time of irrigation water substantially.

Keywords : hydraulic properties , polystyrene , viscous flow approach , water flow



### The influence of microplastics on vertical water flow in sediment columns [334293]



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Germany **CRC 1357 Microplastics** 

### Aim

Mechanistic understanding of behaviour and transport of microplastics (MP) in soils (Fig.1)

- Model system soil column
- Here: analysis of flow patterns of water in sediment columns with MP



### **Column experiments**

### Setup

- <sup>1</sup> Irrigation of sediment columns with and without MP (Fig. 2)
- Tracing the "new" water with NaCl by conductivity measurement Parameters
- Column diameter 8.4 cm, height 10 cm, fill up to 8 cm
- Duration of irrigation: 8760 sec
- Flow rate: 2 ml/min (21.7 mm/h)
- NaCl<sub>(aq)</sub> concentration in irrigation water: 1 g/l
- MP type: amorphous polystyrene (diameter 75-125 μm, 200-400 μm)
- MP addition: wg-0.1%, wg-0.5% (Fig. 2)
- Sediment texture: 88.87% sand, 11.13% silt [1]
- Sediment texture measured with Laser Diffraction Particle Size Analyzer (LS 13320 Beckmann Coulter<sup>™</sup>) [1]

### Quantitative analysis of MP leaching from the sediment column

### (I) Filtration of drainage samples

Vacuum filtration



Fig 4: Filtered sample

### (II) Staining with Nile Red Nile Red solution: 30 mg/L

in ethanol/acetone [2], [3]

Fig 5: Stained MP sampl



Fig 6: Staining process



Fig 7: a) sediment sample transmitted light microscope b) MP sample (75-125µm, 200-400µm) green fluorescence at exposure time 250 ms c) MP sample green fluoresc 148 ms d) sediment sample green fluorescence at exposure time 148 ms scence at exposure time

### **First results**





Fig 10: Comparison of tracer breakthrough times of MP particles (MP\_small = 75-125µm, MP\_large = 200 400µm), 0.1% MP, 0.5% MP, 1% MP, quartz particles (QZ\_small = 63-112µm, QZ\_large = 200-500µm) and

### **Conclusion and Outlook**

Large variability of tracer breakthrough times No size effect

Calculation of flow parameters based on Viscous Flow Approach [4] Processing of microscopy images for MP quantification

thout external material (n

C



a, G., Gibson, M. I., Thompson, Drnik, J., Prume ,J. A., Talber J. Forschungsgemeinschaft [4] Germann P (



Setup

0.5% MP

0.1% MP

88.9% sand 1.1% silt pump NaCl water 2 ml/min (21.7 mm/h) DE CO g/1

Fig 2: Experimental setup

(III) Detection of MP

Detection of MP with fluorescence microscopy

### Session 24.10\_Ma. Chaired by Sascha Müller, Copenhagen

Monitoring microplastics and their associated chemicals in deep water coral habitats Lim Aaron, Mateos Cárdenas Alicia, Wheeler Andy. Paper number 333710

**Transport of Microplastics in a choked coastal lagoon in south Brazil** Santos Ítele, Fernandes Elisa, Jalón-Rojas Isabel, Silva Pablo, Pinho Grasiela. Paper number 334426

**Adsorption behavior of chlorinated phenols on polyethylene in Danube River water** Lončarski Maja, Tubić Aleksandra, Vasiljević Sanja, Agbaba Jasmina. Paper number 334578

### Monitoring microplastics and their associated chemicals in deep water coral habitats

Lim Aaron, Mateos Cárdenas Alicia, Wheeler Andy.

Although microplastic pollution is ubiguitous, accurate quantification is still required and plastic associated chemicals from environmental samples remain largely unexplored. Given the difficulties associated with deep water data acquisition (e.g. costly and opportunistic sampling, weather dependency and engineering restrictions), much of the research carried out on marine plastics to date are either restricted by low spatial or temporal resolution, are isolated studies or are subject-specific in nature due to a lack a multidisciplinary approach. Deep-water microplastic monitoring is treated like a black-box system, an area of science still in its infancy even though the deep sea is estimated to be the major global microplastic hotspot. Preliminary video data collected by a Remotely Operated Vehicle (ROV) from an earlier project led by the team previously showed that large plastic items are abundant, especially fishing items, in deep water Irish coral reefs from the Porcupine Bank Canyon and Moira Mounds. The new study presented here expands on such previous knowledge of the area and focusses on microplastics by integrating a large spatial range, temporal resolution and novel methodologies. Microplastics and their associated chemicals are being analysed from samples collected by eight Benthic Lander systems and sediment traps deployed in 2019 and 2020. QA/QC techniques are given special importance to ensure the reliability of the analytical results produced. The main outcomes of this study are to (1) accurately quantify the abundance and fate of microplastics and their associated chemicals in deep sea Irish canyons, (2) the interactions and impacts to the health of cold-water corals present in Special Areas of Conservation (SACs) and (3) the potential cause for observed variability throughout time.

Keywords : associated chemicals , canyons , corals , deep sea , microplastics

# Monitoring Microplastics and their Associated

# $\bigcap$ hemicals in Cold Water Coral Habitats



Alicia Mateos Cárdenas, Aaron Lim and Andy Wheeler

C?



marinegeology.ucc.ie



- The deep sea is now Preliminary data collected are knowledge gaps considered a major plastic by UCC Marine Geology associated chemicals and microplastic and their hotspot (1,2), however there Team in 2019 showed that marine habitats the potential impacts to large plastic items are regarding the presence of
- This new project will areas

present in our sampling

abundance and their associated chemicals in cold investigate microplastic

### water coral habitats

ogen science, 1(4), 140317. Kane, L.A., Clare, M. A., Miranomes, E., Wogelius, R., Rothwell, J., J. Garreau, P., & Pohl F. (2020), Seaftoor microplastic horspots controlled by deep sea circulation. Science, 3686495). 1140-1145. Woodall, L. C., Sanchez-Vidal, A., Canals, M., Paterson, G. L., Coppock, R., Sleight, V., ... & Thompson, R. C. (2014). The deep sea is a major sink for microplastic debris. Royal Society on one science with vitamat











Photos credit: UCC Marine Geology Group Poster created with Canva

## <u>This project</u> (2020-2023)

Cold water corals

- <u>Study sites</u>: Porcupine Bank Conservation (SACs) status based on their Special Areas of Province (Fig.1), selected Canyon and Belgica Mound 0 Cold water corals are
- <u>Sediment samples:</u> collected water depth using landers (24 bottles per from cold water coral reefs lander) between 600-1100 m

• HPLC, GC-MS

 Landers constantly measure temperature, current speed environmental variables and direction: coral health microplastic presence & will be analysed based on



Microplastics associated chemicals and their

## <u>Techniques and methodologies:</u>

V characterisation microplastics extraction and Sample processing and

μ-FTIR, Raman, SEM

(Fig.2)

abundant in these SACs

chemicals from deep-sea Analysis of plastic-associated (ongoing)

transitional) to be spatially crosstrameworks (live, dead Size and status of coral referenced with lander data

S D

### **Transport of Microplastics in a choked coastal lagoon in south Brazil**

Santos Ítele, Fernandes Elisa, Jalón-Rojas Isabel, Silva Pablo, Pinho Grasiela.

Microplastics in the marine environment are known to have impacts on marine life, ecosystems, and ultimately to human life. Therefore, it is important to understand how these materials behave, once in the aquatic environment, and the main physical forcings responsible for their transport. In this study, the transport of microplastics in Patos Lagoon was studied based on the coupling between the TELEMAC-3D hydrodynamic model results and the microplastic transport model TrackMPD, which takes into consideration specific physical properties of microplastic particles (as density and shape) and transport processes associated to them. Simulations were carried out for the 2013 year, which is considered an ENSO-neutral year. Hydrodynamic results showed that current velocities inside the lagoon present spatial variability, with weaker current velocities (max 0.35 m.s-1) in the inner Lagoon and stronger current velocities (3 m.s-1) at the Patos Lagoon mouth. Differences were also observed between the margins as a function of the Lagoon morphology and predominant wind direction, with current velocities around 0.35 m.s-1 on the west and 0.20 m.s-1 on the east margin. These spatial differences in current velocity inside the lagoon suggests the occurrence of areas of microplastic displacement and/or accumulation. The hydrodynamic response of the system to the main physical forcing (wind and freshwater discharge) promotes alternate in/out fluxes into the Lagoon and that is expected to affect the residence time of plastic debris inside the system. Simulations with the TrackMPD model are now being developed and processed and from the results we expect to understand the dynamics of microplastics and define the preferential areas of deposition inside Patos Lagoon. These results are the very first step towards the understanding of how microplastics are transported in coastal systems in South Brazil and will provide valuable information for the management of the plastic problem in the region.

Keywords : Brazil, Coastal Lagoon, Microplastics, Transport

### 6'\_\_\_\_Transport of Microplastics in \_\_\_\_\_ a choked coastal lagoon in South Brazil

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### Introduction

Microplastics (MPs) are known to have impacts on marine life, ecosystems, and ultimately to human life. Research studies have addressed this problem in different terrestrial and marine environments. In transitional aquatic systems, such as coastal lagoons, however, they are still understudied. These areas are of high importance as they could be serving as a buffer zone for the accumulation and redistribution of plastic particles from continental sources to the marine environment.

Here, we are studying the transport of MPs in the Patos Lagoon, *the world's largest chocked coastal lagoon*, located in South Brazil and this poster shows the very early stages of the modelling effort to try to understand the dynamics of MPs in this environment.

Patos Lagoon has 10,360 km<sup>2</sup> and a drainage basin of 140,000 km<sup>2</sup>, with big cities bordering its margins. It is connected to the South Atlantic Ocean by a long and narrow entrance channel, located in its southernmost part, a nursery ground area where port activities take place.

### Methods

The transport of MPs in Patos Lagoon will be studied based on the coupling between TELEMAC-3D hydrodynamic model results and the plastic debris transport model TrackMPD. The latest was specifically designed for modelling plastic debris, taking into consideration a wide range of physical properties and transport processes associated with these particles (Jalón-Rojas, 2019).

Simulations will be carried out for 2013, which is considered an El Niño Southern Oscillation (ENSO)-neutral year, fact of relevance since Patos Lagoon is known to be affected by ENSO cycles. This study will serve as a baseline for other future scenarios.

### Preliminary Application of Track MPD in Patos Lagoon

**TELEMAC-3D** hydrodynamic results showed that current velocities inside the lagoon presented spatial variability, with overall weaker current velocities (0.35 m.s<sup>-1</sup>) in the MB of the Lagoon.

Differences were observed between the margins of Patos Lagoon due to the Lagoon's morphology and predominant wind direction, with current velocities around 0.35 m.s<sup>-1</sup> on the west margin and 0.20 m.s<sup>-1</sup> on the east margin.

These hydrodynamical results served as initial guides to check if these variables were having any effect on the particles placed on the MB.

The **TrackMPD** simulation showed that during the simulated period, 22% of the particles ended up being exported out of the modeled domain. The particles that stayed, were rearranged to what it seems four probable regions of higher accumulations. These regions are highlighted in the Figure 2G, which is relative to the last simulated time step.

These particle results when compared to the hydrodynamic results were correspondent to the low-speed profile of the currents inside the MB of the Lagoon, reflected by the low number of particles that left the domain during the simulated period. Also the patterns of accumulation in this scenario corresponded to specific areas of even lower current speeds and recirculation cells.

### Conclusions

Although there is still a range of variables and different hydrodynamic scenarios to be considered for realistic MPs simulations in Patos Lagoon, this preliminary scenario serves to show a preliminary application of the TrackMPD model in the region.

The accordance between areas of low current speed and accumulation showed by the two models gives the indication that TrackMPD is working as it should when applied to a complex coastal environment.

The objective forward is to make better and more realistic simulations that will serve as the first step towards the management of MPs in Patos Lagoon, helping researchers and policy makers to better address this problem.







Fig. 1 - Patos Lagoon. Red dots are the main cities bordering the lagoon. The yellow circle indicates the entrance channel.





To keep the discretization of important features in Patos Lagoon the domain for TrackMPD had to be divided (due to computational constrains) into two regions with different resolutions: The Main Body (MB) using a mesh of 500x500 and the Estuary Region (ER) with a mesh of 1000x1000.

Scenarios of robust MPs releases and behaviors are still being developed. This poster presents a preliminary example of the TrackMPD application considering the 2D trajectory of 300 floating particles randomically placed over the MB of the Lagoon for 2013.



Fig 2 – A) Distribution of the particles released in the MB of the Lagoon and associated bathymetry. B-G) Temporal evolution of the transport of these particles during the simulated year associated with current velocities and directions of currents in the MB. Each image is referent to the last day of the respective month. Colored squares are possible accumulating zones.

To watch the animation of the simulated period access the video using the QR-code or the link below:









### Adsorption behavior of chlorinated phenols on polyethylene in Danube River water

Lončarski Maja, Tubić Aleksandra, Vasiljević Sanja, Agbaba Jasmina.

Microplastics (MPs) and chlorinated phenols (CPs) are ubiquitous contaminants in aquatic ecosystems. Therefore the study of organic pollution adsorption affinity toward different types of microplastics is vital for comprehensive assessment and risk management in the environment. Since microplastics may act as vectors for the transport of contaminants in water matrices main scope of this study was to understand adsorption behaviour of four CPs (4-chlorophenol-4-CP, 2,4dichlorophenol-2,4-DCP, 2,4,6-trichlorophenol-2,4,6-TCP, and pentachlorophenol-PCP) on polyethylene of different origin (powdered standard polyethylene (PEp) and polyethylene isolated from two personal care products (PE<sub>P</sub>CPs1 and PE<sub>P</sub>CPs2)). All experiments were carried out in a Danube river water, and the initial concentration of selected CPs was 100 µg/l. The adsorption of selected chlorinated phenols on microplastic was determined by using kinetic and isotherm studies. The obtained results of the kinetic adsorption study indicated that the adsorption equilibrium between the CPs and polyethylene MPs was established after 24 h of contact time. In all cases, pseudo-second order kinetic model fitted data the best (R2=0.973-0.999) indicating that chemisorption is a dominant adsorption mechanism. A better understanding of the adsorption mechanism for chlorinated phenols on microplastic was determined by using Freundlich and Langmuir adsorption isotherms. Based on the obtained results Langmuir adsorption model fitted data better indicating that adsorption of the chlorinated phenols occurs at a specific site on the microplastics, with no further adsorption occurring at the same site. Maximum adsorption capacities determined by the Langmuir adsorption model also indicated that phisyco-chemical properties of the organic compound have the highest impact on adsorption behaviour in the case of chlorinated phenols toward microplastic. Therefore, the highest adsorption capacity was determined for 2,4-DCP (qmax=218.1 µg/g) and the lowest for PCP (qmax=189.0 µg/g). The results of this study also indicated that polyethylene MPs can serve for the transport of the chlorinated phenols through freshwater bodies.

Keywords : adsorption , chlorinated phenols , freshwater bodies , microplastic

### ADSORPTION BEHAVIOR OF CHLORINATED PHENOLS ON POLYETHYLENE IN DANUBE RIVER WATER

Maja Lončarski<sup>1</sup>, Aleksandra Tubić<sup>1\*</sup>, Sanja Vasiljević<sup>1</sup>, Jasmina Agbaba<sup>1</sup>

<sup>1</sup>University of Novi Sad, Faculty of Sciences, Department of Chemistry, Biochemistry and Environmental Protection, Trg Dositeja Obradovića 3, 21000 Novi Sad, Republic of Serbia

### **INTRODUCTION**

A significant amount of plastic comes into the environment, where during the time, large pieces under physical, chemical and biological processes break down into smaller ones. Plastic components detected in the environment less than 5 mm are named microplastic. Microplastics (MPs) and chlorinated phenols (CPs) are ubiquitous contaminants in aquatic ecosystems.

Since microplastics can act as vectors for the transport of different contaminants in water matrices main scope of this study was to understand adsorption behaviour of four CPs on polyethylene of different origin.

### MATERIAL AND METHODS

Four chlorophenols:

- 4-chlorophenol,
- 2,4-dichlorophenol,
- 2,4,6-trichlorophenol and
- pentachlorophenol

All experiments were carried out in a Danube river water, and the initial concentration of selected CPs was 100  $\mu g/l.$ 

Selected microplastics:



standard substance (PEp)

PE isolated from two

personal care products (PE\_PCPs\_1 and PE\_PCPs\_2)

The adsorption of selected chlorinated phenols on microplastic was determined by using kinetic and isotherm studies. Chlorinated phenols were analyzed by GC with MSD, after acurate sample preparation.

### **RESULTS AND DISCUSION**





The obtained results of the kinetic adsorption study indicated that the adsorption equilibrium between the CPs and polyethylene MPs was established after 24 h of contact time.

### ACKNOWLEDGEMENTS:

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University of Novi Sad, Faculty of Sciences, Department of Chemistry, Biochemistry and Environmental Protection Trg Dositeja Obradovica 3, 21000 Novi Sad, R. Serbia tel - x381 (0/21 485 278 In all cases, pseudo-second order kinetic model fitted data the best (R<sup>2</sup>=0.973-0.999) indicating that chemisorption is a dominant adsorption mechanism.  $P_{PEP} \bullet PE_PCPs_1 \bullet PE_PCPs_2$ 



Figure 2. Plots for the sorption kinetics, based on the pseudo-second order model, of (a) 4- CP; (b) 2,4-DCP; (c) 2,4,6-TCP and (d) PCP on PEp, PE\_PCPs\_1 and PE\_PCPs\_2 particles in Danube water (n = 3, mean value SD) Based on the obtained results Langmuir adsorption model fitted data better indicating that adsorption of the chlorinated phenols occurs at a specific site on the microplastics, with no further adsorption occurring at the same site.



Figure 3. Sorption isotherms of (a) 4- CP; (b) 2,4-DCP; (c) 2,4,6-TCP and (d) PCP on PEp PE\_PCPs\_1 and PE\_PCPs\_2 particles in Danube water (n = 3, mean value SD)

Table 1. Values of the Freundlich and Langmuir model parameters for the sorption of 4-CP, 2,4-DCP, 2,4,6-TCP and PCP on PEp, PE\_PCPs\_1 and PE\_PCPs\_2

	Material	Freundclih model			Langmuir model		
Compoud		K <sub>F</sub> (µg/g)/(µg/l) <sup>n</sup>	n <sub>F</sub>	R <sup>2</sup>	К <sub>L</sub> (I/µg)	q <sub>max</sub> (µg/g)	R <sup>2</sup>
4-CP	PEp_D	8.50	0.42	0.944	0.057	62.81	0.996
	PE_PCPs_1_D	18.9	0.49	0.948	0.069	153.3	0.988
	PE_PCPs_2_D	15.8	0.49	0.948	0.077	124.7	0.932
2.4-DCP	PEp_D	23.8	0.47	0.929	0.064	218.1	0.980
	PE_PCPs_1_D	30.9	0.42	0.946	0.230	138.8	0.902
	PE_PCPs_2_D	29.5	0.43	0.913	0.130	152.4	0.949
2,4,6-TCP	PEp_D	22.0	0.43	0.819	0.080	138.1	0.901
	PE_PCPs_1_D	21.2	0.44	0.830	0.069	148.3	0.906
	PE_PCPs_2_D	11.1	0.60	0.928	0.030	191.7	0.938
PCP	PEp_D	28.8	0.35	0.863	0.148	119.1	0.969
	PE_PCPs_1_D	19.5	0.53	0.990	0.062	181.5	0.996
	PE PCPs 2 D	16.3	0.56	0.983	0.047	189.0	0 998

Maximum adsorption capacities determined by the Langmuir adsorption model also indicated that phisyco-chemical properties of the organic compound have the highest impact on adsorption behaviour in the case of chlorinated phenols toward microplastic.

### CONCLUSION

The highest adsorption capacity was determined for 2,4-DCP ( $q_{max}$ =218.1µg/g) and the lowest for 4-CP ( $q_{max}$ =62.81 µg/g). The results of this study also indicated that polyethylene MPs can serve for the transport of the chlorinated phenols through freshwater bodies.

Day 3, Wednesday 25th. November 2020								
9h-10h	25.1_0	25.1_Me	25.1_Ma					
10h-10h15	25.1_Gaia: 3 sessions brief							
10h30-11h30	25.2_0	25.2_Me	25.2_Ma					
11h30-11h15	25.2_Gaia: 3 sessions brief							
12h-13h	25.3_0	25.3_Me	25.3_Ma					
13h-13h15	25.3_Gaia: 3 sessions brief							
13h15-13h45	Poster.25.4_O	Poster.25.4_Me	Poster.25.4_Ma					
14h-15h	25.5_O	25.5_Me	25.5_Ma					
15h-15h15	25.5_Gaia: 3 sessions brief							
15h30-16h30	25.6_O	25.6_Me	25.6_Ma					
16h30-16h45	25.6_Gaia: 3 sessions brief							
17h-18h	25.7_0	25.7_Me	25.7_Ma					
18h-18h15	25.7_Gaia: 3 sessions brief							
18h30-19h27	The Marine Strategy Framework Directive perspective: a panel conversation							
19h30-20h	Poster.25.8_O	Poster.25.8_Me	Poster.25.8_Ma					
20h-20h30	Poster.25.9_O	Poster.25.9_Me	Poster.25.9_Ma					
20h30-21h00	Poster.25.10_O	Poster.25.10_Me	25.10_Ma					

### Day 3/5, Wednesday 25th November 2020

### Session 25.1\_O. Chaired by Maria Cristina Fossi, Siena

**Proof of the invisible ones: microplastic burden in marine mammals from the German Northand Baltic Seas** *Philipp Carolin, Unger Bianca, Fischer Elke, Siebert Ursula.* Paper number 324305

### Are bioplastics safe for the environment? Degradation of biodegradable microplastics results in secondary nanoplastics that affect aquatic microorganisms

Tamayo Miguel, Martín-Betancort Keila, Pulido Reyes Gerardo, Gonzalez-Pleiter Miguel, Martínez-Campos Sergio, Verdu-Fillola Irene, Edo Carlos, Rosal Roberto, Fernandez-Piñas Francisca. Paper number 333536

**The abundance and distribution of plastics in surface water of the Caribbean** *Courtene-Jones Winnie, Penn Emily, Thompson Richard.* Paper number 334015

### **Proof of the invisible ones: microplastic burden in marine mammals from the German North- and Baltic Seas**

Philipp Carolin, Unger Bianca, Fischer Elke, Siebert Ursula.

Microplastics are known to be ubiquitous. It is thus not surprising that laboratory studies and field experiments demonstrated the ingestion, accumulation, cell migration and the egestion in a variety of different invertebrate species. Marine mammals as top predators are thus known to accumulate contaminants and pollutants through the food web. However, information on the occurrences and effects of microplastics in marine mammals are still scarce in the north-western Atlantic region. For revealing potential burdens, the actual presence of microplastics has to be proven. This is the first study, dealing with all three species regularly occurring in German North- and Baltic Seas: harbour porpoises (Phocoena phocoena), harbour seals (Phoca vitulina) and grev seals (Halichoerus grypus) and documents the quantity and quality of microplastics in the gastrointestinal tract. Intestine samples, whole stomachs, and faeces samples collected between 2014 – 2019 were analysed after a new established protocol of sample handling. The low share of secondary pollution and the low costs for isolating microplastic particles are significant benefits of this protocol. First preliminary results already show the identification of PE, PET, PP and ethylene-vinyl acetate copolymers by µRaman spectroscopy, and a higher share of particles compared to fibres. Up to now, the highest share of fragments (n = 28) and fibres (n= 16) was found in harbour porpoises. Whereas, the minimum of five fragments and no fibres were found in a sample of a harbour seal. The sample handling and proceeding are transferable to other mammal species, and allows new insights in predator-prey relationships. The first findings already indicate that microplastic in marine mammals from German waters is a health-relevant topic and further studies are needed.

Keywords : Baltic Sea , Grey seal , Harbour porpoise , Harbour seal , Marine Mammals , Microplastic , North Sea , Pollution Burden ,  $\mu$ Raman spectroscopy

### Are bioplastics safe for the environment? Degradation of biodegradable microplastics results in secondary nanoplastics that affect aquatic microorganisms

Tamayo Miguel, Martín-Betancort Keila, Pulido Reyes Gerardo, Gonzalez-Pleiter Miguel, Martínez-Campos Sergio, Verdu-Fillola Irene, Edo Carlos, Rosal Roberto, Fernandez-Piñas Francisca.

There is an increasing concern about the potential occurrence of nanoplastics as a result of degradation processes suffered by microplastics in the environment. Biodegradable plastics are emerging as an alternative to conventional plastics mainly because they undergo rapid biotic and/or abiotic hydrolytic degradation. In this work we investigated the most relevant physicochemical features and the biological effects of nanoplastics released from microplastics, made of the biodegradable polymer polycaprolactone (PCL), that underwent artificial abiotic degradation up to one year under environmentally representative conditions. The physicochemical characterization of the nanoplastics obtained from the abiotic degradation of microplastics was performed by DLS (size distribution), ELS (surface net charge or  $\zeta$ -potential), NTA (number of particles per unit volume), FTIR (chemical bonds), MALDI TOFF (polymer chain length distribution), DRX and DSC (crystallinity) and SEM (morphology). Base on the outcomes of these techniques, a mathematical model has been developed in order to estimate the potential occurrence of micro- nano-plastics and free oligomers on the aquatic environment. The ecotoxicological effects of released nanoplastics after two weeks in aqueous suspension was also assessed using two cyanobacteria: the filamentous cyanobacterium Anabaena sp. PCC7120 and the unicellular Synechococcus sp PCC7942. After abiotic incubation of the microplastics in aqueous suspension, the supernatant containing released nanoplastics, resulted to be toxic towards both microorganisms. Nanoplastics induced a significant decrease in the growth as well as several physiological alterations, studied by flow cytometry, of the two photosynthetic organisms. However, after removing the nanoplastics by ultrafiltration, only a mild toxicity remained, affecting few physiological parameters which are probably more sensitive to the leachates. In conclusion, nanoplastics released from biodegradable microplastics exhibited toxicity towards aquatic photosynthetic organisms. Our findings indicated that biodegradable plastics may not be as harmless as previously suggested.

Keywords : biodegradable , Cyanobacteria , degradation , ecotoxicity , flow cytometry , mathematical modeling , polycaprolactone , secondary nanoplastic

**The abundance and distribution of plastics in surface water of the Caribbean** Courtene-Jones Winnie, Penn Emily, Thompson Richard.

Plastic pollution is considered one of the most serious threats to sustainable use of marine and coastal resources of the Caribbean. To date the majority of studies in this region have focused on beached litter with research into the abundance and distribution of floating plastics severely lacking. As part of 'eXXpedition Round the World', manta trawls were conducted during November - December 2019 (n = 20), to sample surface (micro)plastics within the Caribbean Sea: around the Lesser Antilles (Antigua, Bonaire and Aruba) and the San Blas islands. Samples were sieved into three size classes (335µm – 0.99mm, 1.00mm – 4.74mm, ¿ 4.75mm), categorised according to morphology and polymer composition as determined by FTIR analysis. Plastics  $\geq$  335µm were identified in all except two of the trawls, with fragments dominating at each location. Concentrations ranged between 0 – 0.0051 particles m<sup>-3</sup>, with the highest accumulation observed in coastal water of the San Blas islands (486 plastics), where foam and pellets were also found in relatively high abundance. Across all sites, 18 polymer types were identified, comprising both negatively and positively buoyant plastics. Polyethylene and polypropylene dominated with other polymers displaying a more heterogeneous distribution. Data were analysed to identify any geographical differences between polymer composition, size and morphology, and plastic abundance was explored in relation to environmental variables to evaluate whether these factors may influence the distribution of surface plastics. The data presented are some of the first baseline estimates of floating plastics in these regions of the Caribbean. Despite regional ocean dynamics, which are hypothesised to transport and aggregate plastics in the Caribbean Sea, the values we report are comparable to similar studies conducted in the wider North Atlantic Ocean. These results are the first to be presented from a larger, global dataset; the fieldwork for which is currently ongoing.

Keywords : Caribbean , distribution , fragments , microplastic , surface water

### Session 25.1\_Me. Chaired by Pennie Lindeque and Rachel Coppock, Plymouth

Potential bioavailability of inorganic compounds from Tire and Road Wear Particles to fish in simulated gastric and intestinal fluids.

Masset Thibault, Ferrari Benoit, Oldham Dean, Dudefoi William, Minghetti Matteo, Breider Florian. Paper number 334177

### Characterisation of tire and tire wear with Py-GC/MS – Identification and quantitative application to complex environmental samples

Goßmann Isabel, Halbach Maurits, Scholz-Böttcher Barbara. Paper number 334214

### Multi-marker DNA reveals the diversity of plastic-associated organisms in the Western South Atlantic and Austral Oceans

Lacerda Ana Luzia, Kessler Felipe, Secchi Eduardo, Proietti Maira, Taylor Joe Daniel. Paper number 334545

### Potential bioavailability of inorganic compounds from Tire and Road Wear Particles to fish in simulated gastric and intestinal fluids

Masset Thibault, Ferrari Benoit, Oldham Dean, Dudefoi William, Minghetti Matteo, Breider Florian.

The potential impact of Tire and Road Wear Particles (TRWP) on aquatic organisms has recently gained attention since the occurrence of TRWP in the aquatic environment has been demonstrated in surface water and sediments of numerous regions. However, data regarding the bioavailability and toxicity of contaminants associated with these particles are still lacking. Several metals enter in the composition of tires including Zinc, used as a catalyst during the vulcanization process of rubber. This study aimed (i) to characterize the elemental composition of Cryogenically Milled Tire Tread (CMTT) and TRWP and (ii) to assess the solubilization potential of inorganic contaminants from CMTT and TRWP in Simulated Gastric Fluids (SFG) and Simulated Intestinal Fluids (SIF) designed to mimic Oncorhynchus mykiss gut conditions. Our results show an important difference in the elemental composition of CMTT and TRWP due to the input of elements such as Ti, Cr, Mn, Sr, Ba and Pb from the road constituent to the TRWP. In vitro digestion experiments of both CMTT and TRWP showed that solubilization of all metals were enhanced by up to 10-fold in simulated gut fluids compared to solubilization in phosphate buffer solution. Solubilization kinetics to the SGF and SIF were biphasic with a higher solubilization rate of elements within the first hour and a slower solubilization rate up to 26h. For most metals, solubilization rates were higher for TRWP than for CMTT. After digestion time of 26h, from 1% (Co) to 10% (Zn) of total metal in CMTT were solubilized in the gut fluids and the concentration of metals were about  $1 - 100 \mu g/L$  except for Zn with 7000 µg/L. Our results emphasise the importance of pH, bile surfactants and organic matter in the solubilization of metals from TRWP and brings new insights on the bioavailability of inorganic pollutants from TRWP.

Keywords : bioavailability , heavy metals , organic pollutants , simulated gut fluids , Tire particles

### Characterisation of tire and tire wear with Py-GC/MS – Identification and quantitative application to complex environmental samples

Goßmann Isabel, Halbach Maurits, Scholz-Böttcher Barbara.

Tire wear particles (TWP) are assumed to be the most dominant source of microplastics (MP) ending up in the marine environment. The constantly released TWP in admixture with road particles are considered be a threat to ecosystems [1]. The identification potential of TWP pyrolysis products and their respective characteristic fragment ions were proven based on earlier publications [2]. The indicator fragments were included to the Py-GC/MS method for simultaneous mass quantitative analysis for various types of MP established by Fischer & Scholz-Böttcher [3]. Different types of truck and car tire treads were characterized, accordingly. An average truck and car tire was defined each and used for further quantification. The approach was applied to different complex environmental samples (road dust, sea salts, blue mussels, and sediments). The resulting TWP mass loads were compared, differentiated into car and truck impact, and related to thermoplastic MP share. Resulting data show an expected variability in rubber composition of tires regarding to their designated use. Truck tires consist almost exclusively of natural rubber (NR). Car tires show greater quantities of synthetic rubber such as styrene-butadiene rubber (SBR). TWP were present in all analysed compartments except the blue mussels. Generally, car tire wear concentrations exceeded truck tire wear by far, proving the estimated ratio of passenger car tires to truck tires of 10 to 1 based on production and usage data [4] experimentally. Although TWP mass loads surpassed the thermoplastic MP share close to the TWP source, thermoplastic MP clearly dominated in the aquatic samples indicating a poor long-distance transport potential of TWP. 1. Wagner, S. et al., 2018. Water Research; 139; 83-100. 2. Eisentraut, P. et al., 2018. Environmental Science & Technology Letters; 5; 608-613. 3. Fischer, M., Scholz-Böttcher, B.M., 2019. Analytical Methods; 11; 2489-2497. 4. Bertling et al., 2018. Frauenhofer UMSICHT.

Keywords : Microplastics , Py GC/MS , Truck and passenger car tire wear , TWP

### Multi-marker DNA reveals the diversity of plastic-associated organisms in the Western South Atlantic and Austral Oceans

Lacerda Ana Luzia, Kessler Felipe, Secchi Eduardo, Proietti Maira, Taylor Joe Daniel.

Floating plastics in aquatic systems act as artificial substrates for a large taxonomic diversity of micro and macro-organisms. As plastics can last for many years in the oceans and be transported between regions, they have the potential to increase dispersal of these species; however, the diversity and ecological function of epiplastic communities are not known for many regions. Our study aimed to identify the diversity of plastic-associated organisms in the Western South Atlantic (WSA) and around the Antarctic Peninsula (AP), and show how plastic characteristics and geographic location may influence community composition. Sampling was conducted in the open ocean using manta net (330µm mesh). The size, color and chemical composition of sampled plastics were characterized. DNA was extracted from the plastics biofilm and a metabarcoding approach targeting both prokaryotes and eukaryotes (16S, 18S and ITS2) was used to characterize the diversity of life associated with plastics. Our data showed a range of different prokaryote (Bacteria and Archaea) and eukaryotes (e.g. Protists, Fungi, Metazoa, Macroalgae) associated with plastics in both regions; in general, plastic characteristics did not influence community diversity, even within regions, contrary to what has been suggested by some colonization studies. We found microbial groups in our oceanic samples that have been previously described, under lab conditions, as plastic biodegraders (Aspergillus and Cladosporium species). We also identified groups that had not been described associated with plastics, and for the first time documented the diversity of plastic-associated fungi in these regions (Lacerda et al., 2020). Our work shows that epiplastic communities within these regions are highly diverse. This work has increased our knowledge about plastic-associated organisms in Southern Hemisphere. It is essential that we continue to monitor these epiplastic communities to better characterize the potential impacts of plastics and their associated biota on marine ecosystems, including bioinvasions.

Keywords : ecology , metabarcoding , Plastic pollution , plastisphere

### Session 25.1\_Ma. Chaired by Bart Koelmans, Wageningen

Nanopolystyrene particles impact fish cell lines González-Fernández Carmen, Esteban M. Angeles, Cuesta Alberto. Paper number 333378

**Occurrence and effects of microplastics in tissues of coastal wildlife** *Haave Marte, Olsen Anne-Berit, Schönheit Jürgen, Nilsen Hanne, Gomiero Alessio.* Paper number 334465

### Label-free nano- and microplastics detection and identification *in vivo* using dark-field hyperspectral microscopy

*Fakhrullin Rawil, Nigamatzyanova Läysän.* Paper number 334525

### Rapid, automated analysis of microplastics using laser direct infrared imaging and spectroscopy

*Kerstan Andreas*. Paper number 334644

### Nanopolystyrene particles impact fish cell lines

González-Fernández Carmen, Esteban M. Angeles, Cuesta Alberto.

Nowadays, nanoplastics (NPs) are the most abundant nanomaterials on the market. This generates, in addition to the indirect production derived from the mechanical, physical or biological degradation of plastic debris, a direct contribution of NPs to the oceans. Determining the effects that the presence of NPs can have on the environment and the organisms that inhabit it is of vital importance since, due to their small size, they are capable of crossing cell membranes and, depending on their physicalchemical characteristics, be accumulated in organisms. The aim of the present work is to evaluate the potential effect of different functionalized NPs on the gilthead seabream Sparus aurata new cell line derived from the brain (SaB-1). The seabream is one of the most commercial fish species of the Mediterranean area and thus it is of great economic interest. SaB-1 cells were exposed to different 50 nm polystyrene (PS) NP types (PS-Plain, PS-COOH, PS-NH2). The LC50 (12 µg/mL) concentration was used to evaluate the impact of NPs for 24 hours of exposure. A battery of gene markers of oxidative stress (mta, sod, cat, gr and gst) and apoptosis (bcl2) was studied through qPCR to identify the effect of NP on cells. Preliminary results evidenced significant impacts of NPs exposure on cell viability, which differed depending on the NP charge, being PS-NH2 that promoting the highest cell mortality. Apoptotic processes were strongly activated by NPs. A down-regulation of bcl2 transcription was observed upon exposure to PS-NH2 and PS-Plain. Conversely, the transcription of mta was up-regulated by all NPs (PS-Plain; PS-NH2; PS-COOH) while gr was significantly increased by PS-COOH and decreased by PS-Plain. The present work evidence that the effect of NP is strongly influenced by particle's type and more research on fate and impact of nanoplastics at cellular level is needed.

Keywords : cell lines , fish , nanoplastics , Polystyrene , seabream

### **Occurrence and effects of microplastics in tissues of coastal wildlife**

Haave Marte, Olsen Anne-Berit, Schönheit Jürgen, Nilsen Hanne, Gomiero Alessio.

Microplastic is omnipresent in biota, and concerns of human exposure through food. Investigations of wildlife relevant for human consumption and the effects in animals exposed to plastics is warranted. We investigated the concentrations of microplastics (MP) in tissues of fish, seabirds and marine mammals from a plastic polluted area near Bergen, Norway. A standardized autopsy included evaluation of condition, bacteriological and histopathological analyzes. Tissues were analyzed for MP (¿10µm) by pyrolysis Gas-Chromatography/Mass-Spectrometry and inspected by polarized-light -microscopy. We analyzed samples of the GI tract, liver and muscle/fillet from three flatfish, three cod, three seabirds, three otters and one seal, kidneys from seabirds, otters and the seal, gills from the fishes. No large plastic items were observed in the gastrointestinal tracts. Eight of thirteen animals had MP in one or several tissues. MP was found in intestine (5), stomach (4), liver(3), muscle(3). No MP was found in the seal, and only in the stomach wall of one otter. In seabirds MP was found in the intestine, stomach and liver, but not muscle. MP was most common in cod (two of three), but rare in flatfish. The highest MP concentration found was 3.4 µg/g wet weight in cod liver. Three of nine polymers were found: Polyvinylchloride¿polystyrene¿¿polyethylene terephthalate. Other investigated polymers were below the Limit of Quantification (LOQ). In four parallel analyses of cod muscle and two analyses of liver in cod, MP was quantified in one parallel per individual. No MP was observed by microscopy. The results show levels around the current LO. Replicates indicate uneven tissue distribution. No adverse effects were observed related to the presence of MP. The sample size was small, and conclusions cannot be drawn regarding risks. Controls showed very low MP. Further studies are needed elucidate the current wildlife and human exposure.

Keywords : food , histopathology , Microplastic , organ , Risk Assessment , tissue , uptake , web , Wildlife

### Label-free nano- and microplastics detection and identification *in vivo* using dark-field hyperspectral microscopy

Fakhrullin Rawil, Nigamatzyanova Läysän.

Microscale and nanoscale polymer particle pollution is among most serious ecological threats, severely affecting environments, live organisms and human well-being. To tackle nano- and microplastics pollution one needs a powerful methodology to visualise and identify small polymer particles in cells, tissues, organisms and environmental specimens (e.g. water, soil, sediments, etc). We have developed a novel methodology to visualise and identify synthetic polymer nanoscale (down to 100 nm) and microscale particles utilising hyperspectral dark-field microscopy in 400-1000 nm wavelength range (visible-near infrared). To demonstrate the feasibility of our technique we have used polystyrene particles with diameters between 100 nm  $- 1 \mu m$ , polymethacrylate 1  $\mu m$  and melamine formaldehyde 2 µm microspheres. These nano- and micro particles in pure and mixed suspensions were effectively imaged and identified chemically using the libraries of spectral signatures and image-assisted analysis. We also succeeded in imaging and hyperspectral identification of pure and mixed nano- and microplastics in Caenorhabditis elegans nematodes in vivo, to demonstrate the ingestion and distribution of nano- and microplastics in tissues. We found that dark-field hyperspectral microscopy can be successfully applied for differentiating between chemically-different microplastics confined within live invertebrates. This simple optical technology allows for quantitative identification of microplastics taken up by live nematodes. This label-free non-destructive methodology will find applications in environmental nano- and microplastics detection and quantification, studies of microplastics biodistribution in tissues and organs and nanotoxicology. The authors acknowledge funding by Russian Federation presidential grant MD-2153.2020.3.

Keywords : Caenorhabditis elegans , Dark , field hyperspectral microscopy , Microplastics , Minimum detection size , Polystyrene

### Rapid, automated analysis of microplastics using laser direct infrared imaging and spectroscopy

Kerstan Andreas.

Environmental researchers wish to measure the size, shape, and chemical identity of every plastic particle in a sample. Because smaller particles are thought to be the most biologically relevant, this analysis must extend to particles on the micron scale. Unfortunately, traditional techniques such as visual inspection are slow, manually intensive, and prone to operator bias. As a result, investigators have recently turned to chemically specific vibrational spectroscopy, which can be used in a microscope format for particle analysis at greater speeds. Spectral microscopes acquire a spectrum guided by a visible-light image to determine a particle's chemical identity along with its size and shape. Still, these instruments have drawbacks. Raman microscopes struggle to identify fluorescent particles, while array-based FTIR (Fourier transform infrared) microscopes generate a large number of spectra which are redundant or taken in the empty space between particles. Finally, the massive datasets generated by these microscopes introduce processing and storage challenges. Since IR spectroscopy is a useful method for microplastics analysis, alternative methods of generating and focusing IR light on a sample may overcome some of these difficulties with existing techniques. A Quantum Cascade Laser (OCL) is a tunable semiconductor laser than generates light in the mid-Infrared regions commonly known as the fingerprint region. Such a light source allows tight focusing of the bright infrared light at the precise center of each particle and this can facilitate rapid and accurate identification of micron-scale particles. In this paper, we present results of studies using an Infrared spectrometer utilizing a Quantum Cascade Laser for the characterization and quantitation of microplastics.

Keywords : Automation , Laser , LDIR , Microplastics , QCL , Speed

### Session 25.2\_O. Chaired by Sonja Oberbeckmann, Rostock

**Microplastics as vector for microbial communities** *Oberbeckmann Sonja, Labrenz Matthias.* Paper number 331903

### Short-term bacterial colonization experiment on biodegradable and non-biodegradable microplastics incubated *in situ* into wastewater treatment plant effluents

Martínez-Campos Sergio, Gonzalez-Pleiter Miguel, Fernandez-Piñas Francisca, Rosal Roberto, Leganés Francisco. Paper number 334136

### Meta-analysis based global survey of the microbial life associated with microplastics in the aquatic environment

*Keating Ciara, Trego Anna C., Chen Bozhen, Li Keda, Ijaz Umer Z., Gauchotte-Lindsay Caroline.* Paper number 334438

**The influence of plastic surfaces on the conditioning film and subsequent biofilm succession** *Rummel Christoph, Lechtenfeld Oliver, Kallies René, Benke Annegret, Herzsprung Peter, Rynek Robby, Wagner Stephan, Potthoff Annegret, Jahnke Annika, Schmitt-Jansen Mechthild.* Paper number 334684

### Microplastics as vector for microbial communities

Oberbeckmann Sonja, Labrenz Matthias.

Microorganisms play crucial roles in all biogeochemical processes, and shifting microbial communities have potential implications for the marine ecosystem. Therefore, the impact of the ubiquitous plastic pollution on composition and function of marine microbial communities represents a highly relevant research topic from an ecological, environmental, and microbiological perspective. Previous studies revealed that plastics in the oceans are colonized by diverse pro- and eukaryotic communities, but many open questions, in particular on functional level, remain. This session will focus on microplastic microbiomes from different parts of the world, their metabolic profiles, and how spatial and seasonal factors play a role in shaping these plastic biofilms. We will further look into topics highly discussed in this field of research, namely whether microplastics carry potential harmful microorganisms and traits, and whether marine microorganisms can contribute to biodegradation of plastics. Overall, we aim at a better understanding of the effect plastic pollution can have on microbial ecology and ecosystem functioning.

Keywords : biofilm , marine ecosystem , microorganisms , microplastics

### Short-term bacterial colonization experiment on biodegradable and nonbiodegradable microplastics incubated *in situ* into wastewater treatment plant effluents

Martínez-Campos Sergio, Gonzalez-Pleiter Miguel, Fernandez-Piñas Francisca, Rosal Roberto, Leganés Francisco.

Microplastics (MPs) constitute a global emerging pollution whose ecological effects are not fully understood. Recent studies performed in aquatic ecosystems show that the plastic particles can be colonized by microorganisms (the plastisphere) being the surrounding environment the key factor for the MP-attached microbial community. Nonetheless, the early colonization of MPs in wastewater treatment plant (WWTP) effluents has not been evaluated yet, despite that mature biofilms depend on early colonizers. Also, MPs might be an important hotspot of bacterial pathogens and antibiotic resistance genes (ARGs). Here, we explored the diversity and community composition of bacteria attached on seven types of MPs (including biodegradable and non-biodegradable polymers) incubated in situ into two different WWTP effluents for 48 hours using Illumina MiSeq sequencing of the 16S rRNA. Also, we checked the relative abundance of the ARGs (sull and tetM) using qPCR in comparison with the surrounding water to check a possible increase of ARGs in the MP-attached community. The diversity in the MP-associated microbial community was significatively higher in comparison with water free-living bacteria. RDA analysis confirmed that the sampling site explained the major changes in community diversity. Hydrophobicity of the polymer also affected the diversity between samples but to a lesser extent. A MPs-core microbiome was identified which included some genera associated with the degradation of plastics such as Acidovorax, Aquabacterium, Pseudomonas and Variovorax. sull and tetM genes were detected in the two WWTPs effluents but only sull was clearly concentrated in the MPs. In conclusion, our results highlight the relevance of the early attachment phase in the development of bacterial biofilms on different types of MP polymers and provide novel information about the development of biofilms on MPs and the role that different types of polymers might have facilitating the attachment of specific bacteria, some of which might carry ARGs.

Keywords : Antibiotic Resistance Genes (ARGs) , bacteria , early colonization , metabarcoding , microplastics , WWTP

### Meta-analysis based global survey of the microbial life associated with microplastics in the aquatic environment

Keating Ciara, Trego Anna C., Chen Bozhen, Li Keda, Ijaz Umer Z., Gauchotte-Lindsay Caroline.

Background & Objectives Since the discovery of microplastics in the 1970's researchers have sought to understand the persistence of these particles both in the environment and in living animals resulting in 4.211 peer-reviewed articles. 242 of these published publications were associated with phrases relating to microbial life. Microorganisms are infamous for their potential for degradation of a variety of natural and synthetic compounds. Thus, the microorganisms colonising microplastics are of great interest. However, research also suggests that microplastics can be a novel breeding ground for pathogens and antibiotic resistance genes which are then easily carried through aquatic environments. To this end, the aim of our work was to combine and re-analyse these previously published datasets to provide a global perspective of the microbial communities associated with microplastics from i) seawater and freshwater aquatic systems ii) lab-based incubations, and including iii) a variety of microplastic materials (e.g. polystyrene [PS] and polyethylene [PE]). Methods Briefly, we searched for published articles containing the term 'microplastics' and 16S rRNA gene amplicon sequencing using the Illumina Miseq platform. Where the authors had made the data publicly available (30% of relevant published work) the data was downloaded (800 samples) and processed using QIIME2 following a meta-analysis workflow as described in Keating et al (2020). Results & Conclusions Microbial species diversity was lowest in the samples associated with PS and PE microplastic materials. In general, samples clustered according to geographical location and sample type. Microbial species classed as lottery winners were identified (i.e. a winning colonising species). Winner behavior shifted according to sample type with Flavobacteriaceae and Saprospiraceae microbial families showing high winner prevalence in samples associated with specific microplastics (PE, PS). These were also the sole species characterised as part of the core microbiome (75% prevalence) of microplastic colonisers.

Keywords : 16S rRNA gene , Analysis , Aquatic , Freshwater , Illumina Miseq , Meta , Microbial Community , Microbiome , Microplastics , Seawater
# The influence of plastic surfaces on the conditioning film and subsequent biofilm succession

Rummel Christoph, Lechtenfeld Oliver, Kallies René, Benke Annegret, Herzsprung Peter, Rynek Robby, Wagner Stephan, Potthoff Annegret, Jahnke Annika, Schmitt-Jansen Mechthild.

It is still under debate if and how the 'plastisphere', a plastic-specific microbial community, can emerge and which the underlying processes are. The initial conditioning film of adsorbed dissolved organic matter (DOM) is thought to play a key role for microbial pioneer attachment and subsequent early biofilm formation. In this study, we tested the hypothesis that DOM sorbs selectively to substrates that display different surface properties. Further, we tested whether subsequent early microbial attachment is governed in a substrate-dependent manner. We investigated the adsorption behavior of DOM to polyethylene terephthalate (PET), polystyrene (PS), and glass as a reference material. The organic matter (OM) composition was characterized by Fourier-transform ion cyclotron mass spectrometry and compared to the DOM in the original incubation water. Only a fraction of the original DOM adsorbed to the substrates. We identified major differences in the molecular OM composition between the substrates which were additionally modified by a weathering treatment. The biofilm community was investigated after 24 h and 72 h of incubation by 16S and 18S rRNA gene amplicon sequencing. Early biofilm communities showed a clear timedependency, however, we could identify a minimal but detectable substrate-specificity for biofilm attachment after 24 h. Conclusively, the adsorbed OM layer reflects the materials' surface properties to some extent and passes on these surface properites to the OM-water interface which in turn may govern microbial colonization.

Keywords : Conditioning film , corona , Dissolved organic matter (DOM) , Eco , FT , ICR MS , Microbiome , Microorganisms , Microplastic , Plastic pollution , Surface properties , weathering

#### Session 25.2\_Me. Chaired by Johnny Gasperi, Bouguenais

#### Fluxes of Plastic Debris to a choked coastal lagoon: a top-down approach based on socioeconomic data from south Brazil

Santos Itele E. Dos, Fernandes Elisa H., Pinho Grasiela L. L., Abdallah Patrízia R. Paper number 334413

**Spatial distribution of microplastics in the riverine waters of southwest India** *Amrutha K., Kumar Warrier Anish.* Paper number 334419

# **PLASTIC0PYR PROJECT.** Guidelines for monitoring plastic litter in Mountain riverine systems: from macro- to microplastic sizes

Margenat Henar, Guasch Elena, Le Roux Gaël, Martí Eugènia, Gacia Esperança, Hansson Sophia, Grioche Alain, Cabrera María, Martínez Mònica, Lopez Manel, Vila Anna, Romaní Anna. Paper number 334429

#### Microplastics in stormwater runoff and stormwater treating systems

*Adyel Tanveer M.* Paper number 334455

# Fluxes of Plastic Debris to a choked coastal lagoon: a top-down approach based on socio-economic data from south Brazil

Santos Itele E. Dos, Fernandes Elisa H., Pinho Grasiela L. L., Abdallah Patrízia R.

Plastic production has increased exponentially worldwide and therefore, the amount of mismanaged plastic that ends up in the marine environment also increased. To better understand how this pollutant reaches marine ecosystems, it is important to study its sources, pathways and ultimately estimate the amounts of plastics reaching the environment. However, this could be a difficult task to achieve. In order to estimate fluxes of plastic debris in coastal environments, two research lines co-exist, one based on modeling inputs and one based on field measurements. In this study, the amount of plastics reaching Patos Lagoon, was estimated based on modeling the inputs of plastic fluxes using data of national production, consumption, and waste management, between the years of 2010 to 2017. The main types of plastic resins being produced in the Lagoon's area were identified as Polyethylene (PE), Polypropylene (PP) and Polyvinyl Chloride (PVC). The amount of plastic waste in the Lagoon basin was in the range of 144.76 Kton to 261.29 Kton per year. The main source of plastic pollution identified was textiles/clothing and food-related activities. Using well-known percentages of waste conversion to marine debris, it was estimated that the amount of plastic debris entering Patos Lagoon ranges from 21.67 Kton to 108.76 Kton per vear. This means that each person living inside the lagoon basin, would be producing from 6.54 grams/person/day to 32.82 grams/person/day of plastic debris. Furthermore, results presented in this study are comparable to the per capita amount of plastic debris produced worldwide, which placed Patos Lagoon in 16° (10°) when considering the minimum (maximum) amount reported, surpassing countries like the United States, India, Australia and others.

Keywords : Brazil , Coastal Lagoon , Marine Debris , Production , Quantification , Waste

# **Spatial distribution of microplastics in the riverine waters of southwest India** Amrutha K., Kumar Warrier Anish.

Plastic pieces ; 5 mm in size are termed as microplastics and is an emerging pollutant. Microplastics were reported from all types of environments. The ingestion of microplastics by the smaller aquatic organisms mistaking them as food, and their subsequent transfer to higher trophic level organisms through food chain and their ability to adsorb heavy metals and pollutants, make them a global urgent socio-economic problem. River act as an important carrier of microplastics from the continents to the oceans. In this study, we have investigated the levels of microplastic pollution (5mm - 0.3mm) for an Indian river, namely the Netravathi River. Surface water samples were collected from 14 locations right from its highland to its estuary during the monsoon season (June 2019). The extracted microplastics were visually identified, quantified and categorized using a stereo-zoom microscope, which is further validated by FTIR-ATR analysis. The study reveals the presence of microplastics in all the samples with a mean numerical abundance of 282 particles/m3. The high abundance of microplastics were obtained from the estuary region (NW14; 2328 particles/m3) and low abundance from the highland (NW1; 56 particles/m3). The downstream of the Netravathi River flows through relatively higher populated urban areas, and hence, a relatively higher abundance of microplastics is observed in the downstream sites. Fibres (51.59%) were the most abundant category of microplastics present in the samples followed by films (34.92%), fragments (8.13%), foams (5.16%) and pellets (0.20%). Polyethylene dominated the polymer composition in the samples followed polyethylene terephthalate, polypropylene and polyvinylchloride. Mismanaged solid waste and lack of effective wastewater treatment facilities are the main reasons for the presence of microplastic materials in the river. Source-to-sink characterization of microplastics from the river concludes that population density and anthropogenic activities play a major role in the distribution and abundance of microplastics.

Keywords : fibres , India , microplastic pollution , Netravathi River , population density , sink , source

# **PLASTIC0PYR PROJECT.** Guidelines for monitoring plastic litter in Mountain riverine systems: from macro- to microplastic sizes

Margenat Henar, Guasch Elena, Le Roux Gaël, Martí Eugènia, Gacia Esperança, Hansson Sophia, Grioche Alain, Cabrera María, Martínez Mònica, Lopez Manel, Vila Anna, Romaní Anna.

Plastic pollution is a worldwide environmental issue, which affects all natural habitats via the global network of land, rivers, lakes, seas and oceans and atmospheric transport and deposition. Although the sources, size, fluxes, behaviour and effects of plastic pollution in freshwater ecosystems are poorly quantified, it is generally accepted that once present in the aquatic system, plastic particles contaminate shoreline habitats, the water column and the river benthos, with the highest concentration measured in the latter. Recent works indicate that rivers may function as (temporary) sinks for land-based plastic pollution and a potential source of its future remobilization. PLASTICOPYR project responds to the challenge of making tourism compatible with the conservation and improvement of mountain freshwater ecosystems. The general objective of the project is to propose solutions to reduce in a sustainable way the accumulation of plastics in the mountain ecosystems and their subsequent transport to the sea. The project is developing unique protocols to be applied in 3-4 natural areas located in the Pyrenees of Catalonia, Andorra and France that show different levels of human impact (ski resorts, national parks...). Different river environment compartments are sampled to quantify the abundance of plastic particles in the riparian area, water column, benthic area (sediments and biofilm), fishes and atmosphere (rain and dry deposition). With the results, we are going to be able to identify the most impacted areas, the transport and storage areas across the headstream and observe if exists a link between microplastic abundance and the health of the river system. Our efforts will help to improve the plastic waste management by involving different stakeholders (scientific, governments, citizens...).

Keywords : field methodology , management , plastic dynamic , plastic waste

#### Microplastics in stormwater runoff and stormwater treating systems

Adyel Tanveer M..

Plastic pollution is a ubiquitous planetary threat, affecting aquatic ecosystems globally. The current global production of plastic is over 300 Mt per year, only 20% is recycled and the remaining about 80% of which is accumulated in the environment. These plastics eventually break down into microplastics (MPs; <5 mm in size) by photolysis and naturalabrasion. Stormwater runoff is a direct pathway to transport land-based MPs to sensitivewaterways. MPs' accumulation in stormwater treatment green infrastructure or nature-based solutions such as constructed wetlands is emerging. This research aims to providea global overview of MPs' status, including size, shape, concentration, etc. in stormwaterrunoff. Besides, availability of MPs in the water and sediments of stormwater runoff-treatingsystems to be addressed. As stormwater runoff-treating systems are designed to treat nutrient(nitrogen and phosphorus), this research can provide a baseline if available MPs can alterthe performance of such system in short or long term scale. Moreover, this research willdevelop a better understanding of the key stakeholders, i.e., local water utilities, urban landdevelopers, and engineering consultants on MPs management.

Keywords : constructed wetlands , Nitrogen , Phosphorus , Stomwater , Waterways

#### Session 25.2\_Ma. Chaired by Martin Wagner, Trondheim

A meta-analysis of ecotoxicological hazard data for nanoplastics in marine and freshwater systems Yang Tong, Nowack Bernd. Paper number 333255

**Characterization, toxicity and leaching properties of tyre wear particles** *Rozman Ula, Marinšek Marjan, Kalčíková Gabriela.* Paper number 333671

Additive chemicals in plastic consumer products and their role in plastic toxicity Booth Andy, Igartua Amaia, Lyngstad Inger, Wagner Martin, Gomes Tânia, Almeida Ana Catarina, Øverjordet Ida Beate, Sørensen Lisbet. Paper number 334311

# Exposure to polymethylmethacrylate nanoplastics induced severe alterations in the morphology of cnidarians and amphibians

*Venâncio Cátia, Oliveira Miguel, Savuca Alexandra, Melnic Ioana, Martins Manuel, Lopes Isabel.* Paper number 334374

# A meta-analysis of ecotoxicological hazard data for nanoplastics in marine and freshwater systems

#### Yang Tong, Nowack Bernd.

There is an emerging concern about the potential health and environmental impacts of nanoplastics in the environment. Despite the lack of information on the exposure side, there is a growing number of ecotoxicological hazard data available which enable to conduct a hazard assessment for nanoplastics in freshwater and marine systems. Based on a critical evaluation of published studies and building probabilistic species sensitivity distributions (PSSDs), our work presents a comprehensive understanding of the state of art of nanoplastic ecotoxicity. Different freshwater and marine datasets were constructed based on different data quality levels and for each of the datasets, PSSDs were built for both mass- and particle number-based concentrations. Predicted no effect concentrations (PNECs) were then extracted from the PSSDs. Hereby, we report PNECs at 99 µg·L-1 and 72 µg·L-1 respectively for the freshwater and marine dataset after the removal of data measured in the presence of sodium azide (NaN3), which is considered to be a major interfering factor in the ecotoxicity testing of nanoplastics. By comparing the PNECs, we found that nanoplastics are less toxic than microplastics and many engineered nanomaterials. Besides, the effects of size and polymer type on the toxicity were also statistically tested. We observed that there is no significant difference in ecotoxicity for nanoplastics of different size while polystyrene nanoplastics were significantly more toxic than all other tested nanoplastics. In conclusion, the results presented in this work provide a comprehensive description of nanoplastic ecotoxicity based on the current knowledge. This work constitutes a fundamental step towards an environmental risk assessment for nanoplastics in freshwater and marine systems.

Keywords : Freshwater , Nanoplastics , Probabilistic risk assessment , species sensitivity distribution

#### **Characterization, toxicity and leaching properties of tyre wear particles** Rozman Ula, Marinšek Marjan, Kalčíková Gabriela.

Tyre wear particles (TWP) are complex material, composed of natural and synthetic polymers, and various additives, like fillers, softeners, vulcanization agents, etc. While driving, shear and friction forces causes abrasion of tyres, and TWP are released into the environment. Smaller TWP are released in the atmosphere, however, most of the particles remain on the roads and due to runoff end up in the soils or freshwaters near the roadside. TWP, used in this study, were obtained from a local car repair service and originated from an old tyre. They were sieved to obtain particles smaller than 300 um. TWP were characterized using a laser diffraction analyser, optical microscope and fieldemission electron microscope. A laser diffraction analyser was used for particle size distribution and the mean value of number particle size distribution was  $47.4 \pm 22.2 \mu m$ . Morphology characterization reviled irregularly shaped particles, with heterogenous surface. The toxicity of TWP and its leachate towards duckweed Lemna minor was determined with the standardized OECD test. The concentration of TWP was 100 mg/L in all tests. Leachate was obtained after 7 days incubation of TWP in duckweed's growth medium. After incubation, TWP were filtered and 10 fronds were added to leachate – the same as with particle testing. The toxicity test lasted 7 days and afterward the specific growth rate of duckweed, root length and chlorophyll content were determined. In comparison with control TWP did not have negative impact on a specific growth rate and chlorophyll content. However, root length was significantly reduced when duckweed was exposed to particles  $(37 \pm 6\% \text{ compared to control})$ , but not when duckweed was exposed to leachate. Results suggested that TWP have a negative impact on duckweed due to its physical properties (irregular particles with sharp edges) and not due to chemical leaching.

Keywords : duckweed , ecotoxicity , leaching , tyre particles

**Additive chemicals in plastic consumer products and their role in plastic toxicity** Booth Andy, Igartua Amaia, Lyngstad Inger, Wagner Martin, Gomes Tânia, Almeida Ana Catarina, Øverjordet Ida Beate, Sørensen Lisbet.

Toxicity assessment of most plastic and microplastic (MP) materials is typically conducted using pristine reference materials that do not accurately reflect the partially degraded materials normally found in the environment. Importantly, few studies have considered the role of plastic additive chemicals as the possible source of any observed toxic effects from MP exposure. All plastic consumer products contain chemical additives that provide the material with specific properties, including softeners, dves, antioxidants, UV stabilizers, antimicrobials and flame retardants. Many also contain residual chemicals used in polymer and/or product production processes. In the current study, 50 plastic consumer products representing different polymer types with a broad range of additive chemical profiles were characterised and their baseline toxicity towards aquatic species investigated using a tiered approach. All test materials were commercially sourced and cut into small pieces prior to use. First, solvent extracts of the plastic products were subjected to non-target screening using chromatography coupled to mass spectrometry, with compounds tentatively identified based on matches to mass spectral libraries. To allow investigation of a large sample set, data processing, including spectra deconvolution, library matching, logical filtering and searches against online PBT databases, has been automated to a large extent. A broad range of additive chemicals were identified in the different plastic products. As expected, the additive chemical profiles were found to vary significantly between the products, with some containing very little and others containing either a large number of different additives, specific additives at high concentrations, or a combination of the two. All test materials were then subjected to solvent extraction by methanol and transfer to DMSO prior to toxicity screening assessment by the marine Bacteria Luminescence Toxicity (BLT) test and marine microalgae (Skeletonema pseudocostatum).

Keywords : additives , chemical toxicity , effects , leaching , plastic impacts , risk assessment

# Exposure to polymethylmethacrylate nanoplastics induced severe alterations in the morphology of cnidarians and amphibians

Venâncio Cátia, Oliveira Miguel, Savuca Alexandra, Melnic Ioana, Martins Manuel, Lopes Isabel.

Scientific research on the toxicity of nanoplastics (NPLs) towards biota has focused mainly ons PS and PE NPLs, and on marine species, remaining a significant knowledge gap on the potential toxicity of other polymers, equally present in environmental samples (e.g., polymethylmethacrylate -PMMA), and on the toxicity of NPLs on freshwater species. Among freshwater invertebrate species, the cnidarians have been poorly studied in ecotoxicology, despite the advantages they offer as model species. Regarding freshwater vertebrates, amphibian species are known to be at rapid decline due to chemical contamination. Therefore, identifying and assessing the extent of potential effects of contaminants (such as NPLs) on these organisms is critical to their preservation. Accordingly, this work aimed at assessing the effects of sublethal levels of PMMA-NPLs on the body morphology and regenerative capacity of Hydra viridissima (cnidarian) and developmental stage and body morphology of Xenopus laevis tadpoles (frog). Morphological changes in H. viridissima were detected at concentrations ≤80 mg of PMMA-NPLs/L. At 80 mg PMMA-NPLs/L, 80% of the hydranths presented severe changes not yet reported in the literature, specifically the presence of doubled tentacles, elbow-like tentacles and curly tentacles. The tadpoles showed a higher sensitivity to PMMA-NPLs comparatively to the cnidarians. Concentrations of 1 mg of PMMA-NPLs provoked severe injuries in the abdomen zone, namely externalization of the gut in more than half of the organisms. The results obtained in this work suggest that cnidarians and early life stages of amphibians are good indicators for assessing freshwater quality in what concerns NPLs. These results provide important baseline information to further understand the toxic potential of PMMA-NPLs to freshwater biota, since it provided results not reported in the scientific literature to date. This work provides important information liable to be framed into more adequate NPLs' risk assessment schemes for the freshwater compartment.

Keywords : Anura , Hydra , Malformations , PMMA , Regeneration

#### Session 25.3\_O. Chaired by Guillaume Duflos, Boulogne Sur Mer

# The potential of microplastics to hinder microalgal biomass industries: raising awareness towards biosustainability

Cunha César, Lopes Joana, Paulo Jorge, Faria Marisa, Kaufmann Manfred, Nogueira Natacha, Ferreira Artur, Cordeiro Nereida. Paper number 333725

## Comparison of standard methodologies for plastic identification and quantification in field deployed oysters

Ribeiro Francisca, Okoffo Elvis, O'brien Jake, O'brien Stacey, Harris Jonathan, Kazerson Sarit, Mueller Jochen, Galloway Tamara, Thomas Kevin. Paper number 334198

# The impacts of polystyrene microparticles in the fatty acids and carbohydrate composition of the commercial bivalve species *Scrobicularia plana*

Tagliaferro Marina, Rocha Carolina, Knobelspiess Sara, Sahadevan Seena, Marques João C., Gonçalves Ana Marta. Paper number 334450

## Impact of polystyrene microplastics in anemones from the Portuguese coast: behavioral responses

Costa Figueiredo Ana, Oliveira Miguel, Figueira Etelvina, Martins Manuel, Pires Adília. Paper number 334497

# The potential of microplastics to hinder microalgal biomass industries: raising awareness towards biosustainability

Cunha César, Lopes Joana, Paulo Jorge, Faria Marisa, Kaufmann Manfred, Nogueira Natacha, Ferreira Artur, Cordeiro Nereida.

The harsh truth is: microplastics (MPs) are everywhere. These micro-sized polymers are ubiquitously distributed throughout all environmental and urban aquatic pathways. In the last decade, microalgae have been attracting increasing attention as a unique biomass feedstock for biofuel production, representing a sustainable and pollution-free fuel spring that is obtained from renewable sources. Recent reports have documented that wastewater treatment plants, central to urban water distribution to industries, discharge a median value of 2 million MPs/day. Given that water quality is a critical parameter in microalgal-based industries, the microalga Phaeodactylum tricornutum was grown in medium contaminated with environmental- and industrial-relevant (0.5 and 50 mg/L) concentrations of polystyrene and/or polymethyl methacrylate. Biochemical parameters such as cell growth, pH, biomass, photosynthetic pigments, extracellular carbohydrates and proteins were assessed over the microalgal full growth cycle (27 days). Overall, a two-stage response was observed, with a polymerand concentration-specific growth enhancement occurring in an early exponential phase, followed by an adaptive response that lead to a recovery until the stationary phase was reached. Results show that the production of photosynthetic pigments was pH-correlated during the first-stage response, while exhibiting a cell density-independent biochemical regulation in later stages of culturing. The biosynthesis of extracellular carbohydrates also followed the two-stage response, with emphasis on the major decrease exhibited during long-term exposure to MPs. Astonishingly, despite the long-term exposure to MPs not affecting cell abundance all across, a severe decrease in biomass yield (of up to 82%) was observed at the end of the culturing period. Altogether, this pilot laboratory-scale study shows that microalgal exposure to MPs disturbs its biochemical equilibrium, in a time-dependent manner, decreasing economically valuable biomass production. These results indicate that an "invisible", but highly abundant and ubiquitous pollution source in water might be playing an important role in the viability of microalgal-biomass biotechnology industry.

Keywords : Biomass , Microalgae , Microplastics contamination , Polymethyl methacrylate , Polystyrene

# Comparison of standard methodologies for plastic identification and quantification in field deployed oysters

Ribeiro Francisca, Okoffo Elvis, O'brien Jake, O'brien Stacey, Harris Jonathan, Kazerson Sarit, Mueller Jochen, Galloway Tamara, Thomas Kevin.

Marine invertebrates are vulnerable to microplastic exposure and their use in monitoringprogrammes to assess environmental contamination is a common practise. Microplastic con-tamination in ovsters is widespread but without standardized methodologies it is difficult toinfer by how much or to compare between studies. Additionally, current methodology is in-sufficient for determining what fraction exists at a specific size range particularly for smallersizes (<25µm). Fourier Transform-Infrared Spectroscopy is the most common technique foridentification of microplastics in biota. Accelerated Solvent Extraction followed by Pyroly-sis Gas Chromatography Mass Spectrometry is a promising emerging technique for smallersized microplastics and for determining plastic as a mass concentration. Here we presenta comparison between standardized identification (FT-IR) and quantification (Py-GC/MS)methods for microplastics analysis in oysters. This study had two aims: (1)identify and quantify plastic in oysters and compare methodologies and (2)investigate if the surroundingenvironment has an effect on the plastic content found in ovsters after deployment. Ovsterswere purchased from an ovster farm and a subset was set aside as a control. Ovsters weredeployed at the Port of Brisbane (Australia) for 14 days. Following exposure oysters weredissected, digested and divided in two subsets: filtered through a 2.7µm filter (for FT-IR)and through a sequential filtration process: 22µm, 1µm and nanofiltration (for Py-GC/MS).FT-IR analysis showed that 64% of the particles were cellulosic fibres. Py-GC/MS revealedthat the 1µm fraction had the highest total plastic content, followed by nano and 22µm.There was an increase in the average plastic content in deployed oysters compared to the control group. Our results show that the smaller sized microplastics are being overlooked inmost studies using FT-IR and that the two methods are not comparable, but instead com-plementary. The use of deployed bivalves as a proxy for plastic pollution is also discussed.

Keywords : deployment , oysters , Pyrolysis GC/MS ,  $\mu FTIR$ 

# The impacts of polystyrene microparticles in the fatty acids and carbohydrate composition of the commercial bivalve species Scrobicularia plana

Tagliaferro Marina, Rocha Carolina, Knobelspiess Sara, Sahadevan Seena, Marques João C., Gonçalves Ana Marta.

Estuaries are one of the most important ecological and socio-economic ecosystems. Estuaries are hotspots of microplastic pollution. Scrobicularia plana, an infaunal benthic bivalve, is one of the most abundant invertebrates, with an important role in estuarine trophic food webs and is also very appreciated by humans as food source, owning a great economic value. The aim of this study was to determine the potential impacts in the fatty acid (FA) and carbohydrates (CHO) content and thus on the quality of S. plana exposed to increasing concentrations of polystyrene microparticules (1 µm; MP). We used two bivalves' size classes (small; big) and exposed to 0.00, 0.25, 2.5 and 25 µg/L of the MP. Results show changes on FA and CHO profiles among both size classes of S. plana when exposed to polystyrene treatments. At the big individuals the content of saturated FA increased, and monounsaturated FA decreased with increasing MP concentration. At small organisms, monounsaturated FA increased from low to high MP concentrations. High unsaturated FA decreased from low to high MP concentrations (2.5 µg/L and 25 µg/L at small and big size classes respectively). The most abundant CHO was glucose for both size classes. In big size organisms, Rhamnose and Arabinose increased with increasing of polystyrene concentration. In big organisms CHO changes from control to MP treatments. In small size individuals, CHO content was similar among 0.0 to 2.5 µg/L but increased at the highest MP concentration. Results suggest S. plana big size class is more affected by polystyrene microparticles with a stronger effect on CHO profile, whereas both size classes decreased in FA quality when exposed to higher polystyrene concentrations. This study highlights the effects of polystyrene in edible bivalve species and thus on human diet and health.

Keywords : Carbohydrates , contamination , Estuaries , Lipids , microplastics , PUFA

# Impact of polystyrene microplastics in anemones from the Portuguese coast: behavioral responses

Costa Figueiredo Ana, Oliveira Miguel, Figueira Etelvina, Martins Manuel, Pires Adília.

Plastics are emerging drivers of environmental change that may influence marine near-shore ecosystems. Polystyrene (PS) is among the most produced polymers worldwide and is frequently found in the marine environment. Under environmental conditions, PS plastics brittle and break down into smaller particles, becoming more available to biota. Although various studies have been focusing on the effects of microplastics, information concerning the impacts of these contaminants on anemones is scarce. Anemones are among the most abundant groups of animals found in rocky shore habitats, maintaining equilibrium by providing habitat, food and protection for other species. Hence, this study aimed to assess the effects of PS microplastics on behavior (reaction time to touch (RT): mouth tightness aftertouch (MT), and mouth reopening after being closed (MR)) of the anemones Actinia equina and Aulactinia verrucosa. Percentage of nonresponsive anemones (NR) and exported water content after being exposed to air (WCE) was also assessed. The anemones Actinia equina and Aulactinia vertucosa were collected in a rocky shore - Praia da Granja (Portugal), and after depuration and acclimation were exposed for seven days to 3 µm PS microplastics particles (0.0; 0.001; 0.01; 0.1; 1.0; 10 mg/L). Results demonstrated that PS microplastics impacted the behavior of A. equina and A. verrucosa. MT and RT in A. equina decreased in exposed organisms (except at 0.01mg/L for MT and 10 mg/L for RT). MR increased at high concentrations (10 mg/L). The NR was higher in exposed organisms, with a peak at 0.01 mg/L. Concerning A. verrucosa, RT, MR and NR increased with concentration increase; MT increased in lower concentrations (0.1; 0.01 and 0.001 mg/L). WCE increased with the concentration increase for both species. Overall, results demonstrated that PS microplastics may present potential impacts for the studied species and, consequently, put at risk these populations.

Keywords : Actinia equina , Aulactinia verrucosa , Behaviour , Effects , Invertebrate , Microplastics

#### Session 25.3\_Me

**Multilevel Governance – What is the potential for microplastics regulation?** Schweiger Stefan, De Gueldre Greet, Schönbauer Sarah, Kingsbury Tony, Mederake Linda. Paper number 334630

#### **Multilevel Governance – What is the potential for microplastics regulation?**

Schweiger Stefan, De Gueldre Greet, Schönbauer Sarah, Kingsbury Tony, Mederake Linda.

(Micro)plastics regulation can address different stages along the life cycle of a plastic product including plastic production and product design, trade and consumer behaviour, recycling and waste management (so-called "landbased policies"), as well as wastewater management and water and marine protection ("water-based policies"). This session looks at the potential for water policies vs. land-based policies to tackle microplastic pollution from the local to the global level. The chair facilitates an interdisciplinary discussion among the panelists as well as between panel and audience. The discussion is supposed to shed light on the crucial target points for future (micro)plastics regulation.

Keywords : consumer behaviour , microplastics , multilevel governance , product design , recycling , regulation , waste management

#### Session 25.3\_Ma. Chaired by Mateo Cordier, Guyancourt

**A new approach to evaluate the toxicity of "environmental" plastic mixtures** *Magni Stefano, Della Torre Camilla, Binelli Andrea.* Paper number 334112

# Practical considerations for sampling, processing, analysis and reporting microplastic occurrence in increasingly complex media: microplastics in the water treatment process

Cross Richard, Horton Alice, Jurgens Monika, Johnson Andrew, Ball Hollie, Read Dan, Svendsen Claus.

Paper number 334288

# Impact of UV degradation on the fate and potential impact of textile microfibers and their additive chemicals in the marine environment

Sørensen Lisbet, Del Puerto Oihane, Groven Anette Synnøve, Hovsbakken Ingrid Alver, Sait Shannen, Sathananthan Dhiya, Igartua Amaia, Davies Emlyn, Sarno Antonio, Ribicic Deni, Salaberria Iurgi, Brakstad Odd Gunnar, Asimakopoulos Alexandros, Halsband Claudia, Herzke Dorte, Booth Andy. Paper number 334469 **A new approach to evaluate the toxicity of "environmental" plastic mixtures** Magni Stefano, Della Torre Camilla, Binelli Andrea.

Knowledge on the impact of plastics on aquatic organisms are generally based on the effect evaluation induced by standard materials. This reductionist approach ignores the different characteristics of plastics when in the ecosystems, as well as the possible effect of chemicals adsorbed on their surface. In this study, we carried out the quali-quantitative evaluation of 5 plastic mixtures collected along the Lambro River (Northern Italy), that crosses one of the higher anthropized and industrialized Italian areas, contemporarily evaluating their environmental hazard by an ecotoxicological approach. We collected plastics at 5 sampling points along the river using two twin plankton nets with a mesh of 300 µm, repeating the sampling for 3 days in a week. Plastics collected from one net were quantified and characterized through the Fourier Transform Infrared Microscope System ( $\mu$ FT-IR), while plastics from the second one were separated from interfering materials and subsequently used for in vivo exposures. We exposed the freshwater mussel Dreissena polymorpha to these plastic mixtures for 21 days, evaluating the toxicity at the end of exposures using an integrated approach based on the measure of some endpoints from the cellular and molecular levels to the whole organism. The obtained results showed an increasing trend of plastic concentration along the Lambro course, with an estimate of about 4,700,000 plastics released daily into the Po River, the main Italian watercourse. After the exposure to "environmental" plastic mixtures we observed a great mortality coupled with significant decrease in cell viability in mussels exposed to some mixtures, as well as an imbalance of the oxidative status, with a significant increase of both catalase activity and protein carbonylation and a modulation of proteins mainly involved in the oxidative stress response. This work aimed to propose a new tool to address the Environmental Risk Assessment for these emerging pollutants.

Keywords : Biomarkers , Freshwaters , Monitoring , Plastics , Proteomics

# Practical considerations for sampling, processing, analysis and reporting microplastic occurrence in increasingly complex media: microplastics in the water treatment process

Cross Richard, Horton Alice, Jurgens Monika, Johnson Andrew, Ball Hollie, Read Dan, Svendsen Claus.

Microplastics may be considered as plastic anywhere between 1nm and 5mm, presenting an analytical challenge for their detection and quantification. No single method can enumerate all polymers, across this full size range. Clarity on the operational limits that define microplasticsis an essential part of any investigation to allow for comparison between studies. At each stage, from sampling through to analysis, consideration should be made as to how these steps influence quantification of microplastics. Sampling strategies often require filtration, introducing a minimum size for microplastics that must be reported. Similarly, imaging approaches such as Raman or infrared techniques can resolve microplastics to different sizes. The choice of analytical method must be balanced by the study requirements. Practical considerations of cost and time balanced against gains in precision are not trivial when large sample numbers are required. We present our experiences in sampling, method development, and analysis as part of a campaign monitoring 8 water treatment works and 8 wastewater treatment works over 7 months. This campaign (UK Water Industry Research Ltd, "Sink to River-River to Tap") enumerated the number and polymer identity of microplastics in raw water, potable water, influent, effluent and treatment residues. This presentation focuses on practical recommendations, in particular the use of blanks, spike-recoveries and replication, in the context of controlling sources of contamination and the patchiness inherent in homogeneous samples. Efforts are underway to harmonise methods and analytical approaches quantifying microplastics in the environment. In the meantime, there is much value in increasing the longevity of the data generated in this highly productive field of research. By necessity, data comes from a plethora of different sampling, processing and analytical strategies. Sharing experiences from the lab and improving transparency in reporting goes some way to allowing data generated today to retain its value as the field develops.

Keywords : FTIR , microplastic extraction , sampling , wastewater , water treatment

#### Impact of UV degradation on the fate and potential impact of textile microfibers and their additive chemicals in the marine environment

Sørensen Lisbet, Del Puerto Oihane, Groven Anette Synnøve, Hovsbakken Ingrid Alver, Sait Shannen, Sathananthan Dhiya, Igartua Amaia, Davies Emlyn, Sarno Antonio, Ribicic Deni, Salaberria Iurgi, Brakstad Odd Gunnar, Asimakopoulos Alexandros, Halsband Claudia, Herzke Dorte, Booth Andy.

Microfibers (MFs) are frequently reported as the most dominant type of microplastic (MP) found in the marine water column and sediments. A major source of MFs is the use and washing of textiles. Although WWTPs can remove up to 98% of MP, estimates suggest billions of MP are still released from a single WWTP annually. Intrinsic properties (polymer type, density, size) will influence environmental degradation, settling times, and ingestion of MFs by marine organisms. Less well understood is the influence of environmental degradation on the fate of MFs. In the current study, we compare the effect of UV exposure on the degradation and fragmentation of polyester (PET), polyamide (nylon; PA), polyacrylonitrile (acrylic; PAN) and wool fibers. Degradation of MFs was conducted in seawater under environmentally relevant exposure conditions using simulated sunlight. PA, PET and wool MFs exhibited changes in surface morphology after just 2 weeks from the start of exposure, followed by fragmentation after.

Keywords : Additive chemicals , Ingestion , Leaching , Microfibers , Settling , UV degradation

**Poster session 25.4** 

#### Session 25.4\_O. Chaired by Eva Cardona, Menorca

# Sources of macroplastic in the northeast coast of Brazil: an exercise with high school students for creating science-based public policies

Alves Ribeiro Celio, Lacerda Ana Luzia De F., Ferreira Divina Magna, Barbos Raí Bernardo, Bonifácio Francisco José De Sousa, Rodrigues Marcelo. Paper number 334227

**Strategies for the separation of microplastics from water via density modification** *Martínez De Pedro Zahara, Munoz Macarena, Ortiz David, Casas Jose A.*. Paper number 334335

# A method for accelerated processing of microplastic samples using a flatbed scanner and MPScanTool plugin for ImageJ

*De Haan William P., Sanchez-Vidal Anna, Canals Miquel.* Paper number 334428

# Sources of macroplastic in the northeast coast of Brazil: an exercise with high school students for creating science-based public policies

Alves Ribeiro Celio, Lacerda Ana Luzia De F., Ferreira Divina Magna, Barbos Raí Bernardo, Bonifácio Francisco José De Sousa, Rodrigues Marcelo.

This study was developed by high school students and teachers on combining curricular theory and practice in the pedagogical teaching of science, as well as on practicing environmental education in a multidisciplinary way. Before starting sampling, professors and students were trained by an expert on this subject. Sampling were performed during seven months in two sandy beaches, Flecheiras and Emboaca, located in the northeast coast of Brazil. Each point had 60m x 20m transects (in triplicates) for manual collection of macroplastics from the superficial sediment. All collected plastic was separated by category, such as: PET bottles, bottle caps, straws, plastic bags, tires and "unidentified" (adapted from UNEP, 2016). Items were counted and weighed, and then plotted in a spreadsheet for further analysis and creation of a database. The total amount of litter was 4,214 items (71.55 kg) in Flecheiras, and 4,209 items (129.70 kg) in Emboaca. Despite the differences in weight, the number of items was similar between locations. We believe that high abundance of plastics in Emboaca is due the dynamics of marine currents, that brings a great contribution of waste to this region, as this place is out of the village; this distance also limits beach clean up by the municipality. There is a different situation in Flecheiras beach, as this beach is located by the village, and receives thousand of tourists annually; even with daily beach clean up in Flecheiras, the high amount of litter may be due the continuous disposal by beach users (mainly tourists), and also urban drainage. Identifying sources is essential for creating science-based strategies to prevent the entry of plastic waste in coastal zones, and the results and hypotheses formulated in this study are being used to create an official document to the municipal council of Trairi city (where these beaches belong).

Keywords : Plastic pollution , Waste management

# **MICR02020 INTERNATIONAL CONFERENCE**

23-27 NOVEMBER 2020 LANZAROTE AND BEYOND\*

#### FATE AND IMPACTS OF MICROPLASTICS: KNOWLEDGE AND RESPONSIBILITIES

#### Sources of macroplastic in the northeast coast of Brazil: an exercise with high school students for creating science-based public policies

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#### ABSTRACT

This is an ongoing study developed by high school students and teachers on combining theory and practice in the pedagogical process of teaching Science, and practicing environmental education in a multidisciplinary way.

Samples were performed during 7 months (Jan-Jun 2020) in two sandy beaches, Flecheiras and Emboaca beaches, in the northeast coast of Brazil. Transects of 60m x 20m (in triplicates) were done for manual collection of macroplastics. All collected plastic was separated by category (bottles, bottle caps, straws, fragments, plastic bags, tires and "undentified"), and then counted and weighed.

The total amount of litter found until now was 4,214 items (71.55 kg) in Flecheiras, and 4,209 items (129.70 kg) in Emboaca. Despite the diferences in weight, the number of items was similar between locations.

The high abundance of plastics in Emboaca could be due local marine currents as being the major contributor of waste to this region, as it is an isolated beach; the distance from the village also limits beach clean up by municipalities. On the other hand, Flecheiras beach is located in a village and receives thousand of tourists annually; even with daily beach clean ups at this place, Flecheiras presented a high amount of litter, probably due the continuous disposal by beach users and urban drainage. Identifying sources is essential for creating strategies to prevent the entry of plastic waste in costal zones, and the results and hypotheses formulated in this study are being used for the development of an oficial document to the city council of Trairi (where these two beachs belong).



**Strategies for the separation of microplastics from water via density modification** Martínez De Pedro Zahara, Munoz Macarena, Ortiz David, Casas Jose A.

The widespread occurrence of microplastics (MPs) in the aquatic environment represents one of the most important environmental concerns nowadays. Although MPs can enter the environment through multiple pathways, wastewater treatment plants (WWTPs) have been recognized as important sources for MPs introduction into the aquatic systems. It is estimated that rivers, the main recipients of WWTPs discharges, transport up to 90% of the global MPs load into the sea (Schmidt et al., Environ. Sci. Technol. 2017, 51, 12246). Only in Europe, 520000 tons/year of MPs are released in WWTPs effluents (Alimi et al., Environ, Sci. Technol, 2018, 52, 1704). In this context, the development of innovative water treatment processes that allow the effective removal of MPs at WWTPs is crucial. This is an important challenge as the small size of MPs and their low chemical and biological reactivity significantly limit their elimination. The methods developed so far have been mainly focused on their sampling, showing important limitations for water treatment. In our research group, the development of strategies for MPs separation from water via density modification are being investigated. In this work, the removal of polystyrene (PS) and polyester fibers (PE) has been evaluated by their interaction with high-dense hematite (Fe2O3) microparticles. We have found that these mineral particles covered completely both PS and PE MPs leading to a significant increase on their density, which facilitates separation by sedimentation. On the other hand, carbon coating of MPs is being also investigated as an alternative approach to separate MPs. In this case, the removal of polyethylene terephthalate (PET) has been investigated by its coverage with a low-dense activated carbon, which allowed to increase the hydrophobic properties of the particles. These can adsorb air bubbles and float by reducing their density, favoring MPs separation via dissolved air flotation (DAF).

Keywords : activated carbon , hematite , microplastic , water treatment







#### Strategies for the separation of microplastics from water via density modification

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[2] Schmidt et al., Environ, Sci. Technol, 51 (2017) 1224

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# A method for accelerated processing of microplastic samples using a flatbed scanner and MPScanTool plugin for ImageJ

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Plastic pollution has become a major threat to marine and terrestrial organisms and habitats worldwide. In particular, the study of microplastics (; 5 mm in length) in different environmental matrices often requires routine quantification, measurement and classification, especially during the analysis of high-throughput monitoring data. However, most of current protocols rely on timeconsuming and biased counting and classification of microplastics. To address this problem, we propose a novel simple and inexpensive method for accelerating laboratory processing of microplastic samples using a modified common flatbed scanner coupled with a custom-built and open-source ImageJ/Fiji macro plugin, named MicroPlastic Scan Tool (MPScanTool). By the superposition of bright-field and dark-field scans and precise image alignment and we achieve major accuracy (¿90%) detecting microplastics down to 0.15 mm in length at a 1200 dpi (47,2 pixels mm-1) scan resolution, and are able to differentiate between transparent or translucent and opaque microplastics in 95% of the cases. By color calibrating the flatbed scanner (i.e. by means of an IT8.7 Target) and using standard color charts we achieve a color classification of microplastics with 92% of accuracy at an average processing rate of 300 plastics/minute. MPScanTool provides semiautomatic counting and particle categorization of microplastic samples and automatic file management, data processing, and color determination, thus accelerating microplastic sample processing, improving data quality and reducing visual subjectivity during sample treatment.

Keywords : flatbed scanner , ImageJ , microplastics , MPScanTool plugin , processing

# A method for accelerated processing of microplastic samples using a flatbed scanner and MPScanTool plugin for ImageJ (num. 334428)



Universitat de Bar



# Method validation

with 92% of accuracy at an average processing rate of 300 plastics per By the superposition of bright-field and dark-field scans and precise standard color charts we achieve a color classification of microplastics translucent and opaque microplastics in 95% of the cases. By color scan resolution, and are able to differentiate between transparent or microplastics down to 0.15 mm in length at 1200 dpi (47,2 pixels mm-1) calibrating the flatbed scanner (i.e. by means of an IT8.7/2 Target) and mage alignment and we achieve high accuracy (>90%) in detecting

# Summary and outlook

 $\rightarrow$ Our novel, simple and inexpensive method using a common flatbed microplastic sample processing, improving data quality and reducing scanner and MPScanTool macro plugin for ImageJ accelerates visual subjectivity during sample treatment

→The combination of black-background reflective images and greenallows discrimination between transparent/translucent and opaque background transflective images increases microplastic detection and plastics that can further be classified in color categories.

Extensive 2D particle characterization parameters can be extracted standardized color measurements and manual/semi-automatic digital semi-automatically for each individual particle, together with automatic

→Our method presents clear benefits to both the scientific community and citizen science projects that work with high-throughput monitoring Mediterranean Sea" by William P. de Haan et al. and "Paddle surfing monitoring of microplastics in coastal waters in the Northwestern data (see presentations at the MICRO2020 conference "Long-term classification of plastic categories

for science on microplastic pollution" by O. Uviedo et al.

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Acknowledgments

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#### Session 25.4\_Me. Chaired by Ana Liria, Gran Canaria

#### Plastic: an insidious new threat to Amazon's conservation

Siqueira Morais Leonardo Mario, Monteiro Raqueline Pereira, Pegado Tamyris, Queiroz Arnaldo Fabrício, Cirino Saraiva Bianca Cristina, Tavares De Oliveira Maria José, Fenzl Norbert, Giarrizzo Tommaso, Martinelli Filho José Eduardo. Paper number 334430

### Incidence of land uses on the consumption of microplastics by fish communities in Uruguayan lowland streams

*Vidal Camila, Lozoya Juan Pablo, Teixeira De Mello Franco.* Paper number 334449

#### Microplastic pollution in the Arctic seas

Makeeva Irina, Ershova Alexandra, Eremina Tatiana, Tatarenko Yuri. Paper number 334576

#### Plastic: an insidious new threat to Amazon's conservation

Siqueira Morais Leonardo Mario, Monteiro Raqueline Pereira, Pegado Tamyris, Queiroz Arnaldo Fabrício, Cirino Saraiva Bianca Cristina, Tavares De Oliveira Maria José, Fenzl Norbert, Giarrizzo Tommaso, Martinelli Filho José Eduardo.

Microplastic (MP) pollution has been recognized as one of the greatest threats to aquatic biota, becoming one of the most studied issues in the recent environmental impacts literature. The number of publications on MPs has increased exponentially since the 1990's. In developing countries like Brazil, the study of MPs is even more recent, and most of the scientific production is concentrated in the last 10 years. This pattern fits perfectly for the Amazon: up to June 2020, only 34 publications were available for the region, 10 (29.4%) peer-reviewed articles and 24 (70.6%) grey literature documents. Most studies were conducted in Brazilian Amazon (32) and were published between 2010 and 2020, but 83.3% were from the last two and a half years (2018 to 2020). The main subjects investigated were environmental screening and monitoring of microplastics (38%), ingestion and uptake of MPs by biota (35%), physiological effects of MPs (15%) and potential biomonitor species (12%). Plastic fibers was the dominant type of MPs in 50% of the publications and the size analyzed varied from 1 µm to 5 cm. Nearly half of the publications (16) used analytical tools for identifying the plastic polymers. The most common identified polymers were Polyamide, Polyethylene (PE), Polypropylene (PP), Rayon, Polyethylene terephthalate (PET), polystyrene (PS), acrylonitrile butadiene styrene (ABS), polyurethane (PU), Poly(methyl methacrylate) (PMMA) and polyvinyl chloride (PVC). This review shows that microplastic contamination is already a reality for the Amazon environment and related biota. Negative effects at different ecological levels are probably taking place in the ecosystem. However, limited studies, lack of standardized methodologies and methodological flaws make it difficult to establish the fundamental knowledge necessary to evaluate the environmental levels and impacts of MP in the region, becoming urgent the need to improve scientific knowledge and laboratory infrastructure to study MPs in Amazon.

Keywords : Brazil, Microplastic, Plastic debris, Pollution







#### Plastic: an insidious new threat to Amazon's conservation

MORAIS, L.M.S.\*; MONTEIRO, R. P.; PEGADO, T., QUEIROZ, A.F.; SARAIVA, B.C.C.; de OLIVEIRA, M.J.T.; GIARRIZZO, T.; MARTINELLI-FILHO, J.E.

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#### Introduction

Microplastic (MP) pollution has been recognized as one of the major threats to aquatic biota, becoming a dominant issue in the recent literature on environmental impacts. The number of publications on MPs has increased exponentially worldwide since the 1990's. For the Amazonian ecosystem, the study of MPs is even more recent, and all the scientific production is concentrated in the last 10 years. This review aims to describe the current state of scientific knowledge about plastic pollution in the Amazon environment and related biota.

#### Materials and methods

An extensive literature review were conducted using databases such as Google Scholar, Web of Knowledge and ResearchGate. The set of keywords used in the review were searched in English, Spanish and Portuguese. A meta-analysis was carry on by filling out an *Excel* spreadsheet with several variables extracted from the studies.

#### Results

Up to June 2020, only 34 publications were available on MPs at the Amazon or related biota, 10 (29.4%) peerreviewed articles and 24 (70.6%) grey literature documents (figure 1A). Most studies were conducted in Brazilian Amazon (32) and were published between 2010 and 2020, but 83.3% were from the last two and a half years (2018 to 2020) (figure 1B). The main subjects investigated were environmental screening and monitoring of microplastics (38%), ingestion and uptake of MPs by biota (35%), physiological effects of MPs (15%) and potential biomonitor species (12%) (figure 1C). Considering the different environments, most studies were conducted on rivers and beaches (38% each), followed by estuaries (15%) and the Amazon shelf (6%) (figure 1D). A single study considered the whole environment, since it is a review under publication for the whole Peru. MP ingestion was reported for freshwater, marine and estuarine fishes from the amazon coast. A plastic fragment was found in the amazon manatee (Trichechus inunguis). The sea anemone *Bunodosoma cangicum* and the freshwater crab *Dilocarcinus pagei* were also found to accidentally consume MPs. Plastic fibers was the dominant type of MPs in 50% of the publications and the size analyzed varied from 1  $\mu$ m to 5 cm. Nearly half of the publications (16) used analytical tools for identifying the plastic polymers. The most common identified polymers were Polyamide, PE, PP, Rayon, PET, PS, ABS, PU, PMMA and PVC.



Figure 1: the distribution of the 34 publications units on microplastics at the Amazon (both grey and white literature), according to A: publication format; B: publication year; C: research theme and D: target environment.

#### Conclusion

This review shows that MPs contamination is already a reality for the Amazon environment and related biota. Negative effects at different ecological levels are probably taking place in the ecosystem. However, limited studies, lack of standardized methodologies and methodological flaws make it difficult to establish the fundamental knowledge necessary to evaluate the environmental levels and impacts of MP in the region, becoming urgent the need to improve scientific knowledge and laboratory infrastructure to study MPs in Amazon.

#### Acknowledgments





# Incidence of land uses on the consumption of microplastics by fish communities in Uruguayan lowland streams

Vidal Camila, Lozoya Juan Pablo, Teixeira De Mello Franco.

The physical, chemical and diversity characteristics of streams and rivers are influencedby the land use in their basins. A large number of pollutants reach watercourses by super-ficial and subsuperficial drainage or direct disposal. Plastics currently make up the mostimportant waste asset, representing an important part of the transported material as well asaccumulated in water courses. This work analyzes the consumption of microplastics (MPs,<5mm) by the fish communities in streams with two contrasting types of land use. For thispurpose we worked with 3 streams impacted by urbanization and 3 by extensive livestockfarming. The stomach and intestinal contents of 314 individuals of 27 species were ana-lyzed in total, by a modified alkaline digestion, and observed under stereo microscope. TheMPs were recovered and confirmed under a polarized light microscope. A total of 373 MPswere found, of which the majority corresponded to fibers (318) and the rest to fragments(55). A significant difference was found between the percentage of individuals with Mps between both systems (51.6% in ranchers and 76.6% in urban, p=0.014 Mood's Median),but no difference was found in the average MP ingested per individual in both systems. This study establishes the first baseline on MP consumption by fish in Uruguayan streams, and in corporates conditions of the water courses and basins regarding this global environmental problem.

Keywords : fish , gut content , microplastics , pollution , streams

# Incidence of land uses on the consumption of microplastics by fish communities in Uruguayan lowland streams

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Streams and rivers are considered to reflect their location and the activities that take part on their basins, the natural characteristics and geomorphological history (Wetzel et al 2000). Human activities tend to generate impacts and contaminants, and microplastics (MPs) are one of the most important emergent pollutant, in water bodies, land and air (García et al, 2020).

The encounter with the biota that inhabits the different ecosystems it's inevitable, and the consequences resulting from this interactions can vary from ingestion and accumulation in the digestive tract, to even death (Folev et al. 2018).

There is little information on the dynamics that occur in lowland streams and the differential impacts product of varios land uses. This work analyzes the consumption of microplastics (< 5mm) by fish communities in streams with two contrasting types of land use in Uruguay.

#### **Objective:**

Evaluate the incidence of microplastics in the diet of fish communities in streams with contrasting land use (extensive ranching and urbanization).

Specific objective:

- I. Evaluate and compare the presence of microplastics in the stomach contents of fish in three streams with extensive livestock farming and three streams with urhan use
- Estimate the microplastics load at the community level with respect to both types of land use (extensive ranching and urban)

Stomachs were dissected from up to 5 individuals per species, and an alkaline digestion modified from Dehaut (2016) was performed. Each sample was later examined under a magnifying glass, were MPs were observed, measured, photographed and moved to a microscope in order to be exposed to polarized light for corroboration.

#### **Results**:

- A total of 373 MPs were found, mostly fibers (85%), and more commonly blue (67%) →
- MPs were found in 189 individuals from 314 analized (60%), corresponding to 29 species. →

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- → Ranching streams showed a significantly lower percentage of individuals with MPs than Urban streams (Test Mood's median, Chi2=6, p= 0.014) (Figure 2). There were no significant differences regarding average of MPs ingested per individual between the two land uses.
- Ranching streams showed significantly lower MPs per gram of individuals than Urban streams (Test Mood's median, Chi2=6, p= 0.014)
- There is a change in the species with the highest incidence of MPs if we consider average per individual or average per gram (Figure 3)



Figure 1. Microplastics (MPs) found in stomach samples. A, B and C are fibers, and D,E and F are fragments. C and F show the reaction of the MPs to polarized light

Figure 2. Median and extremes for the percetnage of individuals with microplastics for ranching (green) and urban (orange) streams. MPs= microplastics

2 3 12 2 10 MPs nor gr am of body

Figure 3. Average of microplastics (MPs) per individual per species, with its standard error (A). Average of MPs per gram of individual (B). Both graphs present the species in increasing order. In parentheses next to each species, you will find firstly the number of individuals analyzed, and secondly how many sites within each group the species was present, in Ranchers and Urban streams respectively

#### Discussion and conclusions:

- This is the first attempt to understand MPs ingestion in lowland streams in Uruguay ->
- The type of MPs found are consistent with previous works in the region (Pazos et al, 2017; García et al 2020) →
- There is a significant difference between the amount of individuals in fish communities in lowland streams eating MPs, Urban land use is associated with a higher consumption.
- → The high percentage of individuals with MPs in ranching streams leads to believe human presence rather than urban areas might result in a higher presence of MPs in streams. Atmospheric deposition should be considered in future investigations
- -> Analyzing MPs with regard to body weight, at stream and species level, provides a new way of understanding potential impacts. Smaller species like Cnesterodon decemmaculatus, might have the same average consumption than bigger species, but has a much higher MPs per gram of body weight ratio, which could indicate higher stress.
- -> Few studies consider MPs consumption at the community level and only consider individual level, but it should be assessed and it could give estimates to the amount of "active" MPs for trophic transfer.

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#### **Microplastic pollution in the Arctic seas**

Makeeva Irina, Ershova Alexandra, Eremina Tatiana, Tatarenko Yuri.

There is a high potential of microplastics accumulation in the Arctic, namely in the Barents Sea region [Cozar et al., 2017]. The Russian Arctic is a poorly explored region, but due to climate changes and the North Passage (NP) opening the anthropogenic load on this area has been increasing. There is also a lack of methods for environmental monitoring of the Arctic seas. The main purpose of this study was to carry out a first ever survey of all Russian Seas along the North Passage and to develop a sampling technique for microplastic particles content in the surface water layer for the Arctic Seas characterized by very high productivity. Over 120 water samples were taken during the Program "TRANSARCTICA-2019" in the summer 2019 (July – September). Water from the subsurface layer (4 m) was sampled using shipboard pump and then filtered through a special filtering instrument with metal mesh (100 µm). Samples were fixed and treated in laboratory according to the methodology [Zobkov et al., 2018], including organic material digestion, visual quantification of microplastic particles with microscope and spectroscopic analysis. The work was supported by the Ministry of Science and Education of RF, project № FSZU-2020-0009. 14 samples were processed for preliminary assessment of microplastic contamination of all 7 seas along the NP. Average microplastic particles content was 0.03 particles/l. The results of the study showed the relationship between the nature of pollution and the remoteness of the sea from large cities. Seas that have a large transport load are characterized by only higher concentrations, but also by a certain type of microplastics. The highest concentration of microplastics was noted in the Sea of Okhotsk and the Barents Sea, and the lowest - in the East-Siberian Sea, and was 0.357 and 0.001 particles per l, respectively.

Keywords : Arctic Seas , methods of sampling , microplastics


#### MICROPLASTIC POLLUTION IN THE ARCTIC SEAS

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#### INTRODUCTION

Today, pollution of the marine environment with microplastics (plastic particles less than 5 mm) is a global environmental problem. In the aquatic environment, microplastic particles travel with currents throughout the World Ocean until they reach the accumulation zone. There are several such zones ("garbage patches") in the world, and one of them is already being formed in the Arctic region [1]. The Russian Arctic, which is one of the most important highly productive marine ecosystems of the Arctic Ocean (AO), with a constantly increasing anthropogenic load, is still poorly studied. There is still no single standardized method (protocol) for determining the concentration of microplastic particles in the Arctic. In the summer of 2019, the RSHU conducted field research in frames of TRANSARCTIC-2019 programme aiming at quantitative and qualitative assessment of the accumulation of microplastic particles in the water area of the Russian part of the Arctic basin.



#### METHODOLOGY

Sampling was carried out using a shipboard pump built into the flow-through system in the keel part of the vessel and sampling seawater along the vessel's course from the subsurface layer (up to 4 m). Water flowing through a special sampler was filtered through a metal mesh (mesh size - 100 µm). Then the sample was fixed and sent for laboratory analysis.

The analysis of samples was carried out according to the methodology developed by the Atlantic Branch of P.P. Shirshov Institute of Oceanology of Russian Academy of Sciences [2], and consisted of two stages:

• Digestion of organic material;

• Visual identification of microplastic particles using a microscope.

To obtain a preliminary assessment of microplastic pollution of the Arctic seas, 14 samples were treated, taken in all seas of the Arctic basin of the Russian Federation.

Third stage – FTIR analysis for identification of plastic type – was not completed due to COVID-19 restrictions.



RESULTS

#### CONCLUSION

A quantitative assessment of microplastic particles in the Arctic seas showed microplastic particles of various shapes, sizes and colors.

Among them, the most common were filamentous transparent fibers - they were present in each of the samples. Particles of indeterminate shapes were found in the seas with a strong traffic load, namely the Barents, Kara and Okhotsk seas. The particle size varied from 100 to 3500 µm.

The highest concentration of microplastics was observed in the Sea of Okhotsk, and the lowest - in the East Siberian Sea and amounted to 0.357 and 0.001 particles per 1 liter, respectively.

The method of subsurface shipboard pump has been successfully used by researchers Lusher et al., (2015) in the area of the Spitsbergen archipelago in the Barents Sea. However, the results of our expedition showed significantly higher concentrations (30 pcs / m<sup>3</sup> in this study and 2.46 pcs / m<sup>3</sup> in Lusher et al. [3].

Laboratory analysis of the samples according to the method of AB IO RAS [2] showed that this technique needs to be improved for the Arctic seas due to their high productivity and abundance of zooplankton and krill, thus the organic material is not completely digested, which is significant and complicates the analysis process and affects the accuracy of the result.

The work will be continued in order to get the qualitative assessment of types of plastic particles (after the FTIR analysis will become available), as well as to develop recommendations for the determination of microplastic particles in the marine waters of the Russian Arctic.



Coordination of Source and Sou

Stations	Liters filtered, (m <sup>3</sup> )	Number of particles found, (pcs)	Concentration, (pcs / l)		
Okhotsk sea					
Station 1	0,075	9	0,120		
Station 2	0,028	10	0,357		
Bering Sea					
Station 1	0,173	14	0,081		
Station 2	0,450	7	0,015		
Chukchi sea					
Station 1	0,787	5	0,006		
Station 2	1,087	28	0,026		
East-Siberian Sea					
Station 1	2,700	4	0,002		
Station 2	1,800	1	0,001		
Laptev sea		<b>r</b>			
Station 1	1,500	9	0,006		
Station 2	1,950	14	0,007		
Kara Sea					
Station 1	1,688	10	0,006		
Station 2	1,800	16	0,009		
Barents Sea					
Station 1	1,875	38	0,020		
Station 2	3,150	92	0,030		
		and the second	ALL CONTRACTOR		



#### Session 25.4\_Ma. Chaired by Ignacio de Sobrino, Lanzarote

#### Plastic flux for innovation and business opportunities in Flanders (PLUXIN)

Devriese Lisa, Toorman Erik, Janssen Colin, Asselman Jana, Sterckx Sindy, Knaeps Els, Teunkens Bert, Van Damme Stefan, Everaert Gert. Paper number 334360

**Citizen involvement in the fight against plastic pollution in the Biosphere Reserve of Menorca** Marta Pérez, Rebeca Morris, Carlos Salord, Eva Cardona, Victor Carretero, David Doblado, Eva Marsinyach. Paper number 334369

### Beach litter of Karasu Coast in Black Sea (Turkey): Sources, composition, fate and some suggestions

*Yurtsever Meral, Başargan Furkan.* Paper number 334584

#### **Plastic flux for innovation and business opportunities in Flanders (PLUXIN)**

Devriese Lisa, Toorman Erik, Janssen Colin, Asselman Jana, Sterckx Sindy, Knaeps Els, Teunkens Bert, Van Damme Stefan, Everaert Gert.

Plastics are globally dispersed and reported at increasing concentrations in marine ecosystems. Due to their persistence in aquatic environments the global plastic problem will last for decennia. Removal of plastic at the source prior to reaching the marine environment is instrumental. Hence, plastic detection methods and plastic remediation measures are urgently needed and may become obligatory in the future. A first prerequisite to take effective plastic remediation measures is to know where and when action should be taken. However, to date there is a critical knowledge gap about the whereabouts of plastics and about their flux towards the marine environment. This information is crucial to fast track cost-efficient plastic remediation measures. A central objective in the Flanders Innovation & Entrepreneurship (VLAIO) via the Blue Cluster funded project PLUXIN is to develop a two dimensional-horizontal (2DH) plastic dispersal model of the estuarine and harbor environments in Flanders. The model will be calibrated and validated with vertical movement experiments and field sampling data. Plastics will be identified from remote sensing reflectance data through image recognition algorithms ('Machine Learning'), hence resulting in an automated plastic detection method. This information in combination with in situ sampling will validate the 2DHmodel. Flanders is a top region in terms of hydrologic and hydraulic expertise, and hosts key international companies active in offshore and coastal engineering and the circular economy. Flemish knowledge institutes are internationally recognized for their plastics-related expertise. This holistic project will integrate this knowhow and take essential steps to further our expertise. Based on the transdisciplinary PLUXIN research initiative a comprehensive and systematic picture of distribution of plastic in the aquatic environment will be obtained. Remote sensing and in-situ observations in combination with numerical models will contribute to our understanding of the sources, circulation patterns and fate of plastic in the aquatic environment.

Keywords : facilitate plastic business models , integration of experimental and modelling work , interventions , whereabouts of plastic

### Plastic flux for innovation and business opportunities in Flanders (PLUXIN)

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### Rationale

Plastic should be **removed at the source** prior to ending up in the marine environment.

Where, when and how should action be taken?

Critical **knowledge gap** about the whereabouts of plastics and about their flux towards the marine environment.

Legislation is being developed.

Basic data on concentrations and fluxes will create opportunities to develop **new economic activities.** 

The PLUXIN project kicked off in September 2020 and is a **three year** EUR 1.8 million basic research project funded by VLAIO through the Blue Cluster in Flanders with **two main objectives**.

### Way forward

Laboratory experiments, remote sensing and insitu observations in combination with numerical models for a better understanding of sources, circulation patterns and fate of plastic in the aquatic environment.



### **Objectives**

**Scientific objective** - Where is the plastic and how to detect?

- 3D insight in whereabouts of micro- and macroplastic
- Automated detection of plastic via remote sensing

**Valorisation objective** - Identify, map, and initiate pathways for valorisation. Stimulate innovation and economic development in Flanders.



### **Expected** output

Protocol for sampling, detecting and identifying micro- and macroplastic aligned with EU initiatives.
Known quantities of plastic present in rivers and harbours.

Conceptual model to quantify plastic flux.

Quantified flux of plastic to the ocean.

Unraveled accumulation zones of plastic.

Quantified **travel speed** of plastic through the water column.

Hyperspectral information for automated detection.

Automated **detection technology** and method.

### See you at MICRO2022 with our results

# VLIZ











## Citizen involvement in the fight against plastic pollution in the Biosphere Reserve of Menorca

Pérez Marta, Morris Rebeca, Salord Carlos, Cardona Eva, Carretero Victor, Doblado David, Marsinyach Eva.

In recent years various initiatives have emerged on Menorca to alleviate plastic pollution and in turn. these have contributed in bringing to the forefront this growing problem that is significantly affecting the island's coastal and marine ecosystems, and negatively impacting on some economic sectors. As a result, five entities that have been working for years to reduce the presence of plastic waste on the island have launched an alliance in 2020 in order to join forces against this issue. The Plastic Free Menorca Alliance is made up of GOB-Menorca, the Menorcan Institute for Socio-Environmental Studies-Observatory of Menorca, Per la Mar Viva, the Leader Illa Menorca Association and the Menorca Preservation Fund. Among the experiences that will be presented in the MICRO 2020 communication are; GOB Menorca's Commitment2020 project to reduce SUP being used by neighbors' associations and schools; the experience of the citizens' movement and coastal cleaning activities, which have collected over 13 tons of plastic, run by Per la Mar Viva; the Leader Illa de Menorca Local Action Fishing Group's campaign More fish, less plastic working with local fish markets and fishmongers; the Socio-environmental Observatory of Menorca's research applied to raising awareness campaigns and; the role of the Menorca Preservation Fund as an actor in the promotion of all these initiatives through funding. The citizen response obtained in recent campaigns such as those mentioned here shows that there is a growing social concern. Even so, given the data on waste generation on the island, we are far from translating this concern into tangible changes in local plastic consumption habits and in the processes needed to reduce the impact of plastic pollution on our island. This has led to the creation of the Plastic Free Menorca Alliance.

Keywords : Biosphere Reserve , Menorca , plastic pollution

### CITIZEN INVOLVEMENT IN THE FIGHT AGAINST PLASTIC POLLUTION IN THE BIOSPHERE RESERVE OF MENORCA

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### More than a project, an alliance

Over the last few years, five local non-profit organisations have been working to reduce the presence of plastic waste in the Biosphere Reserve of Menorca and as a consequence of this, in 2020 the five organisations decided to create the *Plastic Free Menorca Alliance*, in order to join forces against plastic pollution. The **Plastic Free Menorca Alliance** is made up of **GOB-Menorca**, the **Socio-Environmental Observatory of Menorca**, *Per la Mar Viva*, the Leader *Illa Menorca* Association and the Menorca Preservation Fund and has 4 main objectives:

- 1. Create a **Plastic Free Certification** for all those business and entities wishing to reduce their use of single use plastics and needing some support and advice to do so.
- 2. Develop a **Guide of local businesses and supplyers offering alternatives to SUP** and sharing that information to citizens and businesses.
- 3. Develop a **Guide on alternatives to SUP** and offer information to citizens, business and others local actors in how they can prioritise which alternatives to choose from.
- 4. Be the **coordination platform** for all local actors and entities that are working against plastic pollution on Menorca to ensure we are aligned and maximising both resources and the impact we are all able to have in this area.

### Five entities, one common goal

The entities that make up the Alliance have been working and continue to work towards a Plastic Free Menorca, both as an alliance and under their own projects. Some of the top results achieved so far include:



GOB Menorca's **Commitment2020NoPlastic project**, has resulted in more than 235.000 single use objects, including cutlery and plastic bags, being replaced by more sustainble alternatives in less than two years, as well as **44 entities and 23 schools** signing up to this commitment.

A volunteer led organisation that in less than 3 years has: collected more than 1 tonne of plastic waste from Menorcan waters; more than 13 tonnes of plastic waste from Menorca's coastline; has carried out educational talks on plastic to over 4,000 people and has created an exhibition called "A sea of Plastics" which has had more than 4,000 visitors in less than 1 year.



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OBSAM has more than two decades of experience in **environmental research** on the Menorca Biosphere Reserve and over the last couple of years have been key to the fight against plastic by carrying out relevant studies to support marine conservation projects, including, Plastic Free Menorca. This includes analysing marine and coastal impacts around the island **IUCN's Plastic Waste-Free Islands Med project** and biological monitoring of our *Posidonia oceanica* meadows.

LEADER's "**More fish, less plastic**" worked with local fishmongers to reduce their use of plastic bags and with fishermen to identify and remove ghost fishing nets. To date the campaign results are: participation of **16 shops**, distribution of **80,000 compostable bags**; **1 fish market** making compostable bags obligatory for all stands, more than **70,000 views** of the campaign video and **6 ghost nets removed**. Separately, LEADER also supported the instalation of a water treatment plant.





The fight against plastic pollution, along with marine conservation, are priority areas of work for the MEPF and to date they have granted just over **150.000** € to support this work and the brilliant local organisations working in this area, including those mentioned above, along with **leading the creation of the Plastic Free Menorca Alliance**.

The citizen response obtained in recent campaigns such as those mentioned here shows that there is a growing social concern around this issue. And yet, we are far from being a plastic free island, with high ratios of waste generation, plastic pollution points to the sea, high presence of plastic in our environment and an excessive use of SUP in the individual and economic sphere. For this reason, we feel the Alliance is an important opportunity to create a common framework between NGO's, entities, business, citizens and local government to make a real change and address the need to urgently and drastically reduce the Menorca Biosphere Reserve's plastic pollution.

In particular in light of the extension of the Menorca Biosphere Reserve's expansion towards the sea, making it the largest marine biosphere reserve in the Mediterranean and providing an essential platform to become a model for sustainability.

# **Beach litter of Karasu Coast in Black Sea (Turkey): Sources, composition, fate and some suggestions**

Yurtsever Meral, Başargan Furkan.

In this study, plastic litter on the coastline in the northeast part of the Marmara Region were examined with a monitoring according to OSPAR. Sampling was carried out in Sakarya province Karasu beach (41.1109175, 30.6945646). The collected wastes were examined in detail one by one. categorised, counted, and ATR-FT-IR Spectrophotometer analyzes were performed to determine the polymer derivative of the wastes. In addition, it is known that recyclable large bottles such as glass bottles are collected by some groups in the examination area. Litter found on the beach was also categorized in terms of erosion and it was determined that the existing litter was mostly eroded-aged litter. In this study, waste with a visible size was taken into consideration. For this reason, when evaluated in terms of size of the plastic litter found on the beach, it is often large enough to be included in the mesoplastic and macroplastic group. The size and aging of the litter found on the beaches; It is also closely related to environmental and meteorological factors such as sunlight, seasonal changes, temperature, wave, wind and etc. In the ATR-FT-IR analyzes, the most common plastic litter types on the beach are; polyethylene, polypropylene, polystyrene, polyethyleneterephthalate, polyvinylchloride, polyurethane, white rubber (LDPE+EVA 7%). In addition, it is noteworthy that in some direct FT-IR analyzes of plastic litter found on the beach, it is found that motor oils (Factory Fil Oil SAE) are stuck on these wastes. Considering the touristic activities of the Karasu district, it is estimated that a large part of the waste on the coastline originates from visitors and tourists, while the other part is transported from the seas to the coastline due to waves and currents.

Keywords : ATR , beach litter , engine oils , FT , IR analysis , marine , oil derivative , Pollution



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MICRO2020 International conference

#### Beach litter of Karasu coast in Black Sea (Turkey): Sources, Composition, Fate and Suggestions

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Study Objective: show an example of microplastic pollution in the Black sea

In this study, plastic litter on the coastline in the northeast part of the Marmara Region were examined with a monitoring according to OSPAR.

Keywords; Pollution, marine, beach litter, ATR-FT-IR analysis, oil derivative, engine oils

#### Materials and methods

Sampling was carried out in Sakarya province Karasu beach (41.1109175, 30.6945646).The collected wastes were examined in de- tail one by one, categorised, counted, and ATR-FT-IR Spectrophotometer analyzes were performed to determine the polymer derivative of the wastes. In addition, it is known that recyclable large bottles such as glass bottles are collected by some groups in the examination area. Litter found on the beach was also categorized in terms of erosion and it was determined that the existing litter was mostly eroded-aged litter. In this study, waste with a visible size was taken into consideration. For this reason, when evaluated in terms of size of the plastic litter found on the beach, it is often large enough to be included in the mesoplastic and macroplastic group.

#### Conclusions

The size and aging of the litter found on the beaches; It is also closely related to environmental and meteorological factors such as sunlight, seasonal changes, tem- perature, wave, wind and etc. In the ATR-FT-IR analyzes, the most common plastic litter types on the beach are; polyethylene, polypropylene, polystyrene, polyethyleneterephthalate, polyvinylchloride, polyurethane, white rubber (LDPE+EVA 7%). In addition, it is notewor- thy that in some direct FT-IR analyzes of plastic litter found on the beach, it is found that motor oils (Factory Fil Oil SAE) are stuck on these wastes. Considering the touristic activ- ities of the Karasu district, it is estimated that a large part of the waste on the coastline originates from visitors and tourists, while the other part is transported from the seas to the coastline due to waves and currents.





- 18% of the collected plastic waste is PE plastic pellets (nurdles)..Of the 338 plastics collected, 102 are microplastic and the others are mesoplastic.
- ATR-FT-IR analysis was performed on plastic particles that were collected in the same way and different polymers were found.

· Mostly Low Density Polyethylene and Polypropylene.

1.	lastic types	Number
1	OW DESINTY POLYETHYLENE (PE-LD)	67
POLYPROPYLENE %10 TALC (PP)		26
POLYPROPYLENE (PP)		8
	OW DESINTY POLYETHYLENE + ETHYLENE VINYL ACETATE (LDPE + EVA %7)	8
BLEND OF POLYPROPYLEN and ETHYLENE PROPYLEN (PP + EPDM)		8
POLYURETHANE - REACTION INJECTION MOLDED POLYURETHANE-GLYCOL (PUR-RIM-GLYCOL)		7
P	OLYPROFYLENE (PP) WITH TIO2.5	6
1	IIGHT IMPACT POLYSTYRENE (HIPS)	5
1	POLYDIMETHYLSILOXAN USP REFERENCE (PDMS)	4
P	OLYVINYL CHLORIDE - PHENOLIC (PVC - P).0	4
P	OLYSTYRENE (FS)	2
Ċ	ALCIUM CARBONATE MILICARB-FILTER	2
1	STYRENE BUTADIENE COPOLYMER (SCB) STYROFLEX 3G 46.1	2
A	CRYLONTRILE STYRENE ACRYLATE POLYAMIDE (ASA + PA) TERBLEND N NM-NM-31.1	1
I	HERMOPLASTIC STYRENE COPOLYMER-ELASTOMER (TPE-S) CAWITON PR 010113.0	1
P	OLYETHYLENE POLYPROPYLENE BLEND (EPR) TAFMER PO\$60.0	1
P	OLYVINYL CHLORIDE - HARD (PVC - HARD).2	1
s	TYRENE-LIQ. MONOMER.0	1
P	OLYDIMETHYLSILOXAN	1
H	IGHT DESITY POLYETHYLENE (PE-HD)	1
÷	HERMOPLASTIC POLYURETHANE (TPU)	1

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MICRO2020, Fate and Impacts of Microplastics: Knowledge and Responsibilities

#### Session 25.5\_O. Chaired by François Galgani, Bastia

**Quantitative detection of fluorescent microplastic particles with a full-format camera for atmospheric transport wind tunnel studies** *Esders Eike Maximilian, Babel Wolfgang, Thomas Christoph.* Paper number 334190

**Hyperspectral imaging as new tool to analyze microplastics** *Faltynkova Andrea, Wagner Martin, Johnsen Geir.* Paper number 334414

**Microplastics distribution in bottom sediments of the Baltic Sea Proper** *Chubarenko Irina, Esiukova Elena, Zobkov Mikhail, Isachenko Igor.* Paper number 334437

**Conventional and biological treatment for the removal of microplastics from drinking water** *Cherniak Samuel, Hermabessier Ludovic, Mckie Michael, Yuan Kaya, Andrews Robert C., Rochman Chelsea.* Paper number 334753

# Quantitative detection of fluorescent microplastic particles with a full-format camera for atmospheric transport wind tunnel studies

Esders Eike Maximilian, Babel Wolfgang, Thomas Christoph.

Atmospheric transport of microplastic particles (MPP) is an emerging topic and we need new methods to improve our understanding of suspension, transport and redeposition of MPP. The atmosphere is not a major source or sink for MPP, but recent findings demonstrate the atmosphere as an important and fast local and global transport pathway. Our objective was to develop a quantitative visual detection method for MPP which can be applied in wind tunnel experiments. Our goals were to A) detect the total number of MPP suspended on a glass carrier, B) detect and exclude clusters of particles and their formation or disbanding, and C) filter noise originating from dust or fragmented particles. A full-frame visual camera was employed in combination with fluorescent MPP excited by ultraviolet light. Images were analysed using a newly developed software: the image gets reduced to its green channel, followed by an intensity filter which sets green pixels below a carefully selected intensity threshold to zero, and to one above it. This 1-bit image was then processed for the total count of all MPP using their size, aspect ratio, and circularity. Given the optical configuration MPP with a diameter of 53 µm are represented by about 200 pixels. We present a robust counting method that can quantitatively detect MPP with high precision (SD of 0.31%) at a small processing time of approximately 4s for an image containing several hundred particles. Size, aspect ratio and circularity can be used effectively to filter noise. The method enabled a robust quantitative detection of fluorescent particles of different diameters down to 53 µm in recent wind tunnel suspension experiments for which we will show the results. Future work will include adjusting the detection method for examining the atmospheric transport of much smaller MPP, different shapes, and a variety of flow conditions.

Keywords : atmospheric transport , microplastic suspension , visual detection , wind tunnel

#### Hyperspectral imaging as new tool to analyze microplastics

Faltynkova Andrea, Wagner Martin, Johnsen Geir.

A central challenge in microplastics (MP) research is the analysis of small particles in an efficient manner. To assess the feasibility and applicability of infrared hyperspectral imaging (HSI) to detect and characterize MPs, we conducted a systematic literature and identified XXX studies applying HSI in a MP context. This shows the early stage of application of the technology. We critically assessed the variety of approaches to HSI including instrumentation, data collection and data handling and compare the performance to best available technologies, including Fourier transform infrared spectroscopy and Raman spectroscopy. HSI was found to be a suitable MP analysis method for particles larger than 250  $\mu$ m, with drastic improvements in analysis time as compared with other methods. Primary challenges are the improvement of spatial resolution to detect smaller MPs and development of robust models for analyzing hyperspectral data. Parameters and methodological details which were applied with success are summarized and recommendations are made for future research. While HSI requires specialization for this new application, it represents a promising technology for MP analysis.

Keywords : hyperspectral imaging , methodology , microplastics analysis

#### Microplastics distribution in bottom sediments of the Baltic Sea Proper

Chubarenko Irina, Esiukova Elena, Zobkov Mikhail, Isachenko Igor.

Concentration of microplastics particles (0.2-5 mm, MPs) in bottom sediments from 3 to 215 m depth is analyzed on the base of 53 samples obtained in 8 cruises of research vessels in the Gotland, Gdansk and Bornholm basins of the Baltic Sea in March-October of 2015-2016. MPs concentration varied from 103 up to 10 179 items/kgDW, with the bulk mean of 863 ± 1371 and median of 530 items/kgDW, showing general increase with water depth. As many as 74.5 % of MPs are fibres, followed by films (19.8%), and fragments (5.7%). In size distribution, maximum is in the range of 1-2 mm (36 % of items). Among 21 polymer types found, dominate Polyethylene (11.1%). Polypropylene (8.3%), and Polymer blend (5.6%). Nearly-homogeneous, weakly increasing and relatively small MPs content (mean 286, median 245 items/kgDW) is found on in coastal zone down to 20 m depth, where strong currents and wave-induced mixing are at work. In intermediate layers (down to 50 m depth), the bulk mean MPs concentration remains the same, but the median drops to 186 items/kgDW, indicating patchy contamination pattern. The highest MPs concentrations (mean 1359, median 837 items/kgDW) are observed in relatively calm zone below the pycnohalocline (70-80 m). Statistically significant correlation between water depth and concentration of fibres is found, which proves that the deep sea is an ultimate sink for synthetic fibres. Distribution of MPs is analyzed in relation to the density stratification, the hydrodynamic zones, surface-wave impact on fine sediments, types of bottom sediments, and the grain size distribution (RSF project No 19-17-00041). Correlation between MPs concentration and type of sediments is not significant, highlighting different dynamic behavior of MPs particles and sediment grains. However, the erosion/transition/accumulation zones still can be distinguished for MPs, and are located slightly deeper than those for fine sediments.

Keywords : bottom sediments , microplastics , the Baltic Sea

# Conventional and biological treatment for the removal of microplastics from drinking water

Cherniak Samuel, Hermabessier Ludovic, Mckie Michael, Yuan Kaya, Andrews Robert C., Rochman Chelsea.

Microplastics have been identified as an emerging threat to drinking water supplies as the risk they pose to consumers is currently unknown. Full-scale studies completed in On-tario, Canada observed removals of 60% following both conventional and membrane-based treatment. Significant changes to the particle size distribution occurred following filtration, with larger particles (>125µm) being more effectively removed than smaller particles (10-45µm). As smaller particles have been observed to translocate across the gut, quantifying overall removal may not be indicative of the health implications associated with microplastic consumption. To improve removal through water treatment the mechanisms of removal regarding particle size and composition needs to be elucidated. This is the first known pilot study which examined the removal of microplastics, specifically evaluating coagulation, ozonation, filtration with two different media configurations (GACor anthracite over sand) and operating conditions (biological or non-biological). The majority of particles observed in this study were small (10-45µm) fibers. Polyester was the most commonly observed plastic; cellulose fibers were hypothesized to be anthropogenically derived. Coagulation was found to provide excellent particle removal (70%), particularly with respect to larger particles (>90% for particles>125µm). Ozonation provided limited particle removal (<10%), and further evaluations are being completed to determine the impact of ozonation on particle transformation. Filtration provided minimal additional particle removal (<25%) for particles 10-45µm; continued evaluations of media type and operational impacts are on-going. The result of this study confirmed the ubiquitous nature of microplastics in treated drinking water and its source. Similar to previous studies it was observed that the size distribution changed through treatment, with smaller particles becoming more prevalent following treatment. The implications of this work will be critical to the subsequent understanding of potential health impacts, and to provide guidance regarding potential monitoring and regulatory requirements.

Keywords : Biofiltration , Drinking Water , Microplastic removal

#### Session 25.5\_Me. Chaired by Mateo Cordier, Guyancourt

High prevalence of plastic ingestion by *Eriocheir sinensis* and *Carcinus maenas* (Crustacea: Decapoda: Brachyura) in the Thames Estuary Mcgoran Alex, Clark Paul, Smith Brian, Morritt David. Paper number 329727

**Study of a liquid soap containing polyethylene microspheres: characterization, quantification, separation and use of** *Acrocomia aculeata* (macaúba) as an alternative abrasive *De Freitas Raí, Dias Ribeiro Bernardo, Viana Marcelo Mendes.* Paper number 331579

**Introducing EUROqCHARM. A H2020 CSA focused on assuring reproducible, harmonised and quality controlled assessments of plastic pollution** *Lusher Amy, Van Bavel Bert.* Paper number 334441

**From SimpleBox4nano to SimpleBox4micro: application to a case study on microbeads** *Quik Joris, Meesters Johannes, Koelmans Albert A.*. Paper number 334604

# High prevalence of plastic ingestion by *Eriocheir sinensis* and *Carcinus maenas* (Crustacea: Decapoda: Brachyura) in the Thames Estuary

Mcgoran Alex, Clark Paul, Smith Brian, Morritt David.

The Thames has a large catchment, 16,000 km2 encompassing 15 million residents and passes through major urban centres e.g., Oxford, Reading and London. Consequently, the Thames is vulnerable to domestic and industrial wastewater pollution. It is one of the most microplasticcontaminated rivers in the UK and this high contamination is likely to have a detrimental effect on wildlife in the river. This study presents evidence for microplastic contamination in two resident species of brachyuran crab from the Thames Estuary: the native shore crab, Carcinus maenas (Linneaus, 1758) and the invasive Chinese mitten crab, Eriocheir sinensis (H. Milne Edwards, 1853). Crabs were sampled periodically (ca. every three months) between December 2018 and October 2019 and their gills, gastric mill and gastrointestinal tract examined. A total of 94 C. maenas and 41 E. sinensis were collected and 874 pieces of plastic recovered, ranging 34 µm–34 mm in length. Overall, 71.3% and 100% of C. maenas and E. sinensis, respectively, contained at least one item (fibre, film, fragment or tangle of fibres) in the gill chamber, gastric mill or gastrointestinal tract. The most common items were fibres (78.5%) but in some cases, particularly in the gastric mill, these were aggregated into tangles (7.8%). Almost all E. sinensis contained tangles of fibres (95.1%). whereas, relatively few C. maenas contained similar tangles (10.6%). This work was supported by the National Environment Research Council [grant number NE/ L002485/1] with co-sponsorship from a Fishmongers' Company's Fisheries Charitable Trust CASE Partnership.

Keywords : Carcinus maenas , crab , Eriocheir sinensis , estuarine , ingestion , Microfibres , Microplastics , plastic load , river , riverine , Thames Estuary , UK

# Study of a liquid soap containing polyethylene microspheres: characterization, quantification, separation and use of *Acrocomia aculeata* (macaúba) as an alternative abrasive

De Freitas Raí, Dias Ribeiro Bernardo, Viana Marcelo Mendes.

Microplastics are particles smaller than 5 mm and larger than 1 µm, which became a global alarm only at the end of the 20th century. Due to the increased public and governmental awareness of several countries, microplastics have found restrictions, and new biodegradable alternatives are being suggested for their replacement. As a way to better understand and assist future studies on the subject, this work aims to seek and raise data regarding an industrial liquid soap that contains polyethylene microspheres in its composition, generically called L, which is used by an aeronautical engine repair company. According to the analyses carried out, it was found that the microplastics resemble microfragments with an average length of  $195.2 \pm 100.9 \,\mu\text{m}$  and an average density of  $0.9416 \pm 0.0004$  g/cm3, being classified as high-density polyethylene (HDPE). By gravimetry, the presence of 4.90% w/w of microplastics was determined in the liquid soap L. Centrifugation and membrane filtration processes (porosity of 0.2 µm) were also performed to separate the microplastics from the soap, confirming the filtration method as the most efficient, which was visually verified by SEM and confirmed by thermal analysis due to the absence of PE fusion. By comparison of the liquid soap L'with another liquid industrial soap, generically called Ä; it was found that the first one had an effective COD higher than the second soap, impacting more on the aquatic environment. Moreover, by adding macaúba to the filtered liquid soap L, a soap with 0.53% of renewable carbon was produced, which differentiates it from the conventional liquid soap L, which has 4.20% of nonrenewable carbon, coming from the polyethylene microspheres, which persist, slowly degrade in the environment and are vectors of pollutants.

Keywords : Macaúba. , Microfiltration , Polyethylene microspheres , Scanning Electron Microscopy , Thermal Analysis

#### **Introducing EUROqCHARM - A H2020 CSA focused on assuring reproducible, harmonised and quality controlled assessments of plastic pollution** Lusher Amy, Van Bavel Bert.

Plastic pollution (macro to nano) has become a global environmental and societal concern. Numerous protocols have been developed to monitor plastic debris, but these are rarely comparable. This has hindered gathering of knowledge regarding pollution sources, development of monitoring programmes, risk assessments and the implementation of mitigation measures. To develop long-term solutions to reduce plastic pollution, it is essential to establish harmonised methodologies. EUROqCHARM, a recently funded H2020 project, will address this by critically reviewing state-ofthe-art analytical methods and validate them through an interlaboratory comparison (ILC) study. The EUROqCHARM consortium consists of 15 partners, a scientific advisory board composed of international experts and a network of more than 25 associated laboratories. This presentation will introduce the project and it's aims for the next three years. EUROqCHARM recognises that harmonisation for large scale monitoring requires flexibility, comparability and reliability. As this is a Coordination and Support Action (CSA), our aim is to provide a cross-Europe and international platform validate several methods for monitoring plastic in the environment and put forward recommendations and standards for monitoring. In more detail, this project aims to identify, test and optimise monitoring approaches through quality assured and rigorously validated methods based on current state-of-the-art techniques which cover all relevant environmental matrices. It will focus on harmonizing and possibly standardisation of methods and reporting formats to facilitate data comparability and meta-level analysis on regional, national and international scales. By including multiple national and international organisations and working groups currently participating in the proposal of harmonisation and standardized methods for research and monitoring, a coordinated and strategic action to bring these key players together, merge working group ideas and facilitate a framework for urgently required procedures for monitoring and assessment of plastics in our environment. This presentation is given by the coordination team on behalf of EUROqCHARM partners.

Keywords : collaboration , harmonization , microplastics

### From SimpleBox4nano to SimpleBox4micro: application to a case study on microbeads

Quik Joris, Meesters Johannes, Koelmans Albert A.

Although field studies have detected microplastics, quantitative estimate of exposure concentrations to nano- and microplastics is lacking, e.g. due to fragmented field campaign's. Furthermore, the detection methods applied are generally not applicable to nanoplastics, making it a big data gap in knowledge on fate and occurrence in the environment. A commonly applied tool in assessing fate and exposure to chemicals and particles is the use of multimedia fate models. Here, we present a study on the fate of microbeads at regional scale using an extension of SimpleBox4nano (www.rivm.nl/simplebox4nano) for use with nano- and microplastics. So called Predicted Exposure Concentrations (PECs) are presented for air, soil, water and sediment compartments. The uncertainty in the PECs is due to uncertainty in the emission estimates and uncertainty and variability of input parameters describing fate of the NMPs. In the air and soil compartments the uncertainty in release rates from microbead used as abbrasives explains the most uncertainty in PECs. For the water and sediment compartment particle density, size and the fragmentation or degradatation rate constant explain most of the variation in PECs. As expected a large effect is found based on density, with low density NMPs largely remaining in the water fase in the fresh water compartment and transported to sea. PECs for sea water show a great sensitivity to the fragmentation or degradation rate, where unrealisticly high PECs are calculated when underestimating this process. This multimedia fate model for nano and microplastics provides a convenient tool for assessing exposure to different types of nano and microplastics at a screening level. Specifically, for nanoplastics this model provides an invaluable asset in future risk assessment studies as there is currently no adequate detection method. Further validation of the SimpleBox4nano model using high quality monitoring data is recommended.

Keywords : microbeads , multimedia fate modelling , simplebox4nano

#### Session 25.5\_Ma. Chaired by Lisa Devriese, Oostende

**Overview of the NOAA Marine Debris Program's funded research** *Uhrin Amy, Herring Carlie.* Paper number 334023

#### **Endless journey of macroplastics in rivers**

Tramoy Romain, Gasperi Johnny, Colasse Laurent, Silvestre Marie, Dubois Philippe, Noûs Camille, Tassin Bruno. Paper number 334180

#### Distribution, composition and abundance of microplastics in the bay of biscay

mendoza amaia, osa juan luis, c. Basurko Oihane, Rubio Anna, Santos María, Gago Jesús, Galgani François, Peña-Rodríguez Cristina. Paper number 334246

#### **Effect of exposure duration on caged blue mussels (***Mytilus edulis***) microplastics bioaccumulation** *Kazour Maria, Amara Rachid.* Paper number 334306

#### Confirmation of microplastic accumulation in habitual usage organic and inorganic fertilizers

*Kannan Kayalvizhi R.* Paper number 334427

#### **Overview of the NOAA Marine Debris Program's funded research**

Uhrin Amy, Herring Carlie.

The NOAA Marine Debris Program (MDP) is committed to supporting the scientific community to improve the global understanding of marine debris. Since 2013, the MDP has administered a federal grant competition for original, hypothesis-driven projects devoted to marine debris research. To date, over 2.6 million dollars has been awarded to 10 institutions in support of 11 projects. In recent years, funding priorities have focused on risk assessment, emphasizing risk to commercial species from ingestion, confinement or entanglement, and fate and transport of marine debris in the coastal zone. Our portfolio currently includes projects addressing the risk of microplastic ingestion to sea scallops, larval blue crabs, black sea bass, and Eastern oysters. In addition, currently funded projects are examining the role that circulation and biological processes at river plume fronts play in the fate and transport of microplastics, how hydrologic regimes impact plastic flux dynamics from river and wastewater sources, and how biofouling influences vertical transport of microplastics. Here, we will provide a brief overview of preliminary findings from a subset of these projects and discuss their broader implications.

Keywords : competitive grants , Marine Debris Program , NOAA , research

#### **Endless journey of macroplastics in rivers**

Tramoy Romain, Gasperi Johnny, Colasse Laurent, Silvestre Marie, Dubois Philippe, Noûs Camille, Tassin Bruno.

Rivers are supposed to be the main pathways of plastics from lands into the Ocean (Lebreton et al., 2017; Schmidt et al., 2017). However, there is still a huge lack of knowledge on how riverine litter, including macroplastics, is transferred into the Ocean. Quantitative measurements of macroplastic emissions in rivers even suggest that a small fraction (0.001 to 3%) of the Mismanaged Plastic Waste (MPW) generated within a river basin finally reach the sea (Emmerik et al., 2019; Schöneich-Argent et al., 2020: Tramov et al. submitted). Instead, macroplastics may remain within the catchment and on coastlines because of complex transport dynamics that delay the transfer of MPW from lands into the Ocean (Olivelli et al., 2020; Weideman et al., 2020). In order to better understand those dynamics, we released 1L plastic bottles equipped with GPS-trackers in the Seine River. Between March 2018 and April 2019, 39 trajectories were recorded in the estuary under tidal influence and 11 trajectories upriver, covering a wide range of hydrometeorological conditions. Results show a succession of stranding/remobilization episodes in combination with alternating upstream and downstream transport in the estuary related to tides. In the end, 100% of the tracked bottles stranded somewhere, for hours to weeks, from one to several times at different sites. In addition, date-prints items found in historical polluted sites in the estuary confirm that plastic may remain stored on riverbanks for decades. The overall picture shows that different physical phenomena interact with various time scales ranging from hours/days (high/low tides) to weeks/months (spring/neap tides and highest tides) and years (seasonal river flow). Thus, the fate of MPW is highly unpredictable with a chaotic-like transfer of plastic debris into the Ocean, and their residence time is much longer than the transit time of water.

Keywords : Flood , Plastic debris , Plastic transport , Residence time , Tides , Tracking

**Distribution, composition and abundance of microplastics in the Bay of Biscay** Mendoza Amaia, Osa Juan Luis, C. Basurko Oihane, Rubio Anna, Santos María, Gago Jesús, Galgani François, Peña-Rodríguez Cristina.

Some studies identify the Bay of Biscay as a marine litter accumulation area (Galgani et al., 1995; Lebreton et al., 2012). In order to verify if this is also the case with microplastics (MPs), an overview of the research studies performed on the abundance of MPs in this marine region is presented, with the aim of addressing the general situation in the different marine compartments, highlighting limitations and knowledge gaps. Eighteen studies were located to date on waters, sediments and biota. The comparative assessment on each compartment reflects high spatial and temporal variability in the distribution of MPs, suggesting seasonal and short term influencing factors. MPs appear in all sampled compartments in a wide range of abundances, presenting in at least 50% of the samples in surface waters and sediments, with 100% occurrence in some studies. Polypropylene, polyethylene, polyester and polystyrene are found most frequently, in agreement with the main polymers reported in worldwide studies, as well as with the main polymers consumed worldwide (GESAMP, 2015). The predominance of fragments and fibres over other kind of shapes of MPs in all sampled compartments (surface water, sediments and biota) suggests that the MP source in the Bay of Biscay is more related to the decomposition of larger plastic waste rather than to direct primary inputs such as pellets or microspheres. Studies show the ubiquitous distribution of fibres in the deep marine environment in the BoB, while studies of beach sediments more frequently report MPs in the shape of fragments. The Bay of Biscay can be considered as medium MPs concentration region. However, monitoring programs are needed to compile data over time to make evaluation of trends possible. The present work highlights difficulties encountered comparing studies due to their differing methodologies. Hence, an urgent consensus is needed to standardise procedures.

Keywords : abundance , Bay of Biscay , characterization , marine , microplastics

# Effect of exposure duration on caged blue mussels (*Mytilus edulis*) microplastics bioaccumulation

Kazour Maria, Amara Rachid.

Bivalves have been used to assess microplastics (MPs) from their surrounding waters and recently. bivalve active biomonitoring was proposed as a potential approach to quantify and assess MPs pollution in the field. However, key questions on the deployment duration and onthe potential use of bivalves as a good proxy for MPs in the aquatic environment are still to be resolved. Here, we investigate the effect of exposure duration on caged blue mussels MPs bioaccumulation. The characteristics of ingested MPs were compared with those found in the surrounding sea surface water. Prior deployment, cultured mussels were depurated and transplanted in the marine coastal waters near an abandoned coastal landfill. Then, MPs bioaccumulation was studied 1, 2 and 5-weeks post transplantation. After 72-h depuration period, 98.8% of the MPs were eliminated. 1-week post deployment, the number of ingested MPs by caged mussels increased significantly to an average of 0.93±0.6 items/g with the presence of fragments and/or fibers. During the second and the fifth weeks of deployment, the number of ingested MPs increased progressively, yet not significantly, and reached a threshold of 1.42 items/g. We found differences in the relative abundance of particle sizes and polymer types between seawater and mussels, with mussels over-representing particles<100µm but under-representing some polymers such as PA and PE. Unlike mussels, the amounts and characteristics of MPs in the surrounding waters showed important temporal variations and seemed to be influenced by environmental factors such as the tidal coefficient. Even if mussels are biased toward small fragments, they are more integrative of MPs pollution than punctual water samples. The present study highlights on a 5-weeks period as a minimum exposure for mussels to reach a bioaccumulation steady-state and emphasizes on using blue mussels caging as a tool for biomonitoring microplastics<200µm.

Keywords : Bioaccumulation , Blue mussels , Caging , Depuration , Microplastics

# Confirmation of microplastic accumulation in habitual usage organic and inorganic fertilizers

Kannan Kayalvizhi R.

Abstract Microplastic is a small piece of plastic that is polluting the environment. The National Oceanic and Atmospheric Administration (NOAA) in U.S. classify microplastics as less than 5mm in diameter and into two categories, like primary microplastics are manufactured as direct of human material product and the secondary microplastics are derived from the breakdown of larger plastic debris. This fragmentation is caused by chemical and physical aging, as well as through biodegradation mechanisms. Microplastics have become a global environmental concern because of their ubiquitous presence. Compared to plastic pollution in the marine and freshwater ecosystems, that in the soil ecosystem has been highly neglected. Terrestrial ecosystems under human influence, such as agroecosystems, are likely to be contaminated by plastic debris. In this present study, different types of organic and inorganic fertilizers are collected from various places to analyze the microplastic accumulation from those fertilizers. The organic fertilizers are biogranules, vermicompost, Huminol-G and Microzyme –G and the inorganic fertilizer like super phosphate, urea and potash, collected both organic and inorganic fertilizers were based on their stereomicroscopic structure and also for quantification and characterization of their chemical nature by FTIR and SEM analysis. Overall the results was shows that those fertilizers having microplastic contamination in considerable levels. Further research is needed to fully reveal the fate and ecological risks of microplastics in agricultural soils and necessary action is required to control microplastic pollution in terrestrial ecosystems.

Keywords : Agricultural soils , FT , Inorganic fertilizer , IR , Microplastics , Organic fertilizer , SEM , Terrestrial ecosystems.

#### Session 25.6\_O. Chaired by Mateo Cordier, Guyancourt

**Microplastics originating from road markings: A significant overestimate?** *Burghardt Tomasz, Pashkevich Anton, Mosböck Harald.* Paper number 334279

**Polystyrene foam as a main carrier of microplastics in the urban river of Mongolia** *Kawahigashi Masayuki, Battulga Batdulam.* Paper number 334378

### Impacts of road runoff: how a cocktail of multiple anthropogenic stressors impact macroinvertebrate communities

*Giles Rachel, Rochman Chelsea, Ruppert Jonathan, Wallace Angela.* Paper number 334542

**Microplastics in riverbed sediments from the largest Amazonian rivers, Brazil** Gerolin Cristiano R., Pupim Fabiano, Sawakuchi André, Grohmann Carlos Henrique, Labuto Geórgia, Semensatto Décio. Paper number 334556

#### **Microplastics originating from road markings: A significant overestimate?** Burghardt Tomasz, Pashkevich Anton, Mosböck Harald.

Numerous literature reports point to horizontal road markings as significant sources of microplastic pollution. There are claims that even 7% of all microplastics originate from road markings and according to a German report, the contribution reaches 19.3-121.1 g/person/year. However, one must consider that these estimates presume that "paint sales data is assumed to equal the paint wear minus new and replacement roads". From the perspective of road marking industry, such assumption is incorrect in vast majority of cases: horizontal road markings are renewed when they lose their retroreflective properties, which occurs before paint is worn and converted into dust. To verify the industrial claims, field testing was undertaken; preliminary results are reported herein. Indeed, samples collected from various roads show the presence of several layers of markings. Furthermore, measurements of retroreflectivity done on several roads confirm that renewals took place before there was a loss of glass beads protecting the colour layer from abrasion. Thus, the reported estimates appear to significantly overrepresent the contribution of road markings to microplastic pollution, at least in cases of well-maintained roads. Understanding of the technology of road markings and their renewal process is seen as critical for accurate estimates of their contribution to microplastic pollution. It must be emphasised that appropriate maintenance of horizontal road markings can essentially eliminate this source of microplastics and modern, durable materials further limit such risk.

Keywords : glass beads , microplastics , retroreflectivity , road marking

# Polystyrene foam as a main carrier of microplastics in the urban river of Mongolia

Kawahigashi Masayuki, Battulga Batdulam.

Plastic debris distributed in and around tributaries affects material cycling and biological activities in the downstream aquatic environment such as shorelines, open oceans and lakes. The distribution and dynamics of plastic debris in riverine environments were influenced by population density, the waste management, and the tributary system. The plastic debris can be transformed during their travel due to physical impacts, solar radiation and/or interactions with other anthropogenic litters, affecting to their interaction with colloidal particles in the river system. Polystyrene foam (PSF) is one of major plastic debris in the urban river in the capital city, Ulaanbaatar, Mongolia. PSF easily fragmented into many units as microplastics (MPs), further scattered to downstream with water flow followed by transformation on its surface and interaction with other colloidal particles. The collected PSFs along the shoreline of the urban tributary were carefully prepared through the process of wet decomposition of natural organic substances followed by sonication and filtration using a membrane filter. Plastics captured onto the filter was analysed using a micro-FTIR to obtain spectra and to identify colloidal particles adhered onto PSFs. The carbonyl index (CI), which is a surface degradation index calculated by IR absorbances, showed wide range values, indicating that surface degradation through oxidation and solar radiation was respectively occurred on distributed PSFs. Number of captured MPs in the view of a digital microscope was reaching to over 100 items on a piece of PSF. Various types of MPs adhered onto PSFs. Although there was no clear relationship between CI and abundances and/or types of adhered MPs, complex of MPs with PSFs can be a common status during their transportation in the urban river. Plastic debris actively interacts with a variety of secondary sourced MPs and travels together through direct adhesion or mediated by colloidal substances in the riverine environment.

Keywords : Adhered microplastics , Adhesion , Colloidal particle , Foamed plastic , Surface degradation

### Impacts of road runoff: how a cocktail of multiple anthropogenic stressors impact macroinvertebrate communities

Giles Rachel, Rochman Chelsea, Ruppert Jonathan, Wallace Angela.

Globally, urbanization continues to increase and produce elevated levels of contaminants (e.g., metals, pesticides) in the air, soil, and water within and around urban centers. In particular, heavily trafficked roads are a major source of anthropogenic contaminants, including polycyclic aromatic hydrocarbons (PAHs), heavy metals, tire dust, and road salt. This mixture, herein referred to as road runoff, represents one of the largest contributors of diffuse-source toxicants in urban areas, and yet is seldom studied as a contaminant mixture. Here we investigate how road runoff could impact ecosystem structure and function in urban stream ecosystems. Specifically, we (1) investigate contaminant fluctuations over winter, a time of intense road salt application, (2) assess communitylevel invertebrate responses to contamination loads in urban streams, and (3) assess the effect of road runoff-associated chemicals to early life stage freshwater mussels. Preliminary results demonstrate that contaminant concentrations (chloride, metals, microplastics) in urban sites are greater than rural sites. Additionally, the amount of chloride has a strong relationship with temperature, where chloride concentrations increase with air temperature during the winter season. Last, we detected differences in community composition of benthic macroinvertebrates between rural and urban sites. Future work will determine how early life stage freshwater mussels respond to environmentally relevant concentrations of each road runoff contaminant both independently and combined in a full factorial experiment. This will provide insight into potential stressors that can impact these and other sensitive species. This study builds on our understanding of how multiple anthropogenic stressors impact stream communities, and has the potential to help inform future policy and urban development decisions.

Keywords : community , invertebrate , level responses , multiple stressors , tire rubber , tire wear particles , urban streams

**Microplastics in riverbed sediments from the largest Amazonian rivers, Brazil** Gerolin Cristiano R., Pupim Fabiano, Sawakuchi André, Grohmann Carlos Henrique, Labuto Geórgia, Semensatto Décio.

The Amazon River is the mainstem of the world's largest fluvial system. Despite the common idea that the Amazon River flows in a remote region, about 20 million people live scattered in its rivers margins in Brazil. The presence of microplastics in riverbed sediment samples collected in seven sites from Solimões, Negro and Amazon rivers, upstream and downstream the metropolitan region of Manaus was investigated. Concentrations ranged from 417 to 8178 parts/kg sediment dw (microplastics: 0.063–5 mm), and from 0 to 5725 parts/kg sediment dw (microplastics: 0.063–1 mm). The highest microplastics concentrations were observed in samples from shallow water (water depth of 5–7 m) sites with lower water velocity of the Negro river surrounding Manaus, and the lowest concentration in farthest sample collected in deeper zone (water depth of 34 m) of the Amazon river around 110 km downstream Manaus. The variation of microplastics concentrations within the studied area can be related to hydraulic characteristics defining the erosive-depositional behavior of the sampling sites and their proximity to Manaus. The sediment samples of the Negro river near Manaus presented the higher microplastic concentrations, probably due the combine effects of hydraulic conditions suitable for sediment accumulation and proximity to the urban area of Manaus. The lower concentrations of microplastics in the Solimões and Amazon rivers are attributed to their more erosive and bypassing hydraulic conditions, especially in deeper zones near the thalweg. These results are the first report to show the ubiquitous presence and widespread distribution of microplastics in sediments from the lower Solimões, lower Negro and upper Amazon rivers. The presence of microplastics in sediments from the largest Amazonian rivers points out that we must advance our knowledge about sources and fate of microplastics to infer their impacts on Amazon ecosystems.

Keywords : Amazon , Microplastics pollution , River sediment

#### Session 25.6\_Me. Chaired by Ana-Carolina Ruiz, Mazatlán

**Electrochemical properties of nanoplastics and their importance in transport through terrestrial groundwater ecosystems** *Müller Sascha, Balic-Zunic Tonci, Posth Nicole.* Paper number 334352

**Microplastics in the sediment of shelves and basins offshore of southern California** *Sarner Rachel, Gray Sarah.* Paper number 334371

**Long-term effect of microplastics on soil microbial community in marine coastal environment.** *Bødtker Gunhild, Hovland Beate, Haave Marte, Andersen Gidske, Bastesen Eivind.* Paper number 334537

### **Electrochemical properties of nanoplastics and their importance in transport through terrestrial groundwater ecosystems**

Müller Sascha, Balic-Zunic Tonci, Posth Nicole.

The subsurface is considered a key sink for plastic polymers in the environment. The presence of preferential flow paths, e.g. along cracks and fractures, but also the variety of geochemical and microbiological processes, however, challenge such a hypothesis. This is especially relevant for nanoplastic (NP) transport, as their physical entrapment (straining) may be less effective in subsurface pore space compared to larger microscale plastics. Accordingly, Nanoplastic particle transport is governed by the groundwater ecosystem chemistry that varies with the geological host material. A complex interplay between the NP particle with its surrounding host media (particleparticle, particle-solvent, particle-porous media) is largely driven by hydrogeochemical and microbiological conditions. Electrochemical properties of both the plastic particles and the host medium serve as crucial factors influencing nanoplastic transport behavior. We here use Zetapotential (ZP) to measure the electrochemical properties of nano-sized polystyrene particles in model groundwater systems under changing chemical conditions (pH, different ions, ion valency, ionic strength). Particular focus is on the influence of surface coatings (i.e., COOH groups) as well as common minerals (i.e., Quarz, Feldspars) and sediment types of the subsurface (i.e., Quartenary sands, Miocene sands). A simple 1D- particle interaction model (DLVO-theory) is used to quantitatively derive potential interaction schemes between NP and subsurface media. Those information can guide us in further upscaling experiments from a 1D space to a 2D space (i.e. column experiments). Taking together, these efforts inform whether NP may be harbored or transported under a variety of subsurface conditions and trace the physical and biogeochemical mechanisms mediating this transport behavior.

Keywords : DLVO , freshwater systems , nanoplastic , nanoplastic groundwater , nanoplastic minerals , nanoplastic transport , zetapotential

# Microplastics in the sediment of shelves and basins offshore of Southern California

Sarner Rachel, Gray Sarah.

Every year, at least eight million tons of plastic enters the ocean with some sinking to the sea-floor. Micro-sized fibers, granules, plastic films and spherules of plastics (microplastics) are a large part of this pollution. The objective of this research was to develop a methodology to determine how the abundance of microplastics varied with distance from shore/population and with water depth offshore of San Diego. Samples were collected on the RV Sally Ride and RV Sproul research vessels in 2018 using a multicorer, which was deployed at water depths ranging from 100 to 960 meters. In addition, sediments were collected in San Diego Bay for comparison. To extract microplastics from the samples, approximately 100mL of sediment from the upper layer (0-1 cm) of the cores was processed by density floatation in Zinc Chloride (density 1.5 g/cm<sup>3</sup>). Floating microplastics were transferred onto a gridded filter and systematically categorized and counted under a microscope. Microplastic fibers were found as deep as 960 meters suggesting that plastic pollution is accumulating in the deep basins of the Southern California continental margin. Ongoing analyses of sediments (and analytical blanks) will determine whether microplastic abundance varies with distance offshore or water depth. A better understanding of the microplastic distributions in offshore sediments will help us better predict the impact of plastics on marine life which inhabit the deep sea.

Keywords : basins , benthic environments , California , fibers , methodology , Microplastics , pollution , ridges , San Diego , sediment , shelves , Southern California

### Long-term effect of microplastics on soil microbial community in marine coastal environment.

Bødtker Gunhild, Hovland Beate, Haave Marte, Andersen Gidske, Bastesen Eivind.

Costal environments are hot spots for accumulation of marine plastic litter. An exposed fjord beach in Tysnes, Norway have received ocean transported plastic since the 1950, which has generated heavily microplastic polluted soil at the backshore of the beach. Because the area is part of a nature reserve attempts are made to restore the microplastic polluted soil by different cleaning methods including soil sifting. As part of the assessment of ecological consequences of cleaning, soil samples will be analysed for microbial community composition by NGS before and at intervals after cleaning, over a period of 5 years. The current study presents results from the baseline before cleaning. Results show that the microplastic polluted soil contain a more diverse community of procaryotes (16S) and fungi (ITS) compared to nearby plastic free soil. Although microplastics and surrounding soil shared many of the same OTUs, soil contained a 15-fold higher number of specific 16S OTUs and 3-fold higher number of specific ITS OTUs. Based on relative abundance of phylogenetically assigned OTUs, bacterial phyla dominant in both soils were Proteobacteria, Planctomycetes, Acidobacteria, Verrucomicrobia and Actinobacteria. Dominant fungal phyla were Ascomycota, Basidiomycota, Rozellomycota and Mortierellomycota in addition to unassigned sequences. The relative abundance of fungi varied between the two soils, with overall higher abundance of Ascomycota, Rozellomycota and unassigned sequences in microplastic soil, and Basidiomycota and Mortierellomycota in control soil. The bacterial composition on microplastics were like surrounding soil at phylum level, with enrichments of plastic-degrading genera Pseudomonas and Bacillus observed on individual microplastics. The microplastics colonised by Pseudomonas also showed enrichment of the fungal genera Mortierella and Cenococcum. The results suggest effects of long-term exposure to microplastics on diversity and composition of soil microorganisms. Monitoring after cleaning will give important knowledge on the recovery rate of natural soils after long-term exposure to microplastic pollution.

Keywords : bacteria , environment , fungi , marine , microbial ecology , microorganisms , Microplastic , NGS , soil

#### Session 25.6\_Ma. Chaired by Cristina Panti, Siena

Does micro-litter ingestion affect the health status of European anchovy (*Engraulis encrasicolus*)?

Rodríguez-Romeu Oriol, Constenla María, Carreras-Colom Ester, Dallarés Sara, Padrós Francesc, Carrassón Maite, Solé Montserrat, Soler-Membrives Anna. Paper number 334389

### Preliminary results of macro and micro debris ingested by loggerhead turtles on the Canary Islands

*Ostiategui-Francia Patricia, Liria-Loza Ana.* Paper number 334477

### Preliminary study on the effects of nanopolystyrene on immune system of a freshwater crayfish *Procambarus clarkii* under low-dose exposure

*Capanni Francesca, Manfrin Chiara, Greco Samuele, Tomasi Noemi, Giulianini Piero Giulio.* Paper number 334530

### Long-term monitoring of microplastics in coastal waters in the Northwestern Mediterranean Sea

De Haan William P., Sanchez-Vidal Anna, Canals Miquel, Amblàs David, Calafat Antoni, Cerdà Marc, Pedrosa-Pàmies Rut, Rayo Xavi, Tarrés Marta. Paper number 334552

### **Does micro-litter ingestion affect the health status of European anchovy** (*Engraulis encrasicolus*)?

Rodríguez-Romeu Oriol, Constenla María, Carreras-Colom Ester, Dallarés Sara, Padrós Francesc, Carrassón Maite, Solé Montserrat, Soler-Membrives Anna.

European anchovy is a fish species of great ecological and commercial importance. It has been suggested as a monitoring species in epipelagic environments and particularly, microplastics ingestion has been reported. However, the assessment of the potential impact of micro-litter (including plastic, non-plastic fibres and other particles) ingestion on wild fish is scarce. Thus, the aim of this study is to identify the potential impact of micro-litter ingestion in the condition and health status of this species. Anchovies from three different localities in the NW Mediterranean (including Barcelona) were sampled during 2019. Percentage of micro-litter occupation in stomach was calculated by morphological technique, classification and identification of micro-litter was performed by visual characterisation and FTIR and fish condition and health status was assessed by a multidisciplinary approach integrating the analysis of morphological fish condition indexes, histopathological assessment and enzymatic biomarkers. Preliminary results showed 52.2% of fish containing micro-litter in their gastrointestinal tract, from which 47.7% corresponded to particles (15.6% to films and 32.1% to fragments) and 52.3% to fibres. Mean intensity was 2.33 (SD = 1.58) items/individual. Particles were mostly of polyethylene (79.4%) and fibres of cellulose (78.9%). Average percentage of occupation of microplastics in the stomach was 2.30% ranging from 0.01% to 9.89%. Mild alterations were detected in some internal organs, including changes in the cytoplasm of hepatocytes, small inflammatory foci and presence of macrophages, most of which are also frequent in other wild fish species. Changes in the morphology of adipocytes of the fat tissue around pyloric caeca were also detected in several fish. No apparent correlation between the presence and amount of ingested micro-litter and fish condition indices, histopathological findings or biomarkers was found. These results point towards an absence of clear impact of the ingested micro-litter and a relatively good health status of the studied anchovy populations.

Keywords : Engraulis encrasicolus , health status , litter , micro , microplastic ingestion , NW Mediterranean Sea.

## Preliminary results of macro and micro debris ingested by loggerhead turtles on the Canary Islands

Ostiategui-Francia Patricia, Liria-Loza Ana.

The European Project INDICIT had proposed sea turtles as marine debris indicator on the European waters. In order to determinate microplastic ingestion by sea turtles, INDICIT project had standardised methodologies for sampling and shared data collected in different regions to conduct global and regional analysis. After the validation of the standard protocols for alive and dead animals, stranded sea turtles in Gran Canaria island from May 2018 to September 2020 has been sampled. On dead turtles, necropsies were carried out and complete digestive track was analysed by sections (oesophagus, stomach and intestine). Digestive content was filtered through train of 2 sieves (5mm and 1mm mesh) to separate macro and micro-debris samples. Micro-debris samples were digested with H2O2 for 24h to eliminate organic material. Live turtles arrived to the recovery centre were placed at individual tanks, feed with eviscerated food to avoid contamination, and water tank sampled for one month. Faeces were filtered with 1mm mesh sieve at each tank discharge all month long just after their arrival to the recovery centre. Faeces samples were treated with "2-seive method", similarly than digestive content treatment described above on dead animals. Debris were classified according to the INDICIT protocols attending size, type and colour. In global, 16 dead and 46 live turtles have been sampled for macro and microdebris presence. Results showed a 100% of litter occurrence on loggerhead turtles stranded on Gran Canaria Island, both macro and microdebris. Most abundant colours are white, black and transparent, whereas the most abundant debris type are sheets, threadlike and fragments. For micro-debris, relevant precautions must be taken specially when sampling live turtles.

Keywords : ingestion , Loggerhead , macrodebris , microdebris
## Preliminary study on the effects of nanopolystyrene on immune system of a freshwater crayfish *Procambarus clarkii* under low-dose exposure

Capanni Francesca, Manfrin Chiara, Greco Samuele, Tomasi Noemi, Giulianini Piero Giulio.

Laboratory trials have suggested that plastic particles Procambarus clarkii, Girard, 1852) is an alien species able to tolerate highly perturbed environments. As benthic detrivore, this species can be susceptible to plastic ingestion. Ten adult specimens of P. clarkii, at intermolt stage, were individually exposed to 100 µg of 100 nm anionic carboxylated polystyrene nanoparticles (PS-COOH NPs) in a 72h acute toxicity test. PS-COOH NPs were embedded in an agarose-based food matrix and administered through diet. At the same time, ten control individuals were fed with agarose-food without NPs. An integrated approach has been conceived to assess the biological effects of nanoplastics, by analysing both transcriptomic and physiological responses. Total haemocyte counts, basal and total phenoloxidase activities, glycemia and total protein concentration were investigated in crayfish haemolymph at 0h, 24h, 48h and 72h to evaluate general stress response over time. Transcriptomes of hemocytes and hepatopancreas have been analyzed to ascertain differentially expressed genes (DEGs) in response to PS-COOH NPs-challenge after 72h. While physiological assays did not show substantial alterations between treated and control groups, the RNA-seq analysis revealed few key differentially expressed genes (DEGs). A total of 12, 98 and 32 DEGs were identified in the hemocytes, female and male hepatopancreas, respectively. Interestingly, genes involved in cell-mediated innate immune responses were up-regulated in the hemocytes. In digestive glands, PS-COOH NPs exposure was related to the down-regulation of genes involved in the lipid metabolism in females and the up-regulation of haemocyanin genes in males. This study suggest that a low concentration of PS-COOH NPs exposure may induce light stress in crayfish.

Keywords : decapoda , nanopolystyrene , physiology , transcriptomic

### Long-term monitoring of microplastics in coastal waters in the Northwestern Mediterranean Sea

De Haan William P., Sanchez-Vidal Anna, Canals Miquel, Amblàs David, Calafat Antoni, Cerdà Marc, Pedrosa-Pàmies Rut, Rayo Xavi, Tarrés Marta.

Microplastics (; 5 mm in length) are currently a major environmental threat to marine and terrestrial habitats worldwide. Tracking the sources and fate of microplastics floating at sea has become challenging as they encompass a complex suite of different properties. The current study aims to explore the main anthropogenic drivers of floating microplastics in the Northwestern Mediterranean Sea by analysing a large spatio-temporal dataset at 6 stations located adjacent (4 km from shore) to the main river catchments and urban areas along the North-Catalan coast. A Manta Trawl (200 and 315 µm-mesh) towed by an inspection vessel was used for sample collection, which allowed an extensive characterization of ¿38,000 floating plastics and microplastics over a 6-year (2014-2016 and 2018-2020) time period. We assessed spatio-temporal differences regarding microplastic size, type, morphology, composition and color, and covered seasonal fluctuations in oceanographic settings, river dynamics and socio-economic factors, such as population density. We discover an exponential increase in plastic pollution in recent years and find maximum concentrations over 5,800,000 items km-2 near Barcelona city adjacent to the Besòs River. We found that whether polyethylene and polypropylene fragments and films are the main types of plastics found along the entire study area, strong evidence shows that polypropylene microbeads also highly contribute to the microplastic budget, suggesting that rivers and waste water treatment plants are one of the primary sources of microplastics, and are likely to be transported from North to South by the Northern Current. Our preliminary results show a significant inter-annual variation linked to the size distribution of microplastics and likely as a function of riverine influx and proximity of industrial and recreational areas. Our study aims at contributing to help us better understand the sources and spatio-temporal variability of microplastics in order to develop relevant policies for mitigation.

Keywords : Coastal , Floating plastics , Inputs , Microplastics , Monitoring

### Session 25.7\_O. Chaired by Andy Booth, Trondheim

## Invasive species and microplastics: Can the invasive mussel *Xenostrobus securis* introduce even more microplastic to the marine foodweb?

*Diem Anna, Cacabelos Eva, Gestoso Ignacio, Lekube Xabier, Canning-Clode João.* Paper number 334511

#### Dispersal protocols in microplastics toxicity studies

*Reilly Katie, Sadler Jon, Lynch Iseult.* Paper number 334527

## Quantifying macroplastic accumulation in water hyacinths using three measuring techniques: a comparative approach

Schreyers Louise, Van Emmerik Tim, Kieu-Le Thuy-Chung, Phung Ngoc Ahn, Thanh Nguyen Luan, Castrop Evelien, Strady Emilie. Paper number 334575

## Microplastics in the remote Polar Regions – close encounters with zooplankton in the surface waters around Antarctica

Jones-Williams Kirstie, Manno Clara, Galloway Tamara, Cole Matt, Stowasser Gabi, Waluda Claire. Paper number 334857

## Invasive species and microplastics: Can the invasive mussel *Xenostrobus securis* introduce even more microplastic to the marine foodweb?

Diem Anna, Cacabelos Eva, Gestoso Ignacio, Lekube Xabier, Canning-Clode João.

Plastic pollution has become an increasing worldwide concern, especially as high quantities of this material enters into the marine environment. Once in the oceans, the plastics fragment into smaller pieces and become available not only to top predators but down to micro-sized plankton. Filterfeeders, i.e. mussels play an important role as they are ubiquitous and seem to filter unselectively. Among mussels, Xenostrobus securis is known to be widespread and invasive, especially in the northern coast of Spain. Until now, only limited knowledge is available on the effects of invasive species and their possible role as a vector for microplastic into the marine foodweb. This study aims to answer 3 questions: Does the native crab Pachygrapsus marmoratus prefer the invasive mussel X. securis or the native Mytilus galloprovincialis? Does the crab predation behaviour change when exposed to different microplastic concentrations? And can X. securis introduce microplastic into the marine foodweb? Different experimental aggregations involving both native and invasive mussels were simultaneously deployed in experimental tanks: 50/50 of M. galloprovincialis and X. securis, 60/30 of M. galloprovincialis and X. securis et vice versa, and aggregations formed by each species separately. All aggregations were exposed to four treatments, involving different microplastics (MP) concentrations, namely only microalgae (i.e., without MP), and three MP concentrations (1000parts/mL, 10.000parts/mL and 100.000parts/mL). After an exposure period of 1 hour, one P. marmoratus was added to each aggregation and predation behaviour was observed. After first visual inspections of the mussels, both species seem to have incorporated microplastics. Preliminary results show that P. marmoratus seems to feed on both species, but prefers the native M. galloprovincialis. The final results will show the difference of microplastic incorporation between M. galloprovincialis and X. securis and, furthermore, if there was a trophic transfer from the mussels to the crab P. marmoratus.

Keywords : Invasive Species , Microplastic , Trophic Transfer

#### **Dispersal protocols in microplastics toxicity studies**

Reilly Katie, Sadler Jon, Lynch Iseult.

With increasing awareness of microplastics in the environment there has also been an increase in laboratory studies to further our mechanistic understanding of the potential toxicity issues that may arise as a result of microplastic exposure. Artificially dispersed microbeads that have previously been used in toxicity studies have recently been shown to have detrimental impacts on the health of the test species and affect the outcome of the test methods due to the surfactants used to disperse the particles. However, if dispersants are not used the hydrophobicity of the plastic surfaces means that they agglomerate and remain at the air-water interface of the solution so no exposure of test organisms in the water column occurs. Taking these factors into consideration, how is it feasible to disperse hydrophobic particles more naturally so that the dispersions are representative of the study environment, with the aim to replicate to the best of our current capabilities the interactions between plastics and test species that could be occurring in the environment? In this study, we used methods currently widely used for nanomaterials research by modifying the testing medium that we used to contain varying amounts on natural organic matter to aid the dispersal of the plastics beads into the testing medium for toxicity assays. Using a range of dispersal methods, including the manufacturers' recommended use of the surfactant TWEEN, we compared the potential for the particles to agglomerate during the testing duration using a combination of methods including Disc Centrifuge Sedimentation (DCS) and Dynamic Light Scattering (DLS) along with TEM imaging. This was supported with acute toxicity assessments with Daphnia magna to determine if a more natural method of dispersal is suitable and adequate when compared to the manufacturers recommended dispersal protocol with surfactants such as TWEEN.

Keywords : daphnia magna , dispersion , surfactants , toxicity

## Quantifying macroplastic accumulation in water hyacinths using three measuring techniques: a comparative approach

Schreyers Louise, Van Emmerik Tim, Kieu-Le Thuy-Chung, Phung Ngoc Ahn, Thanh Nguyen Luan, Castrop Evelien, Strady Emilie.

Recent studies suggest that water hyacinths play an important role in the transport of macroplastics in freshwater ecosystems. Forming large patches of several meters at the water surface, the water hyacinths tend to aggregate large amounts of debris, including plastic items. Research on this topic is still novel and few studies have quantified the role of the water hyacinths in plastic entrapment and transport. In this paper, we present and compare the results from three measuring techniques, characterizing the contribution of water hyacinths in macroplastic accumulation for the Saigon River. Through a field campaign conducted in Ho Chi Minh City, Vietnam, we combine the use of visual counting, physical sampling and aerial imagery from UAVs. This enabled to estimate the number of items per size and polymer type entangled in the water hyacinths, along with density and spatiotemporal variation metrics. We also discuss the benefits and limits of the measuring techniques and new methods to detect macroplastics in water hyacinths. Furthermore, our research informs floating macroplastic litter studies more broadly by exploring the potential of UAV-based approaches for monitoring plastic debris in riverine ecosystems.

Keywords : macroplastics , riverine ecosystems , riverine plastics , water hyacinths

## Microplastics in the remote Polar Regions – close encounters with zooplankton in the surface waters around Antarctica

Jones-Williams Kirstie, Manno Clara, Galloway Tamara, Cole Matt, Stowasser Gabi, Waluda Claire.

This study shines a light on the challenges faced when investigating microplastics in perceived "pristine" regions. Sharing results from samples taken using a hydrobios Microplastics sampling net from the surface waters in the Atlantic portion of the Sub-Antarctic to the Antarctic Peninsula. This region is home to some of the highest concentrations of zooplankton biomass but is also threatened by increasing shipping traffic from fishing and the growing tourism market. It was found that 45.6% of the plastic particles isolated from seawater samples were sampling contamination, originating predominantly from the ship. Whilst we found that micro and mesoplastic concentrations in seawater were significantly low  $(0.013 \pm 0.005n/m^3)$  compared to global averages, they were higher along the Antarctic Peninsula than the open ocean (Sub-Antarctic) stations. The potential availability of micro and mesoplastics to pelagic amphipods was explored, using an observed encounter rate (OER) and a possible encounter rate (PER). The total OER (0.8%) was higher than the PER (0.15%), suggesting that even at low concentrations, microplastics are encountered, and potentially consumed, by amphipods. This study highlights the need to prioritise regions of high zooplankton abundance and to investigate both water and biota to build up a picture of plastic pollution and its potential interaction with the Antarctic Ecosystem.

Keywords : amphipods , encounter rate , polar , surface water , synthetic

### Session 25.7\_Me. Chaired by Richard C. Thompson, Plymouth

**Global review about status of plastic presence in marine biota** *Rivas Marga L., López-Martinez Sergio.* Paper number 333015

#### Plastic addtive transfer to fulmars

Kühn Susanne, Booth Andy, Sørensen Lisbet, Van Oyen Albert, Van Franeker Jan. Paper number 333210

## Interacting effects of simulated eutrophication, temperature increase, and microplastic exposure on *Daphnia magna*

Hiltunen Minna, Vehniäinen Eeva-Riikka, Kukkonen Jussi. Paper number 334192

#### Elucidation of the formation mechanism of microplastics

*Kuroda Shin-Ichi*. Paper number 334589

#### National reconnaissance of common plastics in Australian biosolids

Okoffo Elvis, Tscharke Benjamin J., O'brien Jake W., O'brien Stacey, Ribeiro Francisca, Burrows Stephen, Choi Phil M., Wang Xianyu, Mueller Jochen F., Thomas Kevin V.. Paper number 334601

#### **Global review about status of plastic presence in marine biota**

Rivas Marga L., López-Martinez Sergio.

The presence of plastic in the environment is generating impacts on all habitats and has become a major global problem in marine megafauna by macroplastics, through entanglement, ingestion and loss of suitable habitats. In addition to entanglement problems, there is evidence that plastics are entering the food web through their intake by marine organisms, which could ultimately be affecting humans. Much of the available information on the impact of plastic in biota is scattered and disconnected, as different methodologies are used. Here, we review the different approaches and protocols followed to assess macro and micro plastic ingestion in marine vertebrates such as sea turtles, cetaceans and fish in order to offer 28 a global overview of their current status. The analyses of 112 studies indicate the highest plastic ingestion in organisms collected in the Mediterranean and Northeast Indian Ocean, and significant differences among the prevalent polymers, colours an dplastic-types ingested by different species. In sea turtles, it was predominant white plastics (66.6 %), fibers (54.54 %) and LDPE polymer (39.09 %); in cetaceans, white macro and micro plastics (38.31%), fibers (79.95%) and PA polymer (49.6%); and in fish, transparent plastics (45.97%), fibers 66.71% and polyester polymer (36.2%).

Keywords : apical consumers , global change , macroplastic , marine conservation , microplastic , predators.

#### **Plastic addtive transfer to fulmars**

Kühn Susanne, Booth Andy, Sørensen Lisbet, Van Oyen Albert, Van Franeker Jan.

The transfer of plastic additives to stomach oil of northern fulmars (Fulmarus glacialis) was investigated. Procellariiform seabirds retain oily components of their prey in theirs stomach as a means to store energy. In an experiment, a marine litter-derived microplastic reference mixture and separately a marine litter-derived polystyrene sample were added to stomach oil. A total of 15 additives, including plasticizers, antioxidants, UV stabilizers, flame retardants, and preservatives were identified in the original plastic mixtures, and monitored in the leachates. These substances include those known for endocrine disruptive, carcinogenic and/or other negative effects on organisms. Five of the monitored substances were shown to strongly leach from the microplastic reference mixture into the stomach oil during the experiment. Four substances were identified in a marine litter-derived polystyrene foam, of which two leached into stomach oil. Leaching of harmful plastic additives to the stomach oil of fulmars may be of concern, as fulmars regularly ingest plastics which are retained and gradually ground in the gizzard before passage to the intestines and excretion.

Keywords : additive leaching , Fulmarus glacialis , gastric fluid , marine litter , plastic ingestion

## Interacting effects of simulated eutrophication, temperature increase, and microplastic exposure on *Daphnia magna*

Hiltunen Minna, Vehniäinen Eeva-Riikka, Kukkonen Jussi.

The effects of multiple stressors are difficult to separate in field studies, and their interactions may be hard to predict if studied in isolation. We studied the effects of decreasing food quality (increase in cyanobacteria from 5 to 95% simulating eutrophication), temperature increase (by 3°C), and microplastic exposure (1% of the diet) on survival, size, reproduction, and fatty acid composition of the model freshwater cladoceran Daphnia magna. We found that food quality was the major driver of Daphnia responses. When the amount of cyanobacteria increased from 5 to 95% of the diet, there was a drastic decrease in Daphnia survival, adult size, and reproduction but the decrease was not always linear. This was most likely due to lower availability of lipids, eicosapentaenoic acid (EPA), and sterols from the diet. Exposure to a mix of secondary microplastics did not affect Daphnia survival, size, or reproduction. Food quality had an interactive effect with temperature on fatty acid content of Daphnia. Total fatty acid content of Daphnia was almost 2-fold higher at 20°C than at 23°C when fed 50% cyanobacteria. This may have implications for higher trophic level consumers, such as fish, that depend on zooplankton for energy and essential lipids. Our findings suggest that as proportions of cyanobacteria increase, in tandem with water temperatures due to climate change, fish may encounter fewer and smaller Daphnia with lower lipid and EPA content.

Keywords : cladocera , climate change , cyanobacteria , fatty acids , food quality , zooplankton

#### **Elucidation of the formation mechanism of microplastics**

Kuroda Shin-Ichi.

The purpose of this study is to clarify by what mechanism and from what kind of plastic products the secondary microplastics are generated from the perspective of polymer science and polymer engineering. Various approaches of us lead the following results. In the analysis using the pyrolysis method, individual component information was successfully extracted from the plastic mixture samples by means of Pv-APGC-MS measurements with selecting the ions characteristic of the object polymer and recording the ion chromatogram with high mass resolution. In the morphological analysis of secondary microplastics, the microplastics recovered from rivers and beaches showed cracks as in the outdoor-exposed polymers, where light-induced oxidation takes place. On the other hand, no crack was observed in the microplastics recovered from marine. In a study considering microbial corrosion, the "ABC degradation" mechanism was revealed: it was found that a complex degradation takes place where the hydrophilization of polymer proceeds owing to the photooxidative degradation (Abiotic) and microbial corrosion (Biotic), followed by the micro-flake detachment (Cutting) due to the dissociation of the hydrophilic zone caused by water ingress. Chemiluminescence analysis shows that hydrophobic polyolefins such as HDPE and PP tends to decelerate the degradation owing to the suppression of temperature increase and oxygen diffusion due to the presence of seawater. Focusing on mechanical effects, it is was found that fatigue may cause the fragmentation of plastic materials and that the jet mill could efficiently fine-grind plastics. In the future, a database of literature and research results on the formation mechanism of microplastics will be build. In addition, the microplastic formation behavior through long-term outdoor exposure will be observed and the microplastic accelerated generation test methods will be developed.

Keywords : accelerated generation , APGC , cracks , formation mechanism of secondary microplastics , hydrophilic , ion chromatogram with high mass resolution , microbial corrosion , microplastics , MS , oxidative degradation , photo , Py

#### National reconnaissance of common plastics in Australian biosolids

Okoffo Elvis, Tscharke Benjamin J., O'brien Jake W., O'brien Stacey, Ribeiro Francisca, Burrows Stephen, Choi Phil M., Wang Xianyu, Mueller Jochen F., Thomas Kevin V..

The extent and scope of how plastics may be introduced into soil systems is not fully understood, however it has been recognised worldwide that they can be introduced into soils through the practice of applying biosolids (treated sewage sludge) to agricultural land. Plastics entering the soil environment through the land application of biosolids have aroused increasing concern as they pose potential threats to soil organisms and health. Although studies have tried to quantify plastics in biosolids, samples usually suffer from an extensive clean-up process to remove organic materials before plastic particles are separated, counted and identified using spectroscopic techniques such as Raman and Fourier-transform infrared spectroscopy. These techniques are size dependent and in many cases are not able to detect nano-sized plastics leading to underestimation. In this study, biosolids samples collected from WWTPs across Australia were analysed to quantify the mass concentrations and profiles of seven common plastics: - PS, PC, PMMA, PP, PET, PE and PVC in biosolids. Plastics were quantified using a validated quantitative method consisting of pressurized liquid extraction (PLE) followed by double-shot pyrolysis gas chromatography-mass spectrometry (Pyr-GC/MS) (Okoffo et al. (2020) to provide the first per-capita mass loads release of these plastics and to evaluate the mass of plastics discharged into the Australian environment. PE was the predominant plastic detected (median, 7.8 mg/g dw), contributing to 69% of the total of all plastics. The results of this work provides a first quantitative estimate of emission of plastics through biosolids end-use in Australia. Reference Okoffo, E. D., Ribeiro, F., O'Brien, J. W., O'Brien, S., Tscharke, B. J., Gallen, M., ... & Thomas, K. V. (2020). Identification and quantification of selected plastics in biosolids by pressurized liquid extraction combined with double-shot pyrolysis gas chromatography-mass spectrometry. Science of The Total Environment.

Keywords : Australia , Biosolids , MS , Plastics , Pyr GC , Quantification

### Session 25.7\_Ma. Chaired by Winnie Courtene-Jones, Plymouth

**Correlative microscpectroscopy for reliable microplastic particle identification** Brandt Josef, Bittrich Lars, Fischer Franziska, Fischer Dieter, Kanaki Elisavet, Hassellöv Martin, Mattson Karin. Paper number 334394

Marine litter and recyclable waste collection: a case study in the Patos Lagoon – RS, Brazil Ramos Bruna, Proietti Maira. Paper number 334410

**Distribution of current-use pesticides and personal care products sorbed on floating plastics in Ebro Delta and Mar Menor lagoon waters** García-Pimentel María del Mar. Paper number 334418

**Remediation of marine microlitter: proposing photocatalysis technology for the removal of HDPE and PS microplastics from polluted water before its arrival to the oceans** *Cedillo-González Erika, Cristina Siligardi.* Paper number 334574 **Correlative microscpectroscopy for reliable microplastic particle identification** Brandt Josef, Bittrich Lars, Fischer Franziska, Fischer Dieter, Kanaki Elisavet, Hassellöv Martin, Mattson Karin.

Fast and reliable methods for the identification of microplastic (MP) particles in environmental samples are key to a better understanding of sources, pathways and sinks of MP in the environment. Precise knowledge over particle type and size allows drawing conclusions on the impact on the ecosystems the particles are found in. Automated microspectroscopy techniques, such as Micro Fourier Transform Infrared (FTIR) or Micro Raman spectroscopy are conventionally used to determine number, size and chemical classification of MP particles. Both techniques have their advantages and disadvantages; none can be clearly be put in favor over the other. The self-developed software package GEPARD [1] allows combining different microscopy techniques in a correlative manner to exploit the advantages of each method, whilst minimizing analysis effort and time. With GEPARD, a particle based analysis approach is followed, entailing the following steps: (i) Optical image acquisition, using either the built in cameras in microspectrometers or a dedicated light microscope, (ii) automated image segmentation for particle recognition, (iii) spectroscopic scan of the detected particles with either Raman, or FTIR or also both, (iv) spectrum matching using spectra databases and (v) result reviewing and reporting. GEPARD creates and saves comprehensive datasets for each sample, allowing to measure subset of particles with different techniques. E.g., large particles are measured first with FTIR, then small particles with Raman and finally Raman with a different laser for particles showing strong fluorescence. GEPARD's flexible workflow allows for an optimal utilization of each analysis lab's equipment configuration, while generating consistent data sets that can be analyzed independently of any hardware requirements. This opens up novel pathways of data mining (e.g., using machine learning) to maximize the level of information gained from valuable samples. [1] Brandt, J. et al. doi: 10.1177/0003702820932926.

Keywords : correlative microscopy , FTIR , microplastic analysis , microspectroscopy , Raman

## Marine litter and recyclable waste collection: a case study in the Patos Lagoon – RS, Brazil

Ramos Bruna, Proietti Maira.

About 80% of the marine litter present in coastal environments is the result of anthropogenic activities undertaken on land. Solid waste sorting and recycling plays an important role in the waste management chain, as well in society's effort to solve the marine litter problem. When recyclable waste collection fails, its destination is jeopardized and the chances of it being transported through watersheds to the ocean increases, especially under conditions of high rainfall and wind. This study aimed to qualitatively analyse the recyclable waste collection of three municipalities in the Patos Lagoon complex - RS (Porto Alegre, Pelotas, and Rio Grande) that are relevant in terms of population and economy. Data on recyclable waste collection coverage and efficiency were obtained from the Brazilian Institute of Geography and Statistics (IBGE), using waste sorting sustainability indicators (Fechine & Moraes 2015), and a SWOT matrix where we highlight strong and weak points in each city. The state capital, Porto Alegre, presented the most efficient selective collection, but far from ideal; Rio Grande presented the greatest weaknesses, related to the lack of public policies and investment in infrastructure, a situation explained, in part, by the exogenous development of the municipality and multiple coastal zone uses. This result corroborates with data from a local marine litter project (Projeto Lixo Marinho), which found 60% of land-based material at Cassino Beach, Rio Grande. Much of the litter found on the beach could have entered the recycling chain but lost its value upon entering the beach environment. Recyclable waste collection efficiency can be considered an indicator for monitoring land-based marine debris. However, even if recyclable waste collection covers the entire municipal demand, popular participation, reverse logistics initiatives, and shared responsibility are essential for the reduction of marine litter in coastal cities and adjacent bodies of water.

Keywords : Hydrographic Basin , IBGE , solid waste management , SWOT Matrix

## Distribution of current-use pesticides and personal care products sorbed on floating plastics in Ebro Delta and Mar Menor lagoon waters

García-Pimentel María Del Mar.

Plastic acts as passive samplers of organic and inorganic contaminants from the surrounding environment (air, water, soil, sediment, etc) (León et al., 2018) and can be used as integrative matrix for these contaminants in the marine environment. Previous studies have confirmed the concentration of organic contaminants in continental (León et al., 2018) and beached plastics (León et al., 2019) along the SE Iberian coast, but no information is available in relation to their concentrations in floating plastics. In this study the occurrence and distribution of current-use pesticides (CUPs), personal-care products, plastic additives, polycyclic aromatic hydrocarbons (PAHs) and organochlorinated contaminants were characterized in floating plastics sampled in 2019 in surface waters from Ebro Delta and Mar Menor lagoon. The polymeric nature of plastic debris was characterized by attenuated total reflection Fourier-transform infrared spectrometry. Contaminants were extracted from plastic following the proposed method by León et al., (2019). This method consists of three successive ultrasonic extractions with MeOH and the analysis of the extract by stirbar sorptive extraction coupled to gas-chromatography with mass-spectrometry. The presence and distribution profiles of these contaminants was consequence of the predominant anthropogenic pollution sources in each coastal ecosystem (tourism, agriculture, urban nuclei and transport) and some differences were found in their distribution profiles depending on the considered polymer and the physic-chemical properties of the substances.

Keywords : care products , Coastal ecosystems , Current , Personal , Plastic additives , Plastic debris , use pesticides

# Remediation of marine microlitter: proposing photocatalysis technology for the removal of HDPE and PS microplastics from polluted water before its arrival to the oceans

Cedillo-González Erika, Cristina Siligardi.

Currently, our seas are polluted by marine litter. Marine litter is principally composed of plastic. Big items such as nets, bags, and bottles are causing entanglement and asphyxia of marine fauna. Their smaller equivalents, known as microplastics (MPs), are defined by the United Nations as small plastic particles of diameter  $\leq$  5 mm. A full gamma of marine species is consuming MPs. Of more significant concern is that, due to their small size and high surface area, microplastics can adsorb persistent organic pollutants (POPs), transferring them throughout the tropic chain. The solutions to microplastics environmental problem that has been already proposed include plastic recycling, environmental education and implementation of politics for reducing plastic waste generation. However, none of these proposals represents a sustainable solution for microplastic marine pollution. In this work, we propose visible light photocatalysis using N-TiO2, C,N-TiO2 and C,N-TiO2/SiO2 semiconductors for the chemical removal of primary high-density polyethylene (HDPE) and polystyrene (PS) MPs from polluted water. The photocatalysts were characterized by XRD, DRS, FTIR, BET, and FEG-SEM. The HDPE microplastics were characterized by FTIR and optical and electron microscopy. Photocatalysis was performed in aqueous media in order to facilitate the future scaling of the process into wastewater treatment plants. The influence of pH, temperature, semiconductor's form and MPs' size on the photocatalytic removal of MPs was tested. The removal process was monitored by mass loss, FTIR, carbonyl index and optical microscopy. Photolysis was also performed at the same conditions, without semiconductors. From the obtained results, it was found that at the best conditions, removal of up to 70% of the original MPs is possible. Thus, it was demonstrated that photocatalysis of MPs in an aqueous medium is possible, providing an alternative solution to marine microlitter.

Keywords : microlitter , photocatalysis , polyethylene , polystyrene , remediation , removal microplastics

### The Marine Strategy Framework Directive perspective: a panel conversation. Chaired by Bethany Jorgensen, Ithaca.

We are pleased to invite you to join 5 members of the European Commission Technical Group on Marine Litter Microlitter:

Georg Hanke; Elke Fischer; Gunnar Gerdts; Jakob Strand and Francois Galgani.

For 57 minutes, **in Gaia's Room**, they will share the most recent Updates and offer a panel conversation about the ongoing efforts of the Marine Strategy Framework Directive.

**Poster session 25.8** 

### Session 25.8\_O. Chaired by Miguel Tamayo, Madrid

Model polyethylene nanoparticles for the study of (nano)plastics in the ocean Brunel Fabrice, Lansalot Muriel, Bourgeat-Lami Elodie, Monteil Vincent. Paper number 333194

Elimination of nanoplastics from water by by Photocatalytic process: a preliminary study Robert Didier, Alle Paul-Henri, Garcia Munoz Patricia, Keller Nicolas. Paper number 334100

Microplastics contribution to the marine environment through streams in Fildes Peninsula (King George Island, Antarctica) De Feo Bárbara.

Paper number 334440

#### The influence of iron (oxyhdr)oxides on the surface properties of polystyrene in aquatic environments

Schmidtmann Johanna, Ottermann Katharina, Helfricht Nicolas, Papastavrou Georg, Peiffer Stefan. Paper number 334127

**Model polyethylene nanoparticles for the study of (nano)plastics in the ocean** Brunel Fabrice, Lansalot Muriel, Bourgeat-Lami Elodie, Monteil Vincent.

Polyethylene (PE) is the most mass-produced polymer, it represents 90% of the plastic waste accumulated in the center of oceanic gyres (i.e. plastic continents).[1] Following their photochemical degradation, these waste are gradually fragmented into micro-plastics (; 5 mm) and potentially into nano-plastics (; 1 µm).[2-3] Due to their small size, micro / nano-plastics are easily integrated into the food chain with possible toxic effects. [4-5] However, most biological and physico-chemical studies on micro-plastics are carried out with polystyrene nanoparticles because it remains difficult to synthesize PE nanoparticles. Model PE dispersions can be obtained by emulsification of the molten polymer, but this process produces particles of high size and dispersity.[6-7] In the presence of surfactant (cationic or anionic) the nanoparticles obtained are anisotropic and have a very strong supercooling. Temperature-resolved synchrotron scattering was used to propose a novel mechanism explaining the origin of this enhanced supercooling in accordance with the evolution of the particle morphology and surface properties.[6] Our work is currently focused on the synthesis of PE nanoparticles using various stabilization in order to better control their size, morphologies and surface charge. The use of neutral initiator and surfactant allowed us to obtain spherical nanoparticles of controlled size (10 ; D ; 1000 nm) and of low polydispersity and therefore to extend the size range accessible via this synthesis process. These dispersions are well suited for studying the effects of micro / nano-plastics of PE in the environment. [1] Ter Halle et al. (2017) Environmental Pollution 227: 167-174. [2] Gigault et al. (2016) Environmental Science: Nano, 3(2), 346-350. [3] Ter Halleet al. (2017) Environmental science & technology, 51(23), 13689-13697. [4] Grau et al. (2010) Angewandte Chemie International Edition, 49(38), 6810-6812. [5] Billuart et al. (2014) Macromolecules, 47(19), 6591-6600. [6] Brunel et al. (2017) Macromolecules, 50(24), 9742-9749.

Keywords : nanoparticle , polyethylene

## Model polyethylene nanoparticles for the study of nanoplastics in the oceans

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#### Introduction

Polyethylene (PE) is the most mass-produced polymer, it represents 90% of the plastic waste accumulated in the center of oceanic gyres (i.e. "plastic continents").[1] Following their photochemical degradation, these waste are gradually fragmented into micro-plastics (< 5 mm) and potentially into nano-plastics (< 1  $\mu$ m).[2-3] Due to their small size, micro / nano-plastics are easily integrated into the food chain with possible toxic effects. [4-5] However, most biological and physico-chemical studies on micro-plastics are carried out with polystyrene nanoparticles because it remains difficult to synthesize PE nanoparticles. Model PE dispersions can be obtained by emulsification of the molten polymer, but this process produces particles of high size and dispersity.[6-7]



#### Relationship between surface charge, crystallinity and morphology

In the presence of surfactant (cationic or anionic) the nanoparticles obtained are anisotropic and have a very strong supercooling. Temperature-resolved synchrotron scattering was used to propose a novel mechanism explaining the origin of this enhanced supercooling in accordance with the evolution of the particle morphology and surface properties.[6] Our work is currently focused on the synthesis of PE nanoparticles using various stabilization in order to better control their size, morphologies and surface charge. The use of neutral initiator and surfactant allowed us to obtain spherical nanoparticles of controlled size (10 < D < 1000 nm) and of low polydispersity and therefore to extend the size range accessible via this synthesis process. These dispersions are well suited for studying the effects of micro / nano-plastics of PE in the environment.

Average diameter (nm)	PDI	ζ-potential (mV)
47.22	0.14	-73.2
45.44	0.2	-25.2
123.7	0.12	-0.7
71	0.146	24.8
36.19	0.303	51.7

Wide range of surface charges obtained by varying the initiator and the surfactant used during polymerization (anionic, neutral or cationic).

Surfactant exchange by dialysis after polymerization to tune both the charge and the size of the nanoparticles.

#### Perspectives

Synthesis of labeled PE nanoparticles (with <sup>13</sup>C or with a fluorophore) to better overcome the detection problems (by 2D NMR <sup>13</sup>C-13C or by fluorescence microscopy) and study their bioaccumulation depending on their size and their surface properties

Physico-chemical study and modeling of their fragmentation and their aggregation in a complex medium (in presence of salt, extracellular polysaccharide produced by microalgae and inorganic particles) could be performed to simulate the natural processes encountered in the ocean.

#### **References :**

- [1] Ter Halle et al. (2017) Environmental Pollution 227: 167-174.
- [2] Gigault et al. (2016) Environmental Science: Nano, 3(2), 346-350.
- [3] Ter Halleet al. (2017) Environmental science & technology, 51(23), 13689-13697.
- [4] Grau et al. (2010) Angewandte Chemie International Edition, 49(38), 6810-6812.
- [5] Billuart et al. (2014) Macromolecules, 47(19), 6591-6600.

## Elimination of nanoplastics from water by by Photocatalytic process: a preliminary study

Robert Didier, Alle Paul-Henri, Garcia Munoz Patricia, Keller Nicolas.

Many household wastewaters contain Microplastics (MPs) and nanoplastics (NPs). Most of this type of pollution is not stopped by current wastewater treatment plants provoking their entrance into the global aquatic systems. This is an emerging issue and potential threat to marine life and human health. However, advanced technologies for efficient MPs and NPs control and elimination remain largely underdeveloped. Heterogeneous photocatalysis represents an attractive and efficient decomposition technique for the nanoplastic particle degradation. The first objectives of this work were i) to study the feasibility of nanoplastic degradation by photocatalysis and ii) to determine the influence of certain reaction parameters i.e. the flowrate, the pH0 and the light intensity on the photocatalytic degradation of calibrated polymethylmethacrylate (PMMA) and polystyrene (PS) nanoparticles with TiO2-P25/β-SiC foams under UV-A. The first results are very encouraging, because to our knowledge, we are the first to show that it is possible to mineralize PMMA and PS nanobeads by UV-A photocatalysis. We were able to convert in 7 hours, 50% of an aqueous suspension loaded with PMMA nanobeads (TOC = 12 mg/l) by working at an irradiance of 112 W/m2, with a flowrate of 10 mL/min and at initial pH value of 6.3, using TiO2-P25/β-SiC foams in a flow-through mode. We also showed that the photocatalytic treatment can be applied to various polymers, such as polystyrene and PMMA.

Keywords : nanoplastic , photocatalysis , PMMA , PS , water treatment











## Elimination of nanoplastics from water by Photocatalytic process: a preliminary

### study

#### Paul Henri ALLÉ<sup>1,2</sup>, Patricia GARCIA-MUÑOZ<sup>3</sup>, Kopoin ADOUBY<sup>2</sup>, Nicolas KELLER<sup>1</sup>, Didier ROBERT<sup>1</sup>

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#### **INTRODUCTION and PROBLEMATIC**

Nowadays, we can find micro- and nano-sized plastics (MPs and NPs) worldwide in coastal regions and aquatic ecosystems transported by wind and ocean currents. The potential impacts of micro and nanoplastics present in products on the (aquatic) environment and possibly on the human health have generated concerns in worldwide

In the past decade, there has been a major change in the pollution potential for nanoplastic pollution in the oceans, with the shift from natural exfoliators to nanoplastics in skin cleansers. Consequently, certain NPs such as nanobeads derived from toothpaste and facial cleanser are directly introduced into wastewater by human activities (fig 1).



#### **EXPERIMENTAL**

Photocatalytic experiments were performed in recirculation mode photoreactor. Three TiO2-P25/β-SiC foams (diameter 38 mm; length 30 mm) were inserted in the tubular guartz reactor (diameter 40 mm; length 200 mm, volume 250ml) surrounded by 2 to 4 UV-A lamps (1 cm between the quartz tube and the lamps). Each foam sample (10 g) contains  $10 \pm 1$  wt% of TiO2. A volume of 600 ml of aqueous nanoplastic suspension to be treated is introduced into the system.



Fig. 2. Photocatalytic tubular reactor made with (1) a 4 UV-A lightning system + a quartz tube + 3 TiO2/8-SiC foams, (2) polymer nanobeads solution and (3) a peristaltic pump.



PROPOSED SOLUTION

Different methods of nanoplastic removal in wastewater include sorption and filtration, removal based on chemical phenomena, biological ingestion treatments and advanced oxidation technologies (AOTs) has been proposed. Among (AOTs) able to remove nanoplastics from wastewater, heterogeneous photocatalysis represents an attractive and efficient technique for the degradation of these particular pollutants.

TiO<sub>2</sub> is one of the most studied semiconductors due to its non-toxicity, high chemical stability (particularly to photocorrosion) and photocatalytic efficiency. Photocatalytic removal of organic pollutants consists in illuminating TiO<sub>2</sub> with UV-light, to produce hydroxyl and superoxide radicals.

In this work, we use a  $\beta$ -SiC foam supported TiO\_2-P25 photocatalyst integrated into a flow-through photoreactor for nanoplastics removal in wastewater. We took calibrated polymethylmethacrylate (PMMA) and polystyrene (PS) nanobead particles as model systems for microplastic pollutants. The photocatalytic performances have been evaluated by analyzing the time-evolution of the total organic carbon (TOC) upon UV-A irradiation. The influence of some important operating parameters (ie. pH, flowrate, incident irradiance in the photoreactor, particles size...) on the photocatalytic removal of PMMA and PS nanobeads has been studied in order to optimize the process.

#### ANALYSIS

During the process under irradiation, 10 mL of solution were taken at regular time-intervals to evaluate the Total Organic Carbon without filtration. The TOC measurement is not directly related to the concentration of polymer nanobeads during irradiation. However, it can demonstrate the transformation of polymer nanoparticles under the action of photocatalysis.

TOC measurements were determined by a Shimadzu TOC-L apparatus.



The first preliminary results showed that it is possible to oxidize PMMA and PS nanobeads in the presence of  $TiO_2$ -P25/ $\beta$ -SiC foam FLT: TOC (mg/L) (J/bm 202 TIO\_-P25/B-SIC (dark) 30 mL/mi 50 mL/mi TIO\_-P25/8-SIC/UY-A A

Fig. 3. a) Mineralization kinetic of NBs-PMMA on TiO2-P25/6-SiC without and under UV-A irradiation = 10 mL/min, pH0 = 6.3); b) Flow rate effect on the photocatolytic degradation of NBs-PMMA soluti = 6.3). A pseudo-zero order model is applied to the TOC evolution data set. n and photolysis (Flow on on TiO2-P25/8-SiC (

t (min)



Fig.4. a) pH influence and b) UV-A light intensity influence on the photocatalytic mineralization of NBs-PMMA using TiO2-P25/8-SiC foams (flow rate = 10 mL/min). Lines show the pseudo-zero order model fitted to each data set.

#### **RESULTS AND DISCUSSION**

- > The reaction follows a zero-order reaction kinetics (limitation by mass transfer)
- A significant effect of the flow rate on the nanoplastic photocatalytic degradation is observed, which means that an external transfer limitation intervenes, at least at low flowrate values (fig 3b).
- > At free pH 6.3, we are working under optimum conditions for the photo-oxidation. On the contrary, at basic pH, the TiO<sub>2</sub> surface is negatively charged and a part of the ester functions of the nanobeads surface can be hydrolyzed into negative carboxylate groups, causing electrostatic repulsion forces and thus showing a drastic reduction of the TOC conversion (fig 4b).
- Light intensity effect has been studied by varying the number of UV-A lamps irradiating the TiO2-P25/β-SiC foams (Figure 4b), in this case also the results are consistent because the process is more efficient with an irradiance value of 112 W/m2) rather than 56 W/m2.



#### Chemical structure and particles size effects

- > For a similar particle size, PMMA mineralization appears to be faster than PS (kNBs-PPMA = 0.0148 mg/L.min while kNBs-PS1 = 0.0065 mg/L.min).
- Size effect: The results of figure 4 show that the kinetics are faster with the 105 nm PS nanobeads than those of 540 nm. This result seems logical because smaller particles lead to larger contact area with TiO2 photocatalyst.

#### CONCLUSION

These first results are very encouraging, we have shown that it was possible to mineralize nanobeads of PMMA and PS under irradiation of TiO2 in aqueous solution. We also showed that the photocatalytic process can be effective for polymers with various molecular structures, such as polystyrene and PMMA and also with different average sizes of nanobeads. It is now necessary to optimize the process, but also to better understand the degradation mechanisms. However, one of the most important challenges to take up in this work will be to find an analytical procedure that is easy to implement.

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PH Allé, P Garcia-Muñoz, K Adouby, N Keller, D Robert Environmental Chemistry Letters, (2020) 1-6



t (min)

### Microplastics contribution to the marine environment through streams in Fildes Peninsula (King George Island, Antarctica)

De Feo Bárbara.

Plastic is a low cost, durable and versatile material, and has become an indispensable part of our modern life. Their high production level and deficient waste management have led to the accumulation of plastic garbage in aquatic ecosystems around the world, resulting in accumulation in the ocean. In this sense, lotic systems are considered one of the main discharge routes of plastics from terrestrial to marine ecosystems, including Antarctica. Here, human activities have increased due to the establishment of new scientific bases, exploitation of resources, tourism, which have led to increasing waste production and pollution of streams with materials from different sources. The aim of this study is to evaluate the discharge of microplastics from streams with different environmental characteristics and exposure to human activities to the coastal area in Fildes Peninsula. Water samples were taken from different streams in the summer and autumn antarctic campaigns in 2018 and 2019 with a Surber net. The size, colour and density of plastics was analyzed with a stereo microscope and plastic identification corroborated with a microscope with polarized light. Fibers and fragments were found in all samples, while foams and paint fragments were observed in some of them. The total density of microplástics ranged between 10.1 to 1.3 particles/m3. In all the sampled streams, the fiber density was higher compared to the different types of microplastics found. This study establishes the first evidence for the presence of microplastics in freshwater ecosystems in Antarctica.

Keywords : Antarctica , Microplastics , Streams



### Microplastics contribution to the marine environment through streams in Fildes Peninsula (King George Island, Antarctica)

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N° 334440

### 1. INTRODUCTION

Plastic is a low cost, durable and versatile material, and has become an indispensable part of our modern life. Its high production level and deficient waste management have led to the increase of plastic garbage in aquatic ecosystems around the world, and its accumulation in the ocean.

Microplastics (MP) are particles <5 mm. These could be are either pellets intentionally manufactured in that size to make larger plastics objects, or the result of photochemical and mechanical degradation of larger plastic garbage in the environment (Cole et al., 2011).

Although not all plastic comes from land sources, lotic systems are considered one of the main discharge routes of plastics from terrestrial to marine ecosystems (Lebreton et al., 2017)

In Antarctica, human activities have increased due to the establishment of new scientific bases, fisheries and tourism, which have led to increasing waste production and pollution of aquatic systems with materials from different sources (Kelly et al., 2020).



**Figure 1.** Location of the study area (Fildes Peninsula, King George Island. A) Drake stream, that flows into the Drake Passage and B) AINA stream that flows into Maxwell Bay in front of the Artigas Scientific Station (Uruguay). Red dots indicate sampling points.

#### 2. OBJECTIVES

#### General

 Evaluate the discharge of MP from streams with different environmental characteristics and exposure to human activities to the coastal area in the Fildes Peninsula.

#### Specific

- Classify MP according to type (fibers, fragments, foams), size and colour.
- Estimate MP density (items/m<sup>3</sup>) in water samples from the streams studied.

### 3. METHODS

- Water samples were taken in AINA and Drake streams in summer and autumn of 2018 and 2019.
- $\,$  A Surber sampler ( net 300  $\mu m)$  was placed against the current near the mouth of each stream. Filtered volume was estimated with a flowmeter.
- Samples were analyzed via direct observation with a stereo microscope.
- The MP found were measured with a millimeter ruler and classified according to type, size and color. During the observation, a laboratory contamination control was placed near the stereo microscope.
- Fibers and fragments found in the samples were observed under a microscope with polarized light to verify their synthetic nature.



**Figure 2.** Sampling of the AINA Stream with a Surber net.

### 4. RESULTS

 
 Table 1. Density of MP found in AINA stream and Drake stream in Summer (2018, 2019) and Autumn (2018).

YEAR	SEASON	STREAM	Total density (items/m³)	Fibers (items/m <sup>3</sup> )	Fragments (items/m <sup>3</sup> )	Fibers aggregates (items/m³)	Paint fragments (items/m³)	Foams (items/m³)
2018	Summer	AINA	1.28	0.73	0.15	0.03	0.24	0.13
	Autumn	AINA	3.24	2.90	0.34		*	
2019	Summer	AINA	10.13	7.79	1.21	0.09	0.87	0.17
		Drake	9.14	7.16	1.21	( <b>*</b> )	*	0.77

- Fibers and fragments were found in all the analyzed samples, and were more abundant than other MP types in both streams.
- Densities of MP in the streams were higher in 2019, compared to 2018.
- AINA stream had higher MP densities in Autumn 2018, compared to the Summer of the same year.



**Figure 3.** MP found in the samples. A) Fibers, B) foams, C) paint fragment and D) fragment. Black square size is 1.0 x 1.0 mm.

#### 5. CONCLUSIONS

- This study constitutes the first evidence of the presence of microplastics in freshwater ecosystems of King George Island, Antarctica (González-Pleiter et al., 2020)<sup>1</sup>
- Fibers, fragments and foams were found in both streams, while paint fragments were found only in the AINA stream.
- The proximity or distance of the streams to areas of human activities are not the only determining factor for the presence of MP, except perhaps in the case of paint fragments. Wind transport of MP could be a factor involved in explaining our results (González-Pleiter et al., 2020)<sup>2</sup>.

Cole et al., 2017. Marine Pollution Bulletin 62, 2588-2597. González-Pleiter et al., 2020.<sup>1</sup> Marine Pollution Bulletin 161, 111811. González-Pleiter et al., 2020.<sup>2</sup> The Cryosphere. <a href="https://doi.org/10.5194/tc-2020-261">https://doi.org/10.5194/tc-2020-261</a> Kelly et al., 2020. Marine Pollution Bulletin 154, 11130. Lebreton et al., 2017. Nature communications, 8, 15611.

## The influence of iron (oxyhdr)oxides on the surface properties of polystyrene in aquatic environments

Schmidtmann Johanna, Ottermann Katharina, Helfricht Nicolas, Papastavrou Georg, Peiffer Stefan.

Microplastic particles in the environment are expected to undergo numerous processes associated with natural colloids, for example heteroaggregation with iron oxides, which may influence the surface properties and transport behaviour of microplastics. We incubated 1 µm polystyrene (PS) particles with synthesized ferrihydrite, a weakly crystalline Fe(III)-(oxyhydr)oxide that occurs in large quantities in aquatic and soil environments. The surface properties of PS were examined before and after incubation using dynamic light scattering techniques. We observed that heteroaggregation between PS and ferrihydrite strongly depends on the pH value and on the reaction time. The size of aggregates increased with reaction time (between 0 h and 1 week). Strongest heteroaggregation was found at pH values between 6 and 8, close to the point of zero charge. In this pH range, the zeta potential values of incubated PS were nearly the same as those of ferrihydrite. Scanning electron microscopy images showed that the PS surface was covered entirely with ferrihydrite at a neutral pH. Our observations clearly demonstrate that the surface properties of PS particles were modified through interaction with ferrihydrite. Overall, our research suggests that Fe(III)-(oxyhydr)oxides are highly important reactants to control the environmental behaviour of microplastic particles.

Keywords : Adsorption , Colloids , Ferrihydrite , Heteroaggregation



### The influence of iron (oxyhydr)oxides on the surface properties of polystyrene microplastics in aquatic environments [334127]

Johanna Schmidtmann<sup>1</sup>, Katharina Ottermann<sup>2</sup>, Nicolas Helfricht<sup>2</sup>, Georg Papastavrou<sup>2</sup>, Stefan Peiffer<sup>1</sup>

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The interactions of MP particles with other particles in the environment are of great importance. In fact, under certain conditions, the surface properties of PS particles were described by ferrihydrite, which covered the entire PS surface.



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### Session 25.8\_Me. Chaired by Eva Cardona, Menorca

#### Microplastics pollution detected in pristine lakes in the Arctic region

Vianello Alvise, Farstad Nashoug Benedikte, Lorenz Claudia, Pettersvik Arvnes Maria, Gomiero Alessio, Fabres Joan. Paper number 334349

**Analysis of microplastic formation in fragmentation tests** *Sonnenberg Johanna, Benke Annegret, Potthoff Annegret.* Paper number 334163

## Microplastic aggregation and transfer in marine systems (MOTION): Research project presentation

*Khatmullina Liliya, Gorokhova Elena, Chubarenko Irina.* Paper number 334273

#### Engineering the thermal stability of a PET degrading enzyme

*Weigert Sebastian, Höcker Birte.* Paper number 333425

#### Microplastics pollution detected in pristine lakes in the Arctic region

Vianello Alvise, Farstad Nashoug Benedikte, Lorenz Claudia, Pettersvik Arvnes Maria, Gomiero Alessio, Fabres Joan.

Microplastics (MP) have been detected worldwide, even in remote areas. Up to date, it is unclear whether there still exist pristine environments or MP pollution has become a ubiquitous global issue. To better understand the extent of this issue and to assess the baseline concentration of MP in pristine environments, we investigated three lakes in the arctic region (Lofoten Islands, Norway) with low levels of anthropogenic impact. Surface water samples were taken at two points at each of the lakes utilizing a custom-built and already well-established filtration device (UFO system), enabling sampling of MP in the range of 10–5000 µm. MP were extracted by using a multistep enzymatic and catalyzed oxidative treatment and were analyzed by µFTIR imaging followed by automatic identification, quantification and particle analysis pipeline (siMPle). Special care was taken to reduce contamination from atmospheric deposition by collecting air blanks at each location as well as performing a procedural lab blank, and finally subtracting the recorded contamination from the sample's results. Preliminary results revealed low MP concentration ranging from 0 to 45 MP/m3. The average concentration of 20 MP/m3 was comparable to the lower concentrations detected in marine water and drinking water studies. Most of the detected MP (59%) were in the size range of 11-75 µm, and only 10% was larger than 500 µm. Polypropylene (PP) and polyester (PEST) dominated the polymer composition with a lower contribution of polyamide (PA) and polyvinyl chloride (PVC). The polymer composition and overall small particle size are hinting towards atmospheric deposition as a major source of the MP accumulating in these lakes. These results show that MP pollution, in very low concentrations, has reached even pristine freshwater environments in the Arctic. Furthermore, the results highlight the importance of sample contamination prevention and assessment, especially when dealing with low-level MP concentrations.

Keywords : Pristine lakes , small microplastics , surface waters



## **Microplastics pollution detected in** pristine lakes in the Arctic region

<u>Alvise Vianello<sup>1</sup>, Benedikte Farstad Nashoug<sup>2</sup>, Claudia Lorenz<sup>1</sup>,</u> Maria Pettersvik Arvnes<sup>2</sup>, Alessio Gomiero<sup>3</sup>, Joan Fabres<sup>2</sup>, Jes Vollertsen<sup>1</sup>

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Introduction

Microplastics (MP) have been detected worldwide, even in remote areas. Up to date, it is unclear whether there still exist pristine environments or MP pollution has become a ubiquitous global issue. To better understand the extent of this issue and to assess the baseline concentration of MP in pristine environments, we investigated three lakes in the arctic region (Lofoten Islands, Norway) with low levels of anthropogenic impact.

Sampling

Surface water samples were taken at two points at each of the lakes utilizing a custom-built and already wellestablished filtration device<sup>[1]</sup> (UFO system), enabling sampling of MP in the range of 10-5000  $\mu$ m. Field blanks for airborne contamination were collected at each location.





Sample preparation

Map of the sa

The samples and a procedural lab blank were processed through a well established multistep enzymatic-oxidative sample treatment [2,3]



2. Two step enzymatic treatment (Protease; Cellulase & Viscozyme) Sample extraction and concent A sub-sample (min 200µL) was depo on ZnSe window and dried overnight 4. Flotation with SPT ( $\rho = 1.7-1.8 \text{ g cm}^{-3}$ ) 5.Sample evaporation in glass vial -50% EtOH add. (5 mL)

mpling locations (red dots on map)

Sample preparation step

**Results & Discussion** 

TOT MP fibres TOT MP particle MP concentration after blank correction 0 – 46 MPs/m<sup>3</sup> (Average 21 MPs/m<sup>3</sup>) 0 – 120 µg/m<sup>3</sup> (Average 20 µg/m<sup>3</sup>) Eggum In Eggum Out Gronli In Gronli Out Halsan In TOT MP particle 60 50 40 30 Eggum\_In Eggum\_Out Gronli\_In Gronli\_Out Halsan\_In Halsan\_Out

MP conc. and polymer comp. (particle numbers)

#### Conclusion

MP pollution, in very low concentrations, has reached even pristine freshwater environments in the Arctic, although the main sources are likely linked to airborne deposition. Furthermore, the results highlight the importance of sample contamination prevention and assessment, especially when dealing with low-level MP concentrations

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Sampling in Grønlivatnet inlet with UFO system

#### µFTIR-Imaging analysis & siMPle automated analysis

Sampling in Nedre

atnet (Eggum)

The samples and the blanks were analysed by FPA-µFTIR-Imaging (Agilent Cary 620-670). The FTIR-Imaging data were analysed using siMPle, a freeware software for  $\mu FTIR$ -Imaging analysis with MP auto-detection features.



#### Preliminary results revealed low MP concentrations ranging from 0 to 46 MP/m<sup>3</sup> after performing blank correction. The average concentration of 20 MP/m3 was comparable to the lower concentrations detected in marine water and drinking water studies. Most of the detected MP (59%) were in the size range of 11–75 $\mu m,$ and only 10% was larger than 500 $\mu m.$ Polypropylene (PP) and polyester (PEST) dominated the polymer composition with a lower contribution of polyamide (PA) and polyvinyl chloride (PVC). The polymer composition and overall small particle size are hinting towards atmospheric deposition as a major source of the MP accumulating in these lakes.

Sample matrix	MP concentration	Unit			
Treated wastewater	1.9×10 <sup>4</sup> -4.5×10 <sup>5</sup>	MPs/m <sup>3</sup>			
(Simon et al. 2018) [6]	500-1.2×10 <sup>4</sup>	µg/m³			
Stormwater ponds - water	490-2.3×104	MPs/m <sup>3</sup>			
(Liu et al. 2019) [3]	85-1.1×103	µg/m³			
Stormwater ponds - sediment	1.5×10 <sup>3</sup> -1.3×10 <sup>5</sup>	MPs/kg			
(Liu et al. 2019) [7]	115-2.9×104	µg/kg			
MPs concentration in other environmental and anthropogenic matrices according to recent publications					

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We thank the INTERREG FanpLESStic-sea project for financing this study! Find the project here:



#### Analysis of microplastic formation in fragmentation tests

Sonnenberg Johanna, Benke Annegret, Potthoff Annegret.

One of the hotspots of fragmentation of plastics in the sea are beaches where sand acts as an abrasive. Artificial fragmentation tests without abrasives have the advantage that the resulting particles can be easily analyzed. However, these test conditions differ from those on the beach. Therefore, tests in which sand or gravel is used are necessary. We use glass bottles that are partly filled with sand and rotated during the experiment. By setting the rounds per minute, the energy input can be adjusted to a defined value. For the analysis of the formed particles a separation from the nonpolymeric particles is necessary. A visual selection is not an accurate method and cannot consider the particle size range below 1 mm. The analysis of the material loss from the sample is also not sufficient. Thus, we have developed a separation setup to separate the polymer particles from the sand particles. Subsequently, the particle size distribution can be measured using appropriate methods. The separation setup consists of a funnel in which the particles separate due to their different densities. The required high-density liquid is an aqueous solution of potassium carbonate (K2CO3). The potassium carbonate solution is advantageous as the ions also occur in the sea. In addition, the monovalent potassium ions do not react with the particle surfaces, which can be a problem with divalent ions such as zinc. An almost saturated solution with a density of 1.52 g/cm3 is used. Thus, nearly all polymers, including PET and PE, can be separated from the sand particles. In combination with a previous fragmentation step, recovery rates of at least 98 % were achieved in preliminary tests. With this setup we investigate the fragmentation behavior of plastic pellets in different weathering conditions. In addition, naturally and artificially weathered samples are compared.

Keywords : Abrasive , Artificial Testing , Fragmentation , Microplastics , Polystyrene , Potassium carbonate , Separation , Weathering





### **ANALYSIS OF PARTICLE FORMATION IN FRAGMENTATION TESTS**

Johanna Sonnenberg<sup>1</sup>, Leonie Sailer<sup>2</sup>, Annegret Benke<sup>2</sup>, Annegret Potthoff<sup>2</sup>

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#### www.ikts.fraunhofer.de

## Microplastic aggregation and transfer in marine systems (MOTION): Research project presentation

Khatmullina Liliya, Gorokhova Elena, Chubarenko Irina.

Interactions with various suspended soils and natural colloids are important but poorly understood aspects of microplastic (MP) behavior in aquatic environments. The result of these interactions is the formation of aggregates, where MP are incorporated together with live and dead organics, so-called marine snows. The vertical transport of organic substances, including living cells, represents an essential part of the carbon pump exporting organic matter from the photic zone to the deep ocean. Plastic particles have been found in marine snow along the water column as well as in sediments, implying that sinking aggregates could be an efficient export mechanism of MP to the deep ocean. However, it is unclear whether the sinking rate and the associated carbon pump processes can be affected by MP at the ecologically relevant abundances of the plastic litter. In MOTION project, we focus on reviewing interactions between MP and other particles ubiquitously present in pelagia and analyze the kinetics of the aggregate formation, their settling characteristics, and the role of microorganisms in the aggregation. The project is funded by the Swedish Institute.

Keywords : aggregation , marine snow , microplastics , suspended matter

## Microplastic aggregation and transfer in marine systems (MOTION): Research project presentation

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Interactions with various suspended soils and natural colloids are important but poorly understood aspects of microplastic (MP) behavior in aquatic environments. The result of these interactions is the formation of aggregates, where MP are incorporated together with live and dead organics, so-called marine snows. The vertical transport of organic substances, including living cells, represents an essential part of the carbon pump exporting organic matter from the photic zone to the deep ocean. Plastic particles have been found in marine snow along the water column as well as in sediments, implying that sinking aggregates could be an efficient export mechanism of MP to the deep ocean. However, it is unclear whether the sinking rate and the associated carbon pump processes can be affected by MP at the ecologically relevant abundances of the plastic litter. In MOTION project, we focus on reviewing interactions between MP and other particles ubiquitously present in pelagia and analyze the kinetics of the aggregate formation, their settling characteristics, and the role of microorganisms in the aggregation.

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FATE AND IMPACTS OF MICROPLASTICS: KNOWLEDGE AND RESPONSIBILITIE

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#MOTIONproject: link.growkudos.com/1jr11c2yr5s Project DOI: 10.26303/02z9-yn51

# 334273

### Suspended and settling particles in the water column, an estuary example

Particles of different nature present in the water column comprise a complex system driven by environmental factors and interactions between its components



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Passow, 1991 10.1007/BF01313655 Puls et al., 1997 10.1016/S0025-326X(96)00161-0 Sivkov, 1994 (in Russian) <u>search.rsl.ru/ru/record/01000758571</u> Shanks, 2002 <u>10.1016/S0278-4343(02)00015-8</u>
# **Engineering the thermal stability of a PET degrading enzyme**

Weigert Sebastian, Höcker Birte.

One of the most abundant plastics is polyethylene terephthalate (PET), a semi-crystalline polyester made from terephthalic acid and ethylene glycol. Its outstanding properties such as chemical resistance, lightweight and stability facilitate the use of the material in challenging applications like food and beverage packaging and for fibre production. But like for many other plastics these characteristics lead to the accumulation in the environment in the form of microplastic, which causes problems on many different levels. For PET-derived microplastic a new perspective arose with the discovery of PETase in Ideonella sakaiensis.(1) This enzyme is the key for the organism's ability to hydrolyse PET and use it as a nutrition source. It therefore features as the first known PET degrading enzyme with substantial activity at ambient temperatures. Although the enzymes moderate turnover rates fit its biological context, it limits its utilization for microplastic related applications like recycling and decontamination. One way to develop PETase into an enzyme with the desired properties is rational protein design. As a first step we focus on the thermal stability, which is often linked to kinetic stability of an enzyme. Here we used a fully automated algorithm for protein stabilization called PROSS (2) and yielded an increase in Tm of 8°C. We continued to combine this variant with an even more stable enzyme, designed by Cui et al. using their semi-automated clustering algorithm GRAPE, called DuraPETase.(3) Besides ranking the performance of the two algorithms, we are also looking for synergistic mutations from both algorithms to create a further improved PETase. These examples illustrate possible contributions of protein design to facilitate recycling and decontamination in the scope of microplastic. (1) Yoshida et al. (2016) DOI: 10.1126/science.aad6359 (2) Goldenzweig et al. (2016) DOI: 10.1016/j.molcel.2016.06.012 (3) Cui et al. (2019) bioRxiv DOI: doi.org/10.1101/787069

Keywords : enzymatic degradation , enzyme , PET degradation , PETase , protein design , thermal stability



# Engineering the thermal stability of a PET degrading enzyme [ID-333425]

Sebastian Weigert & Birte Höcker Department of Biochemistry III, University of Bayreuth CRC 1357 Microplastics

# Introduction

Polyethylene terephthalate (PET) is a semi-crystalline polyester made from terephthalic acid and ethylene glycol. Its outstanding properties such as chemical resistance, lightweight and stability promote its widespread use with the downside of accumulation in the environment in the form of microplastic causing problems on all levels. For PET derived microplastic a new perspective arose with the finding of the enzyme PETase in *Ideonella sakaiensis*.<sup>(1)</sup> This enzyme is the key for the organism's ability to fully break down PET and use it as a nutrition source. It therefore features as the first known PET degrading enzyme with substantial activity at ambient temperatures, but this limits its utilization for microplastic related applications like recycling and decontamination. One way to develop PETase into an enzyme with the desired properties is rational protein design. Here we use the PROSS<sup>(2)</sup> algorithm to optimize IsPETase. We further compare and combine our results with those from Cui et al<sup>(3)</sup> who applied their GRAPE algorithm, thereby revealing synergistic combinations of mutations from both approaches.



deonella sakaiensis<sup>(3)</sup>(a), solid PET surface before (b) and after incubation with isPETase (c).

# **PROSS** – Algorithm

- Fully automated and unsupervised
- **Phylogenetic analysis**: Depends on sequence alignments to identify potential stabilizing mutations for higher thermostability.
- Rosetta Scoring: Individual and pairwise scoring of the suggested mutation.
- **Output:** Seven variants from conservative to progressive number of mutations. Here we used an intermediate design bearing 7 mutations: **S214H** and six further mutations, here labelled M1-M6.

# **GRAPE** – Algorithm

- Semi-automated, iterative, supervised, experiment based
- Prediction of stabilizing mutants: Based on a combination of statistical energy function, force-field based energy function (Rosetta) plus FoldX and a phylogenetic analysis. After filtering this yielded 21 mutations.
- Biochemical Analysis and Clustering: The 21 mutations were individually analysed and clustered upon Tm, activity and location using Kmeans algorithm.
- **Greedy algorithm:** Efficient combining of synergetic mutations to sample the energetic landscape overcoming epistatic effects. Iterative biochemical analysis, clustering and recombination.
- Output: Final variant with 10 mutations including S214H as suggested by PROSS.



References: <sup>(1)</sup> Yoshida et al. DOI: 10.1126/science.aad6359, <sup>(2)</sup> Goldenzweig et al. DOI: 10.1016/j.molcel.2016.06.012, <sup>(3)</sup> Cui et al. DOI: doi.org/10.1101/787069



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# Session 25.8\_Ma. Chaired by Marta Sales, Menorca

Water quality and chemical weathering of the Teesta River of Eastern Himalaya, India *Tenzin Tsering*. Paper number 334266

# Can specially protected areas be protected from microplastics?

Dimante-Deimantovica Inta, Buhhalko Natalja, Vianello Alvise, Barone Marta, Lanka Anna, Prokopovica Anda, Suhareva Natalija, Vecmane Elina, Burdukovska Valentina. Paper number 334280

# Microplastic pollution in three rivers with varying anthropogenic impact in South Eastern Norway Lorenz Claudia, Dolven Jane K., Værøy Nina, Gomiero Alessio, Stephansen Diana, Olsen Stein B., Vollertsen Jes. Paper number 334347

**Wastewater treatment plants as a source of microplastics to the environment in New Zealand** *Ruffell Helena, Gaw Sally, Pantos Olga, Northcott Grant.* Paper number 334233

# Water quality and chemical weathering of the Teesta River of Eastern Himalaya, India

Tenzin Tsering.

Himalaya region is a pivotal environmental monitoring site because of its sources of freshwater. dynamic vegetation and topography, prone to flood and drought due to climate change. The freshwater from Himalaya serves to the livelihood of billions of people. Herein, for the first time, based on the original data of major ions, the water quality of the Teesta River is studied. The evaluation of the major ion and trace elements against the standard guideline values and the average chemical composition of world rivers were discussed. The predominance of Ca, Mg and HCO3 in all waters reflects the influence of carbonate weathering on the Teesta River. However, an increase in the Na/Ca ratio was linked to the increase of Si downstream, indicating that silicate weathering was predominant in the lowlands of Teesta drainage. The rate of silicate weathering is dependent on an overall balance of key factors including gradient, contact time, temperature and vegetation. The higher concentration of cations was balanced by the SO4 originating from the action of H2SO4 and H2CO3 on carbonates and silicates. Rock weathering (carbonate-silicate weathering) is the key mechanism that controls the major ion chemistry of the Teesta River followed by evaporite dissolution. The findings of the study can support future studies on the geochemistry of rivers in the Himalayas, which could also contribute to water resource management and the preservation of natural resources.

Keywords : Carbonate weathering , Eastern Himalayas , Major ions , Sikkim Himalaya , Silicate weathering , Teesta River chemistry

# AN ASSESSMENT OF CHEMICAL WEATHERING BASED ON ION CHEMISTRY OF THE TEESTA RIVER, EASTERN HIMALAYA Tenzin Tsering\*, Mahmoud S.M. Abdel Wahed, Sidra Iftekhar, Mika Sillanpää \* LUT School of Engineering Science, Lappeenranta University of Technology, P.O. Box 20, 53851, Lappeenranta, Finland. (Tenzin.Tsering@lut.fi)

# INTRODUCTION

# **OBJECTIVES**

Himalayan rivers are a significant source of water for billions of people. The climate change and rapid receding of glaciers lead to the change in water chemistry and the contamination level of pollutant in the rivers. Thus, monitoring of the river's health are crucial. Teesta river is in the Sikkim state in the Eastern Himalaya, serving as a lifeline for millions of people and representing the Sikkim Himalaya overall fluvial system. However, there is a lack of scientific research on water chemistry compared to the other rivers of Indian Himalaya [1]

To study the spatial variations of major ions and trace elements of the Teesta River

To study the weathering pattern based on the major jons in the Teesta River

### **EXPERIMENTAL METHOD**

### SAMPLING

A sampling of the Teesta River was performed in March 2018 and study sites are shown in the map  $(\rightarrow)$ 

ANALYSIS:

- Metals were quantified using Inductively Coupled Plasma-Optical Emission Spectroscopy
- Anion analyses are performed using Ion Chromatography

### **RESULT AND DISCUSSION**



The ternary plot of the Teesta River indicated that Ca<sup>2+</sup> and HCO<sub>3</sub><sup>-</sup> in the Teesta River are the highest composition. A general tendency of evolution from carbonate weathering to silicate weathering as water flow from upstream to downstream was observed and shown by arrows in the ternary plot above.



- The positive correlation between Mg<sup>2+</sup>+Ca<sup>2+</sup> and HCO<sub>3</sub><sup>-</sup> as shown in the binary plot above indicates the carbonate weathering at upstream and downstream sites with additional contribution from silicate weathering at the downstream.
- The excess of the stoichiometry of total ( $Mg^{2+}+Ca^{2+}$ ) than  $HCO_3^{-1}$  is likely balanced by  $SO_4^{2-}$  from sulphide dissolution [2]. Excess HCO<sub>3</sub>-+SO<sub>4</sub> 2- over Ca2+ +Mg<sup>2+</sup> indicates non-carbonate sources (i.e., interaction of H<sub>2</sub>SO<sub>4</sub> and H<sub>2</sub>CO<sub>3</sub> on silicates) and this excess should be balanced by Na<sup>+</sup> and K<sup>+</sup> that were found in higher amounts than Cl<sup>-</sup>

The rate of silicate weathering is more dependent on the overall balance of key factors including gradient, contact time, temperature and vegetation.

# CONCLUSION

- The Teesta River chemistry is mainly governed by natural phenomena, with carbonate weathering at the upstream and downstream is dominated mainly by silicate weathering
- $Ca^{2+}$ ,  $Mg^{2+}$  and  $HCO_3$  were mainly provided from carbonate weathering while  $Na^+$  and  $K^+$ originated from silicate weathering throughout the entire watershed.
- H<sub>2</sub>CO<sub>3</sub> and H<sub>2</sub>SO<sub>4</sub> make significant contributions to the weathering process
- The concentration of major ions and elements is in the range of concentration of the global average rivers.
- The Teesta river is pristine with a less anthropogenic contribution to the river's major ion chemistry

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A geological map of Sikkim with the sampling sites [1]



 Na<sup>+</sup> / (Na<sup>+</sup> + Ca<sup>2+</sup>)
 The Gibb's plot (above) of the Teesta River shows that the river is governed mainly by carbonate and silicate endmembers.

### Table: Chemical Guideline value by WHO and BIS

Parameter	Units	WHO <sup>a</sup> (guideline value)	Note	BIS <sup>b</sup> (Acceptable limit)	BIS <sup>b</sup> (Permissible limit)	Note	This Study <sup>c</sup>	
pН			6.5-8	6.5-8.5	-		7.69	
Turbidity	NTU	5		1	5		7.02	
TDS	mg/L			500	2000		66.20	
Antimony (Sb)	mg/L	0.02					Not detected	
Barium (Ba)	mg/L	0.7		0.7	No relaxation		0.0048	
Boron (B)	mg/L	0.5 (T)		0.5	1		1.01	
Cadmium (Cd)	mg/L	0.003		0.003	No relaxation		Not detected	
Calcium (Ca)	mg/L		<250	75	200		9.56	
Chlorine (Cl)	mg/L	5 (C)	-	-	-		1.1	
Chromium (Cr)	mg/L	0.05 (P)	For total Chromium	0.05	No relaxation	For total Chromi um	0.0015	
Copper (Cu)	mg/L	2		0.05	1.5		Not detected	
Potassium (K)	mg/L		<250				3.33	
Magnesium (Mg)	mg/L			30	100		1.44	
Manganese (Mn)	mg/L	0.4 (C)		0.1	0.3		0.0036	
Molybdenum(M	mg/L	0.07		0.07	No relaxation		0.0029	
Nickel (Ni)	mg/L	0.07		0.02	No relaxation		0.0006	
Sodium (Na)	mg/L		<200				3.40	
Lead (Pb)	mg/L	0.01		0.01	No relaxation		Not detected	
Selenium (Se)	mg/L	0.01		0.01	No relaxation		Not detected	
Sulphate (SO42-)	mg/L		<500	200	400		10.24	

standard drinking water specification (second revision) BIS standard 2012. This study<sup>2</sup> means values of the measured elements. According to WHO, C is a concentration below or at the giv value that might affect the taste, appearance and odour of the water. P stands for provisional value, because there is evidence of hazard but limited information on the effect on health. T . al gu on on the effect on health. T is sional guideline value because the given value is below the level that can be a reatment methods, source protection, etc.

# Can specially protected areas be protected from microplastics?

Dimante-Deimantovica Inta, Buhhalko Natalja, Vianello Alvise, Barone Marta, Lanka Anna, Prokopovica Anda, Suhareva Natalija, Vecmane Elina, Burdukovska Valentina.

Microplastics are a widely known and distributed anthropogenic pollution found in all kind of natural habitats, including lake sediments. In this study we compared upper-layer sediments from 5 lakes in Latvia (Baltic States, Northern Europe) to see if ecosystems in specially protected areas have corresponding amounts of microplastic particles buried in the sediments. Sediment cores from lakes under different anthropogenic load were collected in 2019 using a Kayak/HTH gravity-type corer. The upper sediment laver was used for the further sample treatment and analysis. Samples were preoxidised with H2O2, freeze-dried, separated by heavy liquid application, treated with SDS (NaC12H25SO4) solution and different enzymes (alcalase, cellulase, viscozyme). For the further oxidation process, a Fenton reaction was applied and sample treatment was finished with a final heavy liquid separation. Particles larger than 500 µm were separated by a steel filter and analysed visually, while particles within the size range of 100 - 500 µm were analysed using µFTIR (Fourier Transform Infrared Spectroscopy) system Spotlight 400 (Perkin Elmer). A high variety of polymer groups was detected, however it was considerably lower in remote, specially protected areas where the use of water vehicles is prohibited (11 polymer groups) or restricted (16 polymer groups) compared to lakes located in a city or vicinity of a large city (20 to 23 polymer groups were detected). Nevertheless, the most common microplastic group found in lakes both from urban and protected areas was rubbers (from 15.6 to 49.7%). The second most common polymer group detected was polyvinylpolypyrrolidone (PVP), i.e. from 6.4% to 13.6 %. Other dominant groups were viscose, polyvinylacetate (PVA), polystyrene (PS) and polyethylene (PE) comprising from 18.3% to 43.2%. Remote location same as access restrictions may limit ecosystems exposure to microplastic pollution. Funded by the European Regional Development Fund, 1.1.1.2 Post-doctoral project No.1.1.1.2/VIAA/2/18/359, Latvian Environmental Protection Fund (1-08/86/2019).

Keywords : lakes , Latvia , microplastics , sediments , specially protected areas



# Can specially protected areas be protected from microplastics? 334280

Dimante-Deimantovica, I.<sup>1,2</sup>, Buhhalko, N.<sup>3</sup>, Vianello, A.<sup>4</sup>, Barone, M.<sup>1,5</sup>, Lanka, A.<sup>1</sup>, Prokopovica, A.<sup>1,6</sup>, Suhareva, N.<sup>1</sup>, Vecmane, E.<sup>1,5</sup>, Burdukovska, V.<sup>1</sup>

the Built Environment, Aalborg University, Denmark, <sup>5</sup>University of Latvia, Faculty of Geography and Earth sciences, Latvia, <sup>6</sup>University of Latvia, Faculty of Biology, Latvia <sup>1</sup>Latvian Institute of Aquatic Ecology, Latvia, <sup>2</sup>Norwegian Institute for Nature Research, Norway, <sup>3</sup>Tallinn University of Technology Department of Marine Systems, Estonia, <sup>4</sup> Department of

Pinku

Talsu

Usmas1 Usmas2

PRELIMINARY RESULTS

water is prohibited or restricted (5 polymer

specially protected areas where the access to however it was considerably lower in remote, A high variety of polymer groups was detected,

vicinity of a large city (up to 11 polymer groups groups) compared to lakes located in a city or

areas of various anthropogenic load and nature conservation status (Fig.1.). In this study we compared upper-layer sediments from 5 lakes in Latvia (Baltic States, Northern Europe) representing

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reach even the most protected and remote natural areas at Hypothesis for the research: microplastic particles may the considerable amount

# SAMPLING SITES



# Fig. 1. Sampling sites in five lakes in Latvia

with unique ecosystem (Lobelia-Isoëtes plant complex species) water quality and great transparency. Rare and important biotope Pinku Lake – nature park area. Mesotrophic lake with very good lake for recreation is limited. intensive farming takes place around the lake, access and use of Surrounding area is little modified by human activities, no

sampling sites one within and another outsidenature reserve. Lake Usma – part of it is included in the oldest protected area in Latvia, visiting the nature reserve is prohibited. There were two

Enzyme treatment 2

50°C 40h 100rpm

to it is restricted by fence due to fact it is a part of city drinking (Mazais Baltezers, see below) through a pipe. replenishment water for more than 10 years from a eutrophic lake intensified by water pumping station activities when it received water system. Nevertheless, the lake's ecological deterioration was Seksu Lake is located in the vicinity of the capital city and access

**Oxidation - Fenton** 

50°C 40h 100rpm

Size fractionation

Using 100 µm mesh

recharge of the groundwater through infiltration basins subjected to the Talsu Lake is located in the city of Talsi. It is hypereutrophic lake. and industrial wastewatersfor a long time. Still, used for artificial surrounded by urban areas. It is eutrophic lake, receiving domestic Mazais Baltezers Lake is located close to the capital city and urban pollution from its surroundings and

considered as one of the most polluted lakes in the country

Analysis of particles ≤500 - ≥ 100 µm

Fig. 4. Sampling in Talsu Lake, August, 2020

Using µHIIK

Fig. 5. Samples treatment scheme

Density separation

Evaporation 50% EtOH

# METHODS

gravity-type corer (Fig. 2. -4.). The upper sediment layer (2 to 5 analysed using µFTIR (Fourier Transform Infrared Spectroscopy) analysis. Particles within the size range of 100 - 500 µm were samples were treated and analysed as well. system Spotlight 400 (Perkin Elmer). Plastic polymers possibly cm) was used for the further sample treatment (Fig. 5.) and Sediment cores were collected in 2019 using a Kayak/HTF analysis. For contamination and control – blank and control introduced by sampling device were excluded from further



comprising from 18.3% to 43.2%

(PVA), polystyrene (PS) and polyethylene (PE) from 6.4% to 13.6 %, viscose, polyvinylacetate

groups were polyvinylpolypyrrolidone (PVP), i.e.

dominating in protected lakes. Other dominant protected areas was rubbers (from 15.6 to 49.7%) group found in lakes both from urban and

Nevertheless, the most common microplastic

were detected).

dry sediments ranged from 22 to 1648 particles/g

The abundance of total microplastic particles in

(average 409 particles/g) (Fig. 6. - 7.).



SDS treatment





Enzyme treatment 1

50°C 40h 100rpm



Fig. 3. Sediment cores sampli

everything below 50 µm was removed from further data

microplastic groups among lakes. Oval sign for protected areas Fig. 6. Variation in size distribution (size class step – 50 µm due to possible contamination in laboratory) of differen

sediment disturbance. PVP and viscose is included in group

Mazais Baltezers Lake (MB) results most likely represent

plastic pollution such

as atmospheric

5

may limit

Remote location same as access restrictions ecosystems exposure

Fig. 7. Microplastic groups and their composition among lake

M Batezens Pinku

Seksu Lake

Tals

CALLER L SPUIGO

PRE PRE PRE PRE PRE

microplastic pollution as far as it is related Protected, remote areas are not safe from direct anthropogenic contribution.

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Our thanks to everyone who helped in organizing and participated in sampling. This study is funded by the European Regional Development 1.1.1.2 Post-doctoral project No.1.1.1.2/VIAA/2/18/359 and Latvian

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Environmental Protection Fund (1-08/86/2019).

# Microplastic pollution in three rivers with varying anthropogenic impact in South Eastern Norway

Lorenz Claudia, Dolven Jane K., Værøy Nina, Gomiero Alessio, Stephansen Diana, Olsen Stein B., Vollertsen Jes.

The ubiguitous pollution of various environments with microplastics (MP) is a global issue of growing concern. Freshwater environments, especially rivers, have gathered increasing attention as recent findings point out their role as relevant pathways of MP distribution. To see whether rivers with different characteristics carry different MP loads and polymer compositions, we investigated three rivers (city influenced, agriculture influenced and pristine) in South Eastern Norway regarding their MP concentration, loads, polymer composition and size class distribution. Water samples of 1 m3 volume were taken with a well-established, custom-build filtration device (UFO-system), allowing for sampling MP down to 10 µm. Samples were prepared for analysis applying a multistep enzymatic-oxidative treatment and separated into two size fractions (10–300 µm and 300–5000 µm) which were analysed via state-of-the-art spectroscopic methods and the smaller size fraction additionally applying a thermoanalytical method. MP concentrations and loads in the small size fraction (10–300 µm) were highest in the city influenced river (1067 MP/m3 and 2740 µg/m3) followed by the two rivers in the rural area (138 MP/m3 and 50 µg/m3 and 140 MP/m3 and 450 µg/m3, respectively). Furthermore, the thermoanalytical analysis revealed the presence of tire wear particles in the city influenced river. More than 70% of the MP were between 10 to 100 µm in size, and hardly any MP were larger than 300 µm. MP concentrations of the large size fraction (300–5000 μm) were 150–530 times lower than in the small size fraction. This highlights the importance of analysing the small size fraction (MP 10–300 µm) when considering the impact of land-based discharge of MP to the oceans, as the particle number, weight, and polymer composition of the larger size fraction (MP ¿300 µm) is not sufficient to gain the full picture.

Keywords : GC/MS , Py , small microplastics ,  $\mu$ FTIR imaging



# Microplastic pollution in three rivers with varying anthropogenic impact in South Eastern Norway

<u>Claudia Lorenz</u><sup>1</sup>, Jane K. Dolven<sup>2,3</sup>, Nina Værøy<sup>2</sup>, Alessio Gomiero<sup>4</sup>, COWI SN Diana Stephansen<sup>1</sup>, Stein B. Olsen<sup>2</sup>, Jes Vollertsen<sup>1</sup>

<sup>1</sup> Department of the Built Environment, Aalborg University, Denmark; <sup>2</sup> COWI AS, Oslo, Norway; <sup>3</sup> University of South-eastern Norway, Porsgrunn, Norway; <sup>4</sup> NORCE – Norwegian Research Centre – Environment, Randaberg, Norway

Introduction

The ubiquitous pollution of various environments with microplastics (MP) is a global issue of growing concern. Freshwater environments, especially rivers, have gathered increasing attention as recent findings point out their role as relevant pathways of MP distribution. To see whether rivers with different characteristics carry different MP loads and polymer compositions, we investigated three rivers (city influenced, agricultural influenced and pristine) in South Eastern Norway regarding their MP concentration, loads, polymer composition and size class distribution.



Our study showed that the number of microplastic particles were 7 times larger in the city influenced river (Akerselva) compared with the two rivers from the rural areas (Hobøl and Gryta). The city influenced river also had the most diverse polymer type composition and was the only river to contain tire wear particles.

More than 70% of the MP were between 10 to 100  $\mu$ m in size, followed by 13–28% in the 100–300  $\mu$ m fraction and hardly any MP were larger than 300  $\mu$ m. MP concentrations of the large size fraction (300–5000  $\mu$ m) were 150–530 times lower than in the small size fraction. This highlights the importance of analysing the small size fraction (MP 10–300  $\mu$ m) when considering the impact of land-based discharge of MP to the oceans.

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NORCE

# Wastewater treatment plants as a source of microplastics to the environment in New Zealand

Ruffell Helena, Gaw Sally, Pantos Olga, Northcott Grant.

Microplastics are ubiquitous in the environment, due to the intensification in global commercial demand for plastics since the 1960s. The detection of microplastics in remote locations and in a range of aquatic organisms has raised questions about the sources of entry into the environment. Wastewater treatment plants (WWTPs) are thought to be a major source of microplastics, particularly microfibres sourced from washing machine effluent, into aquatic and terrestrial environments. WWTPs are not designed to remove microplastics from sewage, and microplastics are retained in sewage sludge or released with effluent. There is currently a lack of data in New Zealand on the amounts and types of microplastics entering and being discharged from WWTPs, and the risk they pose to the environment. This study is the first of its kind to characterise the contribution of microplastics to coastal ecosystems from different WWTPs in Canterbury, New Zealand. A field study of four tertiary WWTPs was undertaken in the Canterbury region across the month of June 2018. Representative influent and effluent samples were collected from each WWTP, comparing weekdays to weekends. Microplastics were extracted from the sewage by wet sieving, chemical digest, and vacuum filtration and identified using microscopy and Fourier transform infrared spectroscopy (FTIR). Microplastic concentrations ranged from 0.9 – 4.8 particles/L and 0.4 – 2.0 particles/L in influent and effluent respectively, fragments the dominant morphotype and polyester the most frequently detected polymer. Glitter and sponge particles were among the microplastics isolated. An additional field study focusing on microplastic differences in the effluent from three WWTPs was undertaken bi-monthly from June – December 2018 to assess temporal trends. Microplastic concentration in temporal effluent ranged from 0.2 - 2.1 particles/L. The findings of this study suggest the need for greater regulation of plastic consumer products to mitigate the risk of microplastics to the environment.

Keywords : glitter , Microplastics , New Zealand , plastic pollution , sewage , wastewater treatment plants



# Wastewater treatment plants as a source of microplastic to the environment in New Zealand

Helena Ruffell<sup>a</sup>, Olga Pantos<sup>b</sup>, Grant Northcott<sup>c</sup>, Sally Gaw<sup>a</sup>,\* \*School of Physical and Chemical Sciences, University of Canterbury, 20 Kirkwood Avenue, Christchurch 8041, New Zealand <sup>3</sup>Institute of Environmental Science and Research, 27 Creyke Road, Christchurch 8041, New Zealand <sup>3</sup>Northcott Research Consultants Ltd, 20 River Oaks Place. Hamilton 3200. New Zealand

oplastics were detected at an average concentration of 2.4 and 1.2 cles/L in influent and effluent from Study 1 and 1.3 particles/L in oral effluent from Study 2

Glitter and sponge fragments among microplastics found in influent and

ral of microplastics from influent to effluent ranged from 0 – 72%

Microplastic concentrations decreased from influent to effluent

-8.9 x 10<sup>10</sup> microplastics estimated to enter the Canterbury co through discharge of effluent from the four WWTPs each year

- Fragments the most abundant morphotype

Polyester the most abundant polymer type

### Introduction

vater treatment plants (WWTPs) are thought to be a major source of lastics, particularly microfibres sourced from washing machine effluent, into and terrestrial environments.<sup>1</sup>

WWTPs are not designed to remove microplastics from sewage, and microplastics are retained in sewage sludge or released with effluent.<sup>2</sup>

Previous studies have estimated removal efficiencies of greater than 99% of microplastics from influent to effluent (with microplastics retained in sewage sludge), however this still equates to figures in the millions of particles exiting WWTPs daily in

There is currently a lack of data in New Zealand on the amounts and types of microplastics entering and being discharged from WWTPs, and the risk they pose to the

This study is the first of its kind to characterise the contribution of microplastics to coastal ecosystems from different WWTPs in Canterbury, New Zealand.

### **Aims and objectives**

- Determine the relative abundance, morphotypes, and polymer types of microplastics present in wastewater influent and effluent from four WWTPs sampled on a weekday and weekend

- Determine the seasonal variability of microplastics present in effluent from three WWTPs

### Method

- Four tertiary WWTPs in Canterbury, New Zealand were sampled (Figure 1) - 10 L of influent and effluent each collected as a 24-hour composite sample

- Study 2 sampled effluent from WWTPs 1 – 3 on a weekday of June, August, October, December 2018

- Influent and effluent wet sieved 1 mm and 300 µm

- Material on sieves digested using Fenton's reagent ( 1:1 0.05 Fe(II) solution and 30%  $H_2O_2)$ 

Filters analysed under stereomicroscope to identify potential micropla and catergorised by morphotype

- µ-FTIR analysis of 100% of suspected microplastic particles



Figure 1. WWTP sampling locations in Canterbury, New Zealand. (1) Christchurch WWTP, (2) Kaiapoi WWTP, (3) Lyttelton WWTP, (4) Governors Bay WWTP.

- Study 1 sampled influent and effluent from WWTPs 1 – 4 on a weekday and weekend of June 2018

Vacuum filtered onto GFC filters (Whatman, 47 mm diameter, 1.2 µm pore)



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We thank Bronwyn Humphries (Institute of Environmental Science and Research), Matt Polson, Wayne Mackay, Nick Oliver, Rob Micliegoe, Rob Stainthorpe, Kimberley Kovac-Wilks, Amanda Inglir, Peter MicQiago, IUliversity of Canterbury, Mike Bouek, Lee Law, Paul Woods, Wayne Higgs, Grant Burgeas (Christchurch City Council), Rob Frizzell Wainnakarin District Council), Kris Kaser and Shay Alford (City Cane) for WITP access and technical

Acknowledgements

Results

- 8









Figure 2. Study 1: Concentration of microplastic morphotypes and polymer type proportions in weekday and weekend influent and effuent from Christchurch, Kalapoi, Lyttelton, and Governors Bay WWTPs.





BRIAN



Christchurch

Figure 3. Study 2: Concentration of microplastic morphotypes in Christchurch, Kalapoi, and Lyttelton effluents sampled in June, August, October and December

### Conclusions

No consistent trends in microplastic concentration, morphotype and polymer type were observed between weekday/weekend influent and effluent and temporal effluent samples, suggesting a common and continual source of microplastics into WWTPs.

Wastewater influent and effluent is a complex matrix with many sources making comparison within and between studies difficult

- Glitter and sponge particles should be included as microplastic morphotypes

- Wastewater effluent is a significant source of microplastics to the coastal environment in Canterbury, New Zealand



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**Poster session 25.9** 

# Session 25.9\_O. Chaired by Mateo Cordier, Guyancourt

**Microplastics in the upper tidal Elbe: A comparison between sediments from the Hamburg port area and an adjacent nature reserve** *Motyl Larissa, Ruff Ines, Fischer Elke.* Paper number 329226

**Organic matter digestion methods for microplastic extraction from estuarine samplings** *Fertala Laila, Palazot Maialen, Soccalingame Lata, Kedzierski Mikaël, Bruzaud Stéphane.* Paper number 334114

**Identification of environmentally degraded microplastics using the thermal-damaged plastics library** *Ikezawa Yoshio.* Paper number 334150

# Microplastics in the upper tidal Elbe: A comparison between sediments from the Hamburg port area and an adjacent nature reserve

Motyl Larissa, Ruff Ines, Fischer Elke.

In cooperation with the Hamburg Port Authority (HPA) and as part of the Sullied Sedimentsproject funded by the European Union, sediment samples within the Hamburg port area were taken at three sites of different location within the harbor. The sites Stover Strandänd Wedeläre located upstream and downstream of the port of Hamburg, while Köhlbrandïs located in the centre area of the port. Additionally, another sediment sample was taken in the adjacent Hamburg nature reserve Heuckenlock. For the analysis of microplastic concentrations in the samples, the biogenic organic matter was removed by digestion (H2O2 30%, HCl 10% and NaClO 6-14%) followed by wet sieving and a density separation with NaI (density: 1.5 g/cm3). For identification the Nile red staining method in combination with fluorescence microscopy (Axioscope 5/7 KMAT, Zeiss) was applied. A subset of identified synthetic polymers was investigated for polymer composition via µRaman spectroscopy (DXR2xi Raman Imaging Microscope, Thermo Fisher Scientific) and additional FTIR analyses. A total number of 18 sediment samples from different depths below riverbed level were examined. Overall, 2,756 microplastic particles could be recorded. Highest numbers were found in the sample at 0-2 cm depth below riverbed level in the sediment of Stover Beach (32,356 particles per kg dry sediment). In the sediment of the nature reserve Heuckenlock the highest concentration was found at a depth of 10 cm below riverbed level (3,835 particles per kg dry sediment). The lowest microplastic concentrations were found at the Köhlbrand site (minimum 1,114- maximum 2,776 per kg dry sediment) and in Wedel (minimum 743- maximum 3,133 per kg dry sediment). At the locations Heuckenlock, Köhlbrand and Wedel fragment dominate, while at Stover Strand fibers are the most common shape. Furthermore, a significant correlation between the biogenic organic matter content and the microplastic concentration could be detected.

Keywords : harbor , microplastic , nature reserve , sediment





# Microplastics in the upper tidal Elbe – A comparison between sediments from the Hamburg port area and an adjacent nature reserve

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### Introduction

Rivers have been identified as major pathways for plastic to marine environments, thus, they are of importance to understand the transportation and accumulation behavior of microplastic debris. Sediments may act as a sink for microplastic particles, especially when conditions of discharge and the underlying sediment substrate change within the river course

The upper tidal Elbe, passing the harbour city Hamburg and draining into the North Sea, has so far not been thoroughly investigated with regards to microplastic abundance in sediments. This study and its findings provide a first screening of the river's sediment microplastic concentration.

### Material & Methods

For the analysis of microplastic concentrations, the biogenic organic matter was removed by digestion ( $H_2O_2$  10 % and NaClO 6-14 %). In the following, the sample was wet sieved (mesh size 63 µm) and a density separation with NaI (density: 1.5 g/cm<sup>3</sup>) was conducted. The sample was introduced into a 1 I glass column which was subsequently filled with Nal solution (Fig. 2). For homogenization, the column was shaken twelve times and left for sedimentation for 10 minutes. The sedimented material was slowly removed via a tap at the base of the column. The remaining supernatant was filtered through a paper filter (413, VWR International, 5-13 µm Retention) using vacuum filtration. For identification, the Nile red staining method in combination with fluorescence microscopy (Axioscope 5/7 KMAT, Zeiss) was applied. A subset of particles was investigated for polymer composition via µRaman spectroscopy (DXR2xi Raman Imaging Microscope, Thermo Fisher Scientific). Additionally, particles not identified by Raman spectroscopy were analyzed via Fourier-transform infrared (FTIR) spectroscopy by Thermo Fisher Scientific. Contamination was evaluated using blank samples and values were substracted from sample results Parallel to the microplastic analysis, soil parameters like water content and organic substance were recorded



# Conclusion

- Microplastics could be identified in all samples
- A significant positive correlation between organic matter and particle concentration could be demonstrated
- River sections with low flow capacity could be possible hotspots for the accumulation of microplastic
- A wide occurrence of PS microbeads was documented
- For future studies, hydrological and pedological parameters (e.g. flow velocity and particle size analysis) should be recorded to examine further impacts on microplastic abundances in rivers

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# Study Area & Sampling

A total of 18 Elbe river sediment samples from different depths below ground level (bgl) were taken within the Hamburg port area, Germany, in cooperation with the Hamburg Port Authority (HPA) as part of the "Sullied Sediments" project funded by the European Union. The sites "Stover Strand" and "Wedel" are located upstream and downstream of the port of Hamburg, respectively, while "Köhlbrand" is located in its center. The samples were taken using a van-Veen sampler. At each sampling point 10-11 grabs were conducted resulting in sediment from 0-15 cm depth bgl. The first 0-2 cm from these grabs were removed using a stainless steel spoon and were immediately transferred into brown glasses. The residual sediment (2-15 cm depth bgl) was homogenized and three subsamples were again transferred into separate brown glasses at each location

Additionally, sediment samples were taken in the nature reserve "Heuckenlock" located also within Hamburg and at the southern river branch traversing the city and the port. Six samples were taken using a marching spoon in different depths bgl (10 to 60 cm) and were immediately transferred into brown glasses.



Fig. 1: Study area at the Elbe river within the wider area of the port of Hamburg. Red points mark the sampling sites.



### **Microplastic Abundance**

A total number of 2,756 microplastic particles could be documented. The results were converted into particles per kilogram dry sediment (per kg DS). The highest concentration of microplastics was found at Stover Strand in 0-2 cm depth bgl (32,356 particles per kg DS) and in the three mixed samples in 2-15 cm depth bgl (mean value ± standard deviation: 13,318 ± 5,907 particles per kg DS) (Fig. 3 and 6). The nature reserve Heuckenlock showed the second highest microplastic concentration (430 - ,385 particles per kg DS). (See Fig. 4). Lower microplastic concentrations were found at the Köhlbrand site (1,114 - 2,776 per kg DS) and in Wedel (743 - 3,133 per kg DS) (Fig. 6). In this study, a significant correlation between the amount of organic matter and microplastic concentration was detected (r=0.72, p-value < 0.001). This result suggests, that an increase in organic matter leads to an increase in microplastic particle concentration in the sediment. Furthermore, fragments were the dominant microplastic shape at all sites except for Stover Strand where fibres were prevailing (Fig. 6). The frequency of particles increases with decreasing size, although this is more prominent for fragments than for fibres. This could derive from the fact, that particles within the sediment further degrade into smaller size fractions



Fig. 6: Comparison of total particle concentration by shape and location (not ulated as kg per dry sec

### Spectroscopic Results

For the analysis via µRaman spectroscopy, 34 particles were analyzed from which 33 (97 %) were identified as plastic. The polymer types PS (41 % of all particles), PET (26 %) and PE (21 %) were identified as dominant polymer types (Fig. 7). EVAC, PP and PDMS were found in minor amounts (3 % each). High percentages of low density plastics in the sediment may result from biofouling on particles. Furthermore, 22 particles (11 fragments and 11 microbeads) not identifiable via Raman spectroscopy, were analyzed by FTIR spectroscopy. For the fragments PVC (36 %) was the predominant polymer type followed by PE (18 %), PET (9 %), PS (9 %). The rest of the fragments were not identified as plastic (27 %). Additionally, all 11 microbeads were identified as PS (100 %).



Microbeads

High numbers of microbeads have been discovered at all investigated sites. The polymer type of microbeads typically used in personal care products is predominately PE, more rarely PP or other polymers (Heger et al. 2014). In this study, the microbeads analyzed via µRaman and FTR spectroscopy were all identified as PS. Consequently, the microbeads in the sediments of the Hamburg Elbe River did not originate from personal hygiene products but rather have an industrial origin since other potential sources could be cleaning products, printer toners and industrial products such as abrasive media (Sundt et. al. 2014). As for PS microbeads in specific, they are also frequently used as ion-exchange resin beads (Scherer et. al. 2020)

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# **Results & Discussion**

# Organic matter digestion methods for microplastic extraction from estuarine samplings

Fertala Laila, Palazot Maialen, Soccalingame Lata, Kedzierski Mikaël, Bruzaud Stéphane.

Microplastics (MP) in the marine ecosystems is a global pollution of increasing scientific and societal concern. With the majority of plastics found in these ecosystems coming from land-based sources [1], through rivers and run-off, studies on MP has recently shifted focus toward freshwater ecosystems. The Tara Microplastics 2019 expedition collected samples from 9 of the main European rivers and revealed the presence of plastic particles in all the sampled sites. However, the samples contain a high amount of complex organic matter, hindering the characterization of MP. Therefore, developing methods to accurately and safely extract microplastics from field-collected waters is a key element to assess this pollution. Since literature varies greatly in the optimum method, there is a clear need for the development of a standardized, robust and efficient protocol. Standardizing a protocol will not only allow for efficient sample processing, but also promote consistency in data collection and analysis, and increase comparability between studies. In this work, two digestion protocols using hydrogen peroxide (H2O2)were tested. Protocol A combined H2O2 with an iron catalyst for a more advanced oxidation reaction known as the Fenton's reagent [2]. Protocol B consisted in two successive steps: first, a potassium hydroxide (KOH) treatment then a H2O2 digestion. Both protocols were completed with a density separation step to remove inorganic matter. Both protocols have shown to maintain MP physical integrity, allowed a clear polymer identification by infrared spectroscopy, and resulted in similar and high organic matter removal rates. Taking in consideration these results and the need to efficiently process a large set of samples, a new protocol is suggested combining those two methods, to be applied separately according to the nature of matter present within each sample.

Keywords : hydrogen peroxide , microplastic extraction , Microplastics , organic matter removal , rivers , Tara Microplastics 2019 expedition



# **RESULTS AND DISCUSSION**

- Weighting and visual inspection of MP showed that both protocols maintain MP physical integrity.
- Both protocols allowed the identification of the chemical nature of the polymers by infrared spectroscopy.



	кон → н₂о₂	Fenton's reagent
Effect on MP <ul> <li>Physical integrity</li> <li>Chemical integrity</li> </ul>	+ + + + +	+ + + + + +
Organic and inorganic matter removal efficiency	81%	89%
Time efficiency	+ (~72h in total)	+ + + (~4h)
Technician involvement	Minimum assistance	Full assistance

Although the Fenton's reagent is faster and slightly more efficient at digesting OM, it requires full assistance, hence the simultaneous processing of multiple samples is limited.

### CONCLUSION

Given the wide variety of the Tara 2019 mission samples, a new protocol was necessary to adapt to the varying concentrations and natures of OM

Taking in consideration the need to efficiently process a large set of samples, a new protocol is suggested. Firstly, all samples undergo a KOH digestion. Depending on the amount of remaining organic matter, more advanced digestion methods with H<sub>2</sub>O<sub>2</sub> may be applied successively.

This new protocol is more time efficient, avoids unnecessary steps and allows the processing of a high number of samples.



Tara's Blue Book: Tracing the Origins of Plastic Pollution (2020), 69 p.
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# Identification of environmentally degraded microplastics using the thermaldamaged plastics library

Ikezawa Yoshio.

Microplastics are classified into two types, primary microplastics and secondary microplastics. Primary microplastics refers to substances that are used as law materials in industrial abrasives, scrubbing agents, and so on. Polvethylene (PE) and polypropylene (PP) are frequently used in these applications. Secondary microplastics are usually generated when large plastic products are reduced to a fine size of 5 mm or less by external factors such as ultraviolet radiation in the environment. There are several analytical methods well known in the microplastic researches, and one of them is the Fourier transform infrared spectrophotometer (FTIR). FTIR is generally used in qualitative analysis of plastics and is already utilized in many surveys of the actual condition of microplastic discharges into rivers or sea. However, since actual microplastics are normally degraded in the environment, mainly by ultraviolet radiation, there may be less matches with the infrared spectra referring from a standard FTIR library. Degradation of plastics begins from the formation of carbon radicals with the dissociation of hydrogen from carbon-hydrogen bonds caused by the energy of heat or light. Reaction of the oxygen connecting to those radicals generates different components with bonds like O-H and C=O, which shows specific absorption spectrum. This reaction process causes cross-linking and molecular scission, resulting to the degradation of plastics. Due to the reaction in the plastics, the FTIR analytical result often shows some differences in the infrared spectra between standard plastics and degraded plastics. In order to provide highly reliable identifications, the thermal-damaged plastics library has been introduced and utilized for real microplastics sampled from the environment.

Keywords : analysis , degradation , FTIR , identification , library , thermal

# Identification of Environmentally Degraded Microplastics Using the Thermal-Damaged Plastics Library

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# 1. Introduction

Microplastics are classified into two types, primary microplastics and secondary microplastics. Primary microplastics refers to substances that are used as law materials in industrial abrasives, scrubbing agents, and so on. Polyethylene (PE) and polypropylene (PP) are frequently used in these purposes. Secondary microplastics are usually generated when large plastic products are reduced to a fine size of 5 mm or less by external factors such as ultraviolet radiation in the environment. There are several analytical instruments well known in the microplastic researches, and one of them is the Fourier transform infrared spectrophotometer (FTIR).

FTIR is generally used in the identification of plastics and is already utilized in many surveys of the actual condition of microplastic discharges into rivers or sea. However, since actual microplastics are normally degraded in the environment, mainly by ultraviolet radiation, there may be less matches with the infrared spectra referring from a standard FTIR library. In order to provide highly reliable identifications, the thermal-damaged plastics library has been introduced and utilized for real microplastics sampled from the environment.

# 2. Apparatus / Method

The compact FTIR, IRSpirit (Shimadzu Corporation), was used for the identification of microplastics with the QATR<sup>™</sup>-S ATR measurement accessory for the ATR method. ATR stands for Attenuated Total Reflection. An absorption spectrum of a sample surface can be obtained by placing the sample on the ATR prism and measuring the reflected IR beam at the sample surface. The light penetration depth in ATR is several µm.

# 3. Thermal-Damaged Plastics Library

The thermal-damaged plastics library is a library containing spectral data of 13 types of plastics in the unheated condition and when heated to various temperatures from 200 °C to 400 °C. Degradation of plastics begins Abs from the formation of carbon radicals with the dissociation of hydrogen from carbon-hydrogen bonds caused by the energy of heat or light. Reaction of the oxygen connecting to those radicals generates different components with bonds like O-H and C=O, which shows specific absorption spectrum. This reaction process causes cross-linking and molecular scission, resulting to the degradation of plastics. Due to the reaction in the plastics, the FTIR analytical result often shows some differences in the infrared spectra between standard plastics and degraded plastics.

In fact, although large differences in the progress of degradation can be seen in ultraviolet degradation and thermal degradation, the factors that govern the progress of degradation are essentially the same. So we can see that the changes observed in the infrared spectrum are often similar. Due to this experience, the thermal-damaged plastics library has been introduced and utilized for real microplastics sampled from the environment.

Right figure shows the infrared spectra of acrylonitrile butadiene styrene (ABS) resin when irradiated with ultraviolet light. In the case of ABS, peaks associated with stretching vibration of the O-H radical and C=O radical appear under ultraviolet conditions. Same experiment was tested under heat condition, 250 degrees for 2 h, and similar phenomena was observed in the spectrum.

# 4. Result

Among the microplastics collected at the sea coast, those with sizes of 5 mm or less were measured. 2 sample measurement results are shown below. From the results in left figure, a hit for polypropylene (PP) heated at 200 °C for 4 h was obtained from the thermal-damaged plastics library for the white microplastic, and from right figure hit for polyethylene (PE) heated at 200 °C for 2 h was obtained for the red microplastic. It can be inferred that both microplastics were degraded by oxidative degradation caused by ultraviolet radiation.



# 5. Conclusion

Microplastics collected at a sea coast were measured with a compact FTIR. Simple and easy measurement was possible by the ATR method. Quick qualitative analysis of degraded microplastics was also possible by using the thermal-damaged plastics library. But in case of desiring higher accurate identification, we recommend measuring plastic which has been intentionally degraded by ultraviolet irradiation and comparing that sample with the actual sample. This is our next challenging task to be solved.



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Adsorption of diclofenac and metronidazole on real microplastics of different nature and size *Munoz Macarena*, *Ortiz David*, *Nieto-Sandova l Julia*, *Gomez-Herrero Esther*, *De Pedro Zahara M.*, *Casas Jose A.*. Paper number 334303

**Industrial sources should be the priority control direction of microplastics in wastewater** *Long Zouxia, Wang Wenling.* 

Screening of cyanobacteria producing exopolymers as bioflocculant for microplastics water removal Faria Marisa, Henriques Sérgio, Cordeiro Nereida. Paper number 334754

**Blue-green microalgae-based exopolymers as an efficient bioflocculant for microplastics debris** *Faria Marisa, Ribeiro João, Kaufmann Manfred, Ferreira Artur Ferreira, Cordeiro Nereida.* Paper number 334868

# Adsorption of diclofenac and metronidazole on real microplastics of different nature and size

Munoz Macarena, Ortiz David, Nieto-Sandova l Julia, Gomez-Herrero Esther, De Pedro Zahara M., Casas Jose A..

Microplastics (MPs) are recognized as micropollutants carriers in the aquatic environment. Accordingly, the evaluation of their sorption capacity has received major attention in the last few years. Nevertheless, in most works, pure polymers have been used as surrogates for real MPs, which can lead to significant underestimation for their actual values. On the other hand, one kind of plastic has been usually tested, making difficult comparison purposes. In this work, MPs were obtained by cutting up commercial plastic products like bottles, containers and trays. The particles were further grinded using a cryogenic mill in order to achieve different sizes (25 – 1000 mm). Four representative plastic polymers were evaluated: polystyrene (PS), polypropylene (PP), high density polyethylene (HDPE) and polyethylene terephthalate (PET). Diclofenac (DCF) and metronidazole (MNZ) were selected as target pollutants given their widespread occurrence in the environment but also their significantly different properties (structure, hydrophobicity and size). The equilibrium adsorption experiments (5 days) were performed under ambient conditions using different pollutant concentrations (0.5 – 15 mgL-1). The reactor volume and MP mass were established at 1.5 mL and 10 mg, respectively. The experimental data were well-described by the Langmuir model, which suggests that monolayer adsorption took place in all cases. Nevertheless, important differences were found in the MPs adsorption capacity considering both their nature but also the kind of micropollutant tested. The MPs adsorption capacity followed the order: PS¿HDPE¿PP¿PET. On the other hand, regardless of the MP nature, DCF adsorption was significantly higher than that achieved for MNZ (450 vs. 20 ngg-1). The increase of hydrophobicity of the MP and/or the micropollutant led to an increase on the adsorption capacity, which suggests that hydrophobic interactions govern the adsorption mechanism. Finally, the MP adsorption capacity increased by decreasing the particle size, consistent with the increase of exposed surface area.

Keywords : adsorption , diclofenac , metronidazole , microplastic , pollutant carrier







# Adsorption of diclofenac and metronidazole on real microplastics of different nature and size

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# Industrial sources should be the priority control direction of microplastics in wastewater

Long Zouxia, Wang Wenling.

As one of the important point sources of environmental microplastic pollution, municipal wastewater has received extensive attention and recognition in recent years. However, little is known about the characteristic differences of microplastics between industrial and domestic wastewater. This study taken the Haicang wastewater treatment plant in Xiamen, China as a case study to compare the abundance, particle size, type and color of microplastics in industrial wastewater and domestic wastewater. It was found that the abundance of microplastics in industrial wastewater was more than twice that of domestic wastewater, and there were obviously differences in morphology, type and particle size distribution between them. Industrial sources contributed more to the microplastics pollution of municipal wastewater, and at the same time, more other pollutants were adsorbed by industrial-based microplastics, which may cause greater harm of compound pollution. It is recommended that industrial sources should be the priority control direction of microplastics in wastewater.

Keywords : Industrial sources , Microplastic , Priority control direction , Wastewater

# Screening of cyanobacteria producing exopolymers as bioflocculant for microplastics water removal

Faria Marisa, Henriques Sérgio, Cordeiro Nereida.

Discovered in the early of the 20th century, plastic has become a material used in all industry sectors as it is a low-cost, durable, inert, and corrosion-resistant product. Around 1 million plastic bottles are purchased every minute around the world [1]. In 2021, its expected that number reach half a trillion. with less than half of those bottles ending up getting recycled [1]. Unfortunately, most of this nonrecycled plastic is disintegrated into smaller sized low molecular-weight polymer fragments – microplastics - through various mechanical actions and degradation in the environment. Microplastics have become into a serious problem due to their potential negative effects - from its size to the chemicals released - on the marine ecosystem and biota. In the recent years, the scientific community turned its focus to the development of sustainable methodologies to remove them from the contaminated water [2]. The flocculation technique is the most conventional approach to remove the suspended solids in wastewater treatment with high efficiency. However, inorganic, and organic flocculants are employed in this process and most of them stay in the wastewater after treatment and may cause ecological damage [3]. Cyanobacterial-based extracellular substances have been shown encouraging properties suitable to be applied in industry as gums, bioflocculants, biosorbents and bioemulsifiers [4]. Following our interest on microbial-based biomaterials and the development of an efficient way to remove the microplastics of contaminated water, the main purpose of this research is to assess the bioflocculant activity of different cyanobacteria-based extracellular substances to remove plastic debris of contaminated water and compare with the bioflocculant activity of commercial extracellular substances. These assessments allowed the evaluation of the potential of cyanobacterial-based exopolymers as bioflocculants and their applicability in bioremediation.

Keywords : based extracellular substances , Bioflocculation , Bioremediation , Cyanobacterial , Microplastics

# Screening of cyanobacteria producing exopolymers as bioflocculants for microplastics water removal

# Marisa Faria<sup>a,b,c</sup>, Sérgio Henriques<sup>a</sup>, Nereida Cordeiro<sup>a,c</sup>

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# **INTRODUCTION AND GOALS**

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Microplastics have become into a serious problem due to their potential negative effects - from its size to the chemicals released on the marine ecosystem and biota. The flocculation technique is the most conventional approach to remove the suspended solids in wastewater treatment with high efficiency. However, inorganic, and organic flocculants are employed in this process and most of them stay in the wastewater after treatment and may cause ecological damage <sup>[1]</sup>. Cyanobacterial-based extracellular substances have been shown encouraging properties suitable to be applied in industry as gums, flocculants, sorbents and emulsifiers <sup>[2]</sup>. Following our interest on microbial-based biomaterials and the development of an efficient way to remove the microplastics of contaminated water, the main purpose of this research was to assess the bioflocculant activity of different RPS — release exopolymer substances - (*Gloecapsa* sp. – RPS Gsp., Nostoc commune - RPS NC, Cyanocohniella calida - RPS CC) to remove plastic debris from contaminated water and compare with the flocculant activity of commercial extracellular substances.



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**METHODS** 

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RESULTS

Figure 1. Bright field (A — stained with Alcian Blue) and SEM micrographs (B) of *Gloecapsa* sp., *N. commune* and *C. calida* cells of each extraction steps.

Figure 2. Bioflocculation activity: Xanthan Gum (1), Alginate (2), RPS NC (3), RPS CC 4), RPS Gsp (5), control (C).

 Table 1. Flocculation activity of different RPS— exopolymers under different concentrations of cations at 15, 30 and 60 min.

		Kaolin		1	<b>Microplastics</b>					
	Floc	culating activ	ity (%)	Flocculating activity (%)						
	15 min 30 min		60 min	15 min	30 min	60 min				
			1% CaCl2							
Alginate										
	45.1 ± 1.7	55.5 ± 0.6	69.7 ± 5.6	22.5 ± 8.2	24.2 ± 0.5	27.5 ± 0.2				
Xanthan gum	75.6 ± 16.6	78.3 ± 5.8	77.0 ± 10.2	25.9 ± 3.4	29.6 ± 2.5	35.2 ± 6.2				
RPS NC	19.3 ± 1.7	2.1 ± 0.6	32.7 ± 3.1	17.6 ± 0.1	27.2 ± 1.6	36.8 ± 7.4				
RPS CC	57.1 ± 4.4	61.5 ± 9.6	67.9 ± 0.9	15.5± 0.2	12.7 ± 2.2	23.1 ± 6.3				
RPS Gsp	15.8 ± 1.9	29.4 ± 9.1	24.0 ± 7.1	4.7 ± 0.1	7.7 ± 0.0	4.5 ± 0.1				
			1% FeCl3.7H	20						
Alginate	15.2 ± 1.3	26.3 ± 0.3	28.6 ± 2.0	21.0 ± 2.4	11.5 ± 3.8	4.4 ± 1.0				
Xanthan gum	2.6 ± 0.5	2.8 ± 0.8	3.5 ± 0.3	20 <mark>.3</mark> ±7.7	16.0 ± 2.3	13.4 ± 0.1				
RPS NC	1.0 ± 0.8	0.2 ± 0.157	1.7 ± 0.3	6.3 ± 0.1	11.0 ± 0.7	11.4 ± 4.3				
RPS CC	6.0 ± 2.0	3.3 ± 0.6	3.8 ± 0.5	2.48± 0.9	23.0 ± 0.7	9.9± 3.2				
RPS Gsp	1.8 ± 0.7	0.9 ± 0.8	0.4 ± 0.2	24.4 ± 5.0	28.4 ± 6.2	18.1± 7.1				

# CONCLUSIONS

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*Gloecapsa* sp. and *N. commune* cultures demonstrated to be a potential source of RPS with high flocculant activity to remove microplastics of contaminated water - applicability in bioremediation.

- RPS Gsp exhibited the highest bioflocculating activity to remove microplastics (about 28%) after 30 min with Fe<sup>3+</sup> cation.
- RPS NC displayed a bioflocculating activity to remove microplastics (about 37%) after 60 min when Ca<sup>2+</sup> cation was applied.

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# Blue-green microalgae-based exopolymers as an efficient bioflocculant for microplastics debris

Faria Marisa, Ribeiro João, Kaufmann Manfred, Ferreira Artur Ferreira, Cordeiro Nereida.

A poluição por microplásticos é a principal preocupação ambiental do século 21 devido ao aumento da produção global de plásticos e ao consumo excessivo. Novas estratégias sustentáveis são necessárias para remover esses resíduos plásticos de águas residuais usando materiais biológicos naturais por meio de abordagens limpas [2]. Polímeros extracelulares de microalgas verde-azuladas têm apresentado o potencial floculante promissor como alternativa aos recursos sintéticos devido às suas atrativas propriedades físicas-definidas [1]. No estudo atual, uma cianobactéria Gloecapsa sp. foi avaliada como uma fonte potencial de polímeros extracelulares (EPS), um sabre, polímeros de ligação celular (CPS) e polímeros liberados (RPS), para aplicações em floculantes.Diferentes métodos de extração e purificação foram registrados para CPS e RPS, os quais foram caracterizados em termos de propriedades morfológicas e relevantes. A atividade biofloculante também foi avaliada a partir do sobrenadante e culturas de Gloecapsa e comparada com a atividade biofloculante de produtos comerciais. Os resultados indicam que Gloecapsa sp. foi um bom produtor de EPS, com rendimento de CPS de 0,273 g L-1 e rendimento de RPS de 0,236 g L-1. O Gloecapsa predominante A natureza polissacarídica e aniônica do EPS confere propriedades floculantes atrativas e promissoras, específicas para aplicação no tratamento de efluentes, contribuindo para um ambiente sustentável e limpo. Referências [1] Cruz, D., Vasconcelos, V., Pierre, G., Michaud, P., Delattre, C. (2020) Exopolysaccharides from Cyanobacteria: Strategies for Bioprocess Development. Ciências Aplicadas, 10, 3763. https://doi.org/10.3390/app10113763 [2] Cunha, C., Faria, M., Nogueira, N., Ferreira, A., Cordeiro, N. (2019) Exopolímeros de microalgas marinhas vs de água doce como biossoluções para a tribos de microplásticos. Poluição Ambiental, 249, 372-380. https://doi.org/10.1016/j.envpol.2019.03.046

Keywords : bioflocculant , bond polymers , cell , CPS , EPS , exopolymers , Gloecapsa sp. , released polymers , RPS

# Blue-green microalgae-based exopolymers as an efficient bioflocculant for microplastics debris

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# INTRODUCTION

UNIVERSIDADE da MADEIRA

Microplastics (MPs) pollution is the major environmental concern of the 21st century due to the increasing global plastic production and excessive consumption. New sustainable strategies are required to remove these plastic debris from wastewater using natural biological materials through clean approaches <sup>[1]</sup>. Cyanobacterial-based exopolymers (EPS) have attractive physical-chemical properties that can be applied as bioflocculants, bio emulsifying and biosorbents<sup>[2]</sup>. These properties make EPSs a promising flocculant as alternative to synthetic resources. Gloecapsa sp. exhibited different types of EPS (i) cell-bound or capsular polysaccharides (CPS) and (ii) released polysaccharides (RPS). In the current study, different extraction and purification methods were applied for CPS and RPS of Gloecapsa sp... They were characterized in terms of morphological and structural properties. The bioflocculant activity was also evaluated from the supernatant and Gloecapsa cultures and compared with the bioflocculant activity of commercial products.

# METHODS





" Stirring 300 rpm, 60 min, 0D = 550 nm



Figure 1. Bright field (A1,B1,C1 - stained with Alcian Blue) and SEM micrographs (A2,B2,C2) of Gloecapsa sp. cells of each extraction steps.

Table 1. Bioflocculation activity of Gloecapsa sp. (culture and supernatant) exopolymers under different concentrations of cations at 30, 60 and 120 min.

		Microplastics		
		Flocculating activity (?	6)	
	30 min	60 min	120 min	
1% CaCl <sub>2</sub>				
Supernatant	6.341 ± 0.427	1.289 ± 0.243	7.361 ± 0.584	
Culture medium	8.967± 0.128	1.289 ± 0.243	4.507 ± 0.175	
1% FeCl <sub>3</sub> .7H <sub>2</sub> 0				
Supernatant	0.230 ± 0.046	9.844 ± 1.337	13.286 ± 2.269	
Culture medium	5.421± 0.875	9.889 ± 0.171	15.124 ± 3.300	
0.25% FeCl <sub>3</sub> .7H <sub>2</sub> 0				
Supernatant	31.050 ± 0.676	32.965 ± 1.558	30.212 ±2.407	
Culture medium	27.548 ± 0.225	29.459 ± 1.417	27.234 ± 0.601	

CPS PF

**RPS U** 

CPS UP RPS PE

2200

Figure 3. FTIR spectra of *Gloecapsa* sp. CPS and RPS.

2700

1700

(%)

3700



Figure 2. Bio flocculation activity of supernatant *Gloecapsa* sp. (A) and *Gloecapsa* sp. culture (B).



Figure 4. Agglomeration of MPs on Gloecapsa culture (x400). EPS were stained with Alcian Blue.

# CONCLUSIONS

The results indicate that *Gloecapsa* sp. was a good producer of EPS. The predominant *Gloecapsa* EPS's polysaccharide and anionic nature confers attractive and promising flocculant properties suitable to be applied in wastewater treatment, contributing for a sustainable and clean environment.

700

1200

### REFERENCES

[1] Cruz, D., Vasconcelos, V., Pierre, G., Michaud, P., Delattre, C. (2020) Exopolysaccharides from Cyanobacteria: Strategies for Bioprocess Development. Applied Sciences, 10, 3763. https://doi.org/10.3390/app10113763 [2] Cunha, C., Faria, M., Nogueira, N., Ferreira, A., Cordeiro, N. (2019) Marine vs freshwater microalgae exopolymers as biosolutions to microplastics pollution. Environmental Pollution, 249, 372-380. https://doi.org/10.1016/j.envpol.2019.03.046

# RESULTS

- CPS was removed successfully through reflux extraction;
- EPS showed a polysaccharide and anionic nature;
- CPS yield of 0.273 g L<sup>-1</sup> and RPS yield of 0.236 g L<sup>-1</sup> were obtained.
- A bioflocculating activity of 33% was achieved for Gloecapsa culture to remove microplastics of contaminated water.

Acknowledgements: Oceanic Observatory of Madeira Project (M1420-01-0145-FEDER-000001)

Interreg |





# Session 25.9\_Ma. Chaired by Aquilino Miguelez, Lanzarote

The Sinos River Watershed and Microplastic Pollution: current status, presence evaluation and perspectives

Panizzon Jenifer, Correa Marina Zimmer, Gehlen Günther, Sobral Paula, Jahno Vanusca Dalosto. Paper number 334443

**Microplastic ingestion by perch (***Perca fluviatilis* **Linnaeus 1758) from four Italian lakes** Galafassi Silvia, Sighicelli Maria, Cau Alessandro, Gillibert Raymond, Temperini Maria Eleonora, Ortolani Michele, Bettinetti Roberta, Pietrelli Loris, Pusceddu Antonio, Zaupa Silvia, Volta Pietro. Paper number 334563

# $M^2 \, {\rm \ll OSPAR} \, {\rm \gg}$ in a maximum visual accumulation of plastic debris in the Seine estuary National Nature Reserve

*Colasse Laurent.* Paper number 335075

# Sampling airborne microplastics and fibers: lessons learned from Aveiro, Portugal

C. Prata Joana, L. Castro Joana, P. Da Costa João, Duarte Carlos, Cerqueira Mário, Rocha-Santos Teresa.

Paper number 334322

# The Sinos River Watershed and Microplastic Pollution: current status, presence evaluation and perspectives

Panizzon Jenifer, Correa Marina Zimmer, Gehlen Günther, Sobral Paula, Jahno Vanusca Dalosto.

The Sinos River is the main water body of the Sinos River Basin. Located in the state of Rio Grande do Sul, Brazil, it comprises 32 municipalities and supplies water for more than 1.6 million inhabitants. Human activities are vastly diversified along the watershed, directly reflecting on its environmental quality once it recieves constant domestic and industrial untreated wastewater discharges. Given hydrological and geomorphological characteristics, the basin is subdivided into three sections: upper, middle, and lower. The first section refers to a region closer to the source of the Sinos River, with low anthropogenic influence and mainly rural land-use. The second section has a transition aspect and presents both rural and urban features. Finally, the lower section, has the highest level of anthropic pressure, is densely populated and highly industrialized. To date, studies evaluating the presence of microplastics in this important freshwater ecosystem are scarce. In this sense, aiming to assess the occurrence of microplastics in the Sinos River, surface water and sediment samples were collected at sites in Caraá (upper), Parobé (middle), and São Leopoldo (lower) sections along the riverbank. Samples were preserved in controlled conditions. After laboratory procedures, samples were vacuum-filtrated using cellulose acetate membrane filters. A quali-quantitative analysis was performed and according to specific literature, the filters were observed under a stereomicroscope and the particles were counted and classified. Precautions were taken in all steps to avoid external contamination. It was possible to observe several microplastics in all samples from the three sites analyzed. In general, fibres were the predominant shape found. These findings are part of a larger ongoing research developed at Feevale University and the final results will lead to important contributions regarding microplastic pollution at state and regional levels.

Keywords : Brazil , Environmental Quality , Rio dos Sinos Basin , Rio Grande do Sul



# The Sinos River Watershed and Microplastic Pollution: current status, presence evaluation and perspectives

Jenifer Panizzon\*<sup>1</sup>, Marina Zimmer Correa<sup>1</sup>, Günther Gehlen<sup>1</sup>, Paula Sobral<sup>2</sup> and Vanusca Dalosto Jahno<sup>1</sup> <sup>1</sup>Feevale University. ERS-239, 2755, Vila Nova, 93525-075, Novo Hamburgo, Rio Grande do Sul, Brazil <sup>2</sup>School of Science and Technology (FCT-NOVA), New University of Lisbon. Campus da Caparica, 2829-516, CAPARICA. Portugal

# Introduction

The Sinos River is the main water body of the Sinos River Basin. Located in the state of Rio Grande do Sul, Brazil, it comprises 32 municipalities and supplies water for more than 1.6 million inhabitants.

Study area

b.

Human activities are vastly diversified along the watershed, directly reflecting on its environmental quality once it recieves constant domestic and industrial untreated wastewater discharges.

To date, studies evaluating the presence of microplastics in this region are scarce

> The basin is subdivided into three sections ( • from right to left): upper, middle, and lower.

# Aims

1. Assess the occurrence of microplastics in the Sinos River:

2. Contribute to the scientific knowledge regarding the presence of microplastics in this important freshwater ecosystem.

# Materials and methods

C.

Surface water and sediment samples were collected at sites in Caraá (upper a.), Parobé (middle b.), and São Leopoldo (lower c.) sections along the riverbank. Samples were preserved in controlled conditions. After laboratory procedures, samples were vacuum-filtrated using cellulose acetate membrane filters. A quali-quantitative analysis was performed and according to specic literature, the filters were observed under a

stereomicroscope and the particles were counted and classified. Precautions were taken in all steps to avoid external contamination.

# **References:**

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BENVENUTI, T., KIELING-RUBIO, M. A., KLAUCK, C. R., RODRIGUES, M. A. S., 2015. Evaluation of water quality at the source of streams of the Sinos River Basin, southern Brazil. Braz. J. Biol. 75 (S2), 98-104. KONZEN, G. B., FIGUEIREDO, J. A. S., QUEVEDO, D. M., 2015. History of water quality parameters - a study on the Sinos River/Brazil. Braz. J. Biol. 75, 2, S1-S10

# **Results and discussion**

Microplastics were observed in all samples from the three sites analyzed. In general, fibres were the predominant shape found.

These findings are part of a larger ongoing research developed at Feevale University and the final results will lead to important contributions concerning microplastic pollution at state and regional levels.

# **Acknowledgements:**







# Microplastic ingestion by perch (*Perca fluviatilis* Linnaeus 1758) from four Italian lakes

Galafassi Silvia, Sighicelli Maria, Cau Alessandro, Gillibert Raymond, Temperini Maria Eleonora, Ortolani Michele, Bettinetti Roberta, Pietrelli Loris, Pusceddu Antonio, Zaupa Silvia, Volta Pietro.

Research on microplastic contamination has been mostly focused on marine environments and only recently attention has been paid also to freshwater environments. Among aquatic organisms, fish have received higher attention compared to other taxa in surveys for MPs detection. However, although publications on this topic are rapidly increasing, there is a high dispersion of data and, rarely, the same species is evaluated on a large number of different environments, allowing an analysis that can lay the foundations for its possible use as an indicator species for microplastic contamination of the food webs. In this study, the presence of microplastics in the digestive system of one of the most widespread and commercially exploited freshwater fish, the perch (Perca fluviatilis Linnaeus 1758), was investigated in four of the largest Italian South Alpine lakes: Como, Garda, Orta, and Maggiore. Microplastics occurred in 86% of the analysed specimens, with average concentrations ranging from  $1.24 \pm 1.04$  ind-1 in L. Como to  $5.59 \pm 2.61$  ind-1 in L. Garda. The isolated particles were mainly fragments, except in L. Como where films were more abundant. Most common polymers were polyethylene, polyethylene terephthalate, polyamide, and polycarbonate, although a high degree of degradation was found in 43% of synthetic particles, not allowing their recognition up to a specific polymer. Despite the high level of ingestion, MPs presence seems to do not affect fish body condition, evaluated though the hepatosomatic index and Fulton' body condition factor. Instead, an inverse linear relationship occurred between the number of ingested particles and fullness index, meaning that an empty stomach had a higher probability to contain microplastics. Presented data do not support the existence of a relation between probability of MPs ingestion and length or sex, as found for other fish species of either marine and freshwater environments.

Keywords : emerging contaminants , freshwater fish , MPs ingestion , perch , plastic polymers , water pollution

# Microplastic Ingestion by Perch *Perca fluviatilis*, Linnaeus 1758) from four Italian Lakes

Silvia Galafassi<sup>1</sup>, Maria Sighicelli<sup>2</sup>, Antonio Pusceddu<sup>3</sup>, Roberta Bettinetti<sup>4</sup>, Alessandro Cau<sup>3</sup>, Maria Eleonora Temperini<sup>5</sup> Raymond Gillibert<sup>5</sup>, Michele Ortolani <sup>5</sup>, Loris Pietrelli<sup>6</sup>, Silvia Zaupa<sup>1</sup>, Pietro Volta<sup>1</sup>

Dep. of Human and Innovation for the Territory; <sup>5</sup>Sapienza University of Rome, Department of Physics; <sup>6</sup>Sapienza University of Rome, Department of Chemistry <sup>1</sup>CNR Water Research Institute; <sup>2</sup>ENEA, Department for Sustainability (SSPT); <sup>3</sup>University of Cagliari, Department of Life and Environmental Sciences; <sup>4</sup>University of Insubria

# Background

Microplastic particles (MPs) carry several potential threats to the biota. Together with the physical damage that can occur during ingestion, MPs can release chemicals, both residues from the manufacturing process and pollutants adsorbed during atmospheric agents in the environments. Also, the biofilm on them can host pathogenic or antibiotic-resistant bacteria, potentially dangerous for the organism. Although publications on freshwater MP pollution are rapidly increasing, those in which the same species is sampled in a large number of different environments and those that measure the effects on health are still few. For these reasons the assessment of the different effects induced by natural MPs exposition still needs research efforts.

# Study sites

Four subalpine lakes (L. Garda, L. Como, L. Maggiore, and L. Orta) were sampled with 25 mm mesh gillnets during October 2018.

# Objectives

Evaluation of MPs ingestion in perch in different freshwater environments and their potential effects on fish health and body condition.



from the other lake Polyethylene and polyethylene terephthalate were the two most frequently present, followed by polystyrene,	Fishes from Garda	FI (Fullness index)	K (Fulton's body condition factor)	HIS (Hepatosomatic index)	TW (g)	TL (cm)	MPs (MPs fish <sup>-1</sup> )	Fish with MPs (%)	Fish with no MPs (n)	Fish (n)		Results
MPs fish-1	Lake inges	6.09	1.13	1.07	36.74	14.80	1.24	87	2	15	Como	
	sted more	3.55	1.02	0.81	43.38	16.01	5.59	95	1	19	Garda	
	MPs that	5.03	0.96	0.94	30.99	14.73	1.73	75	თ	20	Maggiore	
	those	4.88	0.82	0.86	17.96	12.79	2.75	88	ω	26	Orta	



No relation was found between MPs presence and length or sex of the fishes. Also, health status indexes as Fulton's body condition factor (K) and hepatosomatic index (HSI) did not show any significant correlation. Red/Pink

Brown

Transparent



# Conclusion

particles isolated.

Maggiore Garda Como

0%

20%

40%

60%

%08

100%

Beads Films Fragments

Orta

polyamide, and polycarbonate.

Como Garda Maggiore Orta

Fragments were the most abundant MPs shape, follower by films and beads (fibers were not considered). A high degree of degradation was present, that did not allow a precise polymer identification for 43% of the synthetic

MPs pollution revealed to be highly present in the Italian subalpine lakes, especially in L. Garda. Particles present in perch GITs were highly degraded, probably due to the permanence in the GIT harsh conditions. Negative correlation with the fullness index points out that also in perch, MPs ingestion can induce false satiety and interfere with feeding, as already reported for other organisms.

# 23-27 NOVEMBER 2020 LANZAROTE AND BEYON

FATE AND IMPACTS OF MICROPLASTICS: KNOWLEDGE AND RESPONSIBILITIES

INTERNATIONAL CONFERENCE

EYOND

# M<sup>2</sup> « OSPAR » in a maximum visual accumulation of plastic debris in the Seine estuary National Nature Reserve

Colasse Laurent.

La pollution plastique est visuellement choquante! C'est généralement le reflet multicolore de nos supermarchés, sur les berges et les rivages ... Nous avions l'autorisation officielle de prélever des échantillons sur un site incontournable d'une rivière: la réserve naturelle nationale de l'estuaire de la Seine en Normandie. La couche d'objets flottants échoués sur le haut rivage nous donne des résultats incroyables! Cela prouve qu'il faut aussi caractériser les déchets du fleuve (OSPAR / MSFD). Notre nouvelle présentation met en valeur visuellement ces déchets et la fragmentation avant la mer! Cette nouvelle approche sera simplifiée et sera importante pour réduire le flux de polymères.

Keywords : Estuary , Fragmentation , nurdles , primary microplastics , River

### **NIVERSITE** M<sup>2</sup> « OSPAR » in a maximum visual accumulation of plastic debris\* UPEC DOOSU SOS mal de Seine DE ROUEN in the Seine estuary National Nature Reserve

L.COLASSE<sup>a,b</sup> R.TRAMOY<sup>c</sup> J.GASPERI<sup>c</sup> B.TASSIN<sup>c</sup>



ONLY one square meter...

Almost a guarter of plastic pollution is from industrial pre-production pellets (Microplastics). \* this maximum visual pollution is not representative of the entire National Nature Reserve

**<u>CONTACT</u>** : Laurent COLASSE

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of 8kg (43 liters)

# Sampling airborne microplastics and fibers: lessons learned from Aveiro, Portugal

C. Prata Joana, L. Castro Joana, P. Da Costa João, Duarte Carlos, Cerqueira Mário, Rocha-Santos Teresa.

Airborne microplastics and fibers can easily be inhaled, which causes increasing concerns over their environmental concentrations and toxic effects. However, the difficulty of sampling air can be felt in the need for specific equipment (e.g. air samplers) as well as the presence of other particulate matter and potential contaminations from the laboratory air. The objective of this work is to identify potential solutions for routine quantification, based on works conducted in Aveiro, Portugal. A first challenge was separating natural from synthetic fibers. A myriad of laboratory tests showed this could not be easily achieved through density separation, chemical digestion, or the use of staining dyes. The identification of fibers as natural or synthetic can only rely on visual characteristics for routine quantification. Based on the observation of multiple textile fibers, a diagram of identification has been produced based on the more regular structure of synthetic fibers. However, the identification of fibers and microplastics was hindered by the presence of other particulate matter. A method based on the removal of organic matter by H2O2 and density separation by NaI was developed and tested, with recovery rates of 94%. The use of both developed techniques in real sampling efforts allowed further improvements in contamination control measures, identifying possible sources as unfiltered work solutions, air deposition of fibers released from paper towels and cotton lab coats, directly weighting filters, and accumulation of dust in the sample inlet requiring proper decontamination procedures. Together, these developments allow a simple, fast, and reliable procedure which can be applied to determine concentrations of airborne microplastics and fibers.

Keywords : airborne microplastics , contamination , sampling , synthetic fibers

# Sampling airborne microplastics and fibers: lessons learned from Aveiro, Portugal

Joana C. Prata<sup>1</sup>, Joana Castro<sup>2</sup>, João P. da Costa<sup>1</sup>, Armando C. Duarte<sup>1</sup>, Mário Cerqueira<sup>2</sup>, Teresa Rocha-Santos<sup>1</sup> 1. Centre for Environmental and Marine Studies (CESAM) & Department of Chemistry, University of Aveiro, 3810-193 Aveiro, Portugal 2. Centre for Environmental and Marine Studies (CESAM) & Department of Environment and Planning, University of Aveiro, 3810-193 Aveiro, Portugal

Sampling airborne microplastics and textile fibers, in both indoor and outdoor environments, presents methodological challenges despite their wide distribution in the environment<sup>1</sup>. These stem from relatively low concentrations, confounding particles, difficult identification, and cross contamination of samples. From trial and error, improvements were achieved in three key levels: strict contamination control measures, sampling and sample preparation, and particle identification.

# **Contamination Control**



6

Filters Burning glass fiber filters, storing and weighting in clean glass Petri dishes.

indoor air. Strict contamination control measures are required to produce reliable results<sup>2</sup>, with the particularity of sampling equipment decontamination<sup>3</sup>.



ough cleaning of

Working under the laminar flow hood, in a room with limited access, wearing



Solutions and materials

Filtering all working solutions, using all glass or metal materials with proper cleaning, covering with lids or aluminum



Conducting blanks throughout the process: field blanks, procedural blanks, and open filters for air deposition.

# Sampling and Sample Preparation

n lab coats.

### Sampling Removing organic matter Sampling indoor or outdoor air with a portable air sampler with quartz fiber filters at a flow rate of 5 L min<sup>-1</sup> over 48-h period<sup>4</sup> days<sup>4</sup>

# Transferring particles from the filter to a solution of 15% H<sub>2</sub>O<sub>2</sub> and incubating at room temperature (20°C) for 8

### **Density separation**

Filtering and transferring again to a solution of saturated Nal, vortexing, leaving to separate for 90 minutes, and filtering the supernatant<sup>4</sup>



xees: 1. Dris et al. 2017 <u>10.1016/j envpol.2016.12.013;</u> 2. Prata et al. 2021 <u>10.1016/j jharmat.2020.123660;</u> 3. Prata et al. 2020 <u>10.1016/j marpolbul.2020.111522;</u> et al. 2020 <u>10.1016/j marz.2019.11.022;</u> 5. Prata et al. 2020 <u>10.1016/j sciotemv.2020.137488</u>. Aledgement: This work was funded by Portuguese Science Foundation (FCT) through scholarship PD/BD/135581/2018 under POCH funds, co-financed by the European Social Fund an es National Funds from MEC. Thanks are due to FCT/MCTES for the financial support to CESMM (UDIPS0017/2020 + UDB/S0017/2020) through national funds.

FCT
**Poster session 25.10** 

## Session 25.10\_O. Chaired by Mateo Cordier, Guyancourt

On the harmonization of microplastic pollution levels in the Mediterranean Sea. A Metaanalysis

Simon-Sánchez Laura, Grelaud Michaël, Ziveri Patrizia. Paper number 334504

**Estimation of tyre and road wear particle emissions in urban aquatic systems** *Gutierrez Victoria, Boxall Alistair, Mcclean Colin.* Paper number 334415

**Towards the understanding of the distribution of microplastic particles in the ice cores** *Lobchuk Olga, Lazaryuk Aleksander, Grave Aleks, Chubarenko Irina.* Paper number 334468

### Microplastic contamination in snow from Val d'Aosta (Italian Western Alps)

Parolini Marco, Fresta Jacopo, Gibellino Cristina, Borgogno Franco, Antonioli Diego, De Felice Beatrice, Canuto Susanna, Albonico Carlo, Concedi Donatella, Romani Alessandra, Rosio Emanuela, Gianotti Valentina, Bongioanni Maurizio, Laus Michele, Ambrosini Roberto, Cavallo Roberto.

Paper number 334398

## On the harmonization of microplastic pollution levels in the Mediterranean Sea. A Meta-analysis

Simon-Sánchez Laura, Grelaud Michaël, Ziveri Patrizia.

The enclosed Mediterranean Sea is considered as one of the greatest accumulation area for plastic pollution. Still, the disparities in the different methodologies used by researchers for sampling, extracting and identifying microplastics have set a barrier for intercomparison of this pollution across the basin. This systematic review aims to present a harmonized database on the occurrence of these pollutants within different abiotic compartments of the Mediterranean Sea, analyzing the different, but similar, methodologies used to quantify their presence. The compiled data also includes the type of particles, polymer, size, potential sources and factors affecting their distribution. The results of this analysis provide an opportunity to interpret the observed discrepancies of microplastic concentrations reported in the Mediterranean Sea. While the analysis of the chemical and morphological characteristics of the particles reported for each environmental matrix (water column, seabed and beaches) sheds light on the transfer and accumulation patterns of these pollutants in the study region. Moreover, we identified a pressing need for international collaboration between Mediterranean countries, especially with North African countries, to provide a full picture of the microplastic pollution status in this basin.

Keywords : Mediterranean Sea , Microplastics , Review

# On the harmonization of microplastic pollution levels in the Mediterranean Sea. A meta-analysis.

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## BACKGROUND

The enclosed Mediterranean Sea is considered as one of the greatest accumulation area for plastic pollution<sup>[1]</sup>. Still, the disparities in the different methodologies used by researchers for sampling, extracting and identifying microplastics (MPs) have limited the accuracy the intercomparision of this pollution in the basin. In previous literature reviews, authors have provided a general overview on the abundance of MPs in Mediterranean rivers<sup>[2]</sup>, surface waters<sup>[3]</sup>, sediments<sup>[4]</sup> and their interaction with biota<sup>[5]</sup>.

The present review aims to:

- Integrate and harmonize MP pollution data from sediments and seawater bodies of the Mediterranean Sea, considering the methods used in the 79 peer-reviewed articles selected for this study.
- Identify areas with higher and lower research efforts to highlight the pressing research needs to understand the fate of MP within this basin.

Systematic review was conducted under the PRISMA Statement<sup>[6]</sup>.



## CONCLUSIONS

The available data on the occurence of MPs in the Mediterranean Sea indicate that these pollutants are effectively dispersed in the surface waters, while coastal sediments and specific offshore hotspots (i.e. Thyrrenian Sea <sup>[7]</sup>) present higher MPs abundances. However, the predominance of sampling stations in the coastal areas might be jeopardizing our general understanding of the MP fate within the Mediterranean basin. In the future, international collaboration between Mediterranean countries is needed to provide a full picture of the microplastic pollution status in this basin, especially in the open sea and the North African coasts.

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Institut de Ciència i Tecnologia Ambientals-UAB







## **Estimation of tyre and road wear particle emissions in urban aquatic systems** Gutierrez Victoria, Boxall Alistair, Mcclean Colin.

Tyre wear particles (TWP) which generate from the contact between the road surface and the tyre have gained increasing attention because of their significant microplastics ( $\leq 5$  mm) contribution to the environment. These particles will end up on the pavement surface where some parts will get trapped and others will enter the surface water through direct road run-off, wastewater effluent and overflows of the sewage system. The considerable amount of particles released to the environment can have toxicological effects on aquatic organisms, due to the chemicals added during the manufacturing process and to the adsorption of contaminants present in the environment. The lack of analytical methods for the detection and accurate quantification of tread wear particles in the environment makes monitoring campaigns and the implementation of mitigation strategies difficult or impossible. Therefore, this study aims to assess exposure to tyre wear particles in urban aquatic environments by analyzing their theoretical fate, transport and retention processes using an integrative modelling approach at high spatial and temporal resolution. Initially, the city of York in the UK was used as a case study. These findings may help to have better management in the future and an improved road run-off treatment.

Keywords : Surface water , TRWP , Urban





## Estimation of tyre and road wear particle emissions in urban aquatic systems

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#### INTRODUCTION

#### PRELIMINARY RESULTS

- Tyre abrasion as a result of the contact between vehicle tyres and the road has been identified as a major source of secondary microplastics in the environment.
- These particles can be transported by rainwater runoff into sewers and surface waters causing toxicity to aquatic organisms.

#### **AIM OF THE PROJECT**

The general aim of this study is to **assess exposure** to tyre and road wear particles (TRWP) in urban aquatic systems by analysing their emission pathways and fate processes at high spatial and temporal resolution.

Initially, the city of York in the UK was used as a case study. These findings may help to have better management in the future and an improved road run-off treatment.

#### METHODOLOGY

- ✓ The traffic data for the city of York was downloaded from the UK Department of Transport (https://roadtraffic.dft.gov.uk/local-authorities/202) and the city of York Council (https://www.york.gov.uk/).
- ✓ The wear and tear was calculated using the specific emission factors per vehicle-km method for urban roads.
- ✓ For the estimation of TRWP emissions in the city of York a high-resolution modelling approach developed in Matlab software (MATLAB ver. R2018a) was implemented.



#### Key factors presented in this model approach:

- Uses a bottom-up mechanistic approach (sourcepathway-receptor).
- > Provides highly resolved local exposure patterns.
- Accounts for local temporal variations in emissions (rainfall events).
- Full characterization of anthropogenic factors such as, sewage systems and waste water treatment plants.
- Division of the study area into sub-catchments (considered here as hydrological zones) and the urban aquatic systems into river sections (RS). These Hydrological zones (HZs) were delimited taking into account all the sections of the city's sewage network, therefore, the pollutants emitted in each HZ will be discharged into their corresponding river section.

• The total amount of TRWP emitted in the city of York was **108.71 tonnes/year** (TRWP emission per capita 0.50 kg/year).

The

 Total emission of TRWP to York's river system over the simulated year was 55.65 tonnes/year (Fig. 1).



Figure 1. Estimated total emissions of TRWP to York's river system during the year 2017 for Cars and Taxis, Buses and coaches, Light good vehicles (LGVs) and Heavy good vehicles (AII\_HGVs).

The river sections receiving the highest runoff emissions are OUSE1, OUSE5 and FOSS3, whereas the lowest emissions are OUSE3 and FOSS2 (Fig. 2).



Figure 2. Mean daily emissions of TRWP to York's river sections (OUSE1, OUSE2, OUSE3, OUSE4, OUSE5, OUSE6, FOSS1, FOSS2, FOSS3 and FOSS4) during the year 2017 per vehicle type (Cars and Taxis, Buses and coaches, Light good vehicles (LGVs) and Heavy good vehicles (AL\_HCVs).



rigure 4. Daily TRWP estimated emissions for the RS FOSS3 over the year 2017 in kg (in grey) and the rainfall pattern for the same year (plotted in blue in the secondary axis).

#### **FUTURE WORK**

The next step in the project is to implement an integrated time— and spatially-resolved surface water fate model proposed by Domercq et al., (2018) to assess the transport and fate of TRWP in the York river system.

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TRWP

concentrate in the Northeast and

Northwest of the city (hydrozones HZ8

emissions

Spatial variation of emissions

highest



er Hydrological zone and River section.

Temporal variation of emissions

Run off emissions vary proportionally to the rainfall frequency for traffic emissions.

Dry days = zero emissions.

Rainy days = the influence of the runoff pattern is predicted and higher emissions peaks are expected. (Fig. 4).

## Towards the understanding of the distribution of microplastic particles in the ice cores

Lobchuk Olga, Lazaryuk Aleksander, Grave Aleks, Chubarenko Irina.

High concentrations of microplastics (MPs) found in marine ice rise questions on the behavior of MPs particles while water freezes, and on the resulting vertical distribution of the particles within the ice. We have performed (i) laboratory tests and (ii) sampling of sea-ice cores (RSF project No. 19-17-00041) to obtain first empirical insights. In laboratory, samples of fresh and salty (35 ppm) water containing various types of MPs particles were frozen. MPs were fibers (polyester), flakes (solid PS), and spherules (foamed PS) of different size fractions, both floating and sinking in the water of the sample. Physically, fresh-water freezes when water column is stably stratified, and the ice forms and grows over quiescent fluid. Fresh-water ice in the experiments was solid, transparent, containing only a few MPs inside, often captured by the air bubbles. Contrary, seawater is permanently mixed due to thermal convection until the very freezing, and then mixing is enhanced by the brine release. Sea-ice is eventually inhomogeneous (layered, with brine channels), not transparent, with much more MPs inside.Sea-ice cores (13 cores, total length of about 6 m) were collected with a ring drill (inner diameter 16 cm) at the section across the Amursky Bay (Sea of Japan) on February 21-27 2020, within flat areas of the ice field; CTD probe was used to measure parameters of the water laver beneath the ice. The state of the ice field and water layer under ice were monitored during the preceding period (30 days). In the lab, the cores were cut into layers of approx. 5 cm thick, weighed (700-900 g), and melted at room temperature. Finally, 122 filters were obtained and examined under microscope. Additional ice cores were taken for layer-by-layer (5 cm thick) determination of the integral salinity and dissolved organic matter. First results are obtained.

Keywords : ice , laboratory tests , microplastic



Abstract number 334468

### Towards the understanding of the distribution of microplastic particles in the ice cores



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#### Laboratory experiment

freezing chamber, V=120 dm3 T=-18 °C 4 thermistors experimental volume -10 dm<sup>3</sup> heating plate, fan €

thermal insulation, EPS 10 cm

Containers were made from PS-foam (5 cm) (3 sections, 12 liters each: 20 x 20 x 30 cm). On the picture:

□ on the left - brine 35 psu (table rock salt);

on the right - distilled water.

Water is poured into dense PE bags.

The left bag leaked - experiments were continued only with distilled water.

Three 3 types of MP were added:

two sinking:

□ yellow PS

bundles of red polyester fibers

floating tiny pieces of PS-foam.

Laboratory experiment results:

all MP particles are trapped in the chains of

□ (a) MP particles were not crystallization

□ (b) were displaced from the ice. They are located substantially below the surface

Some yellow PS pieces remained floating on the surface

#### Introduction

Concentrations of MP in Arctic sea ice are by several orders of magnitude greater than those found in "garbage patches" at the surface of the Pacific Ocean [Obbard et al., 2014; Bergmann et al., 2017] and in its deep-sea sediments [Bergmann et al., 2017], thus the transport of microplastic in the ocean is determined not only by antropogenic presence, but also by hydrophysical processes (in this case, with the global thermohaline circulation) [Cózar et al., 2017].

There are currently no publications on the physical mechanisms of interaction between MP particles and sea ice. The paper [Obbard et al., 2014] presents the first evidence of MP particles presence in sea ice samples from the Chukchi Sea and the Beaufort Sea: all four studied cores (taken at stations separated by hundreds of kilometers) contained MP concentration ranging from 38 to 234 particles per cubic meter of water, that is several orders of magnitude higher than those found in in Atlantic waters north of Scotland (0.34 particles per cubic meter of water) or in waters of the North Pacific Subtropical Gyre (0.12 particles per cubic meter of water) [Obbard et al., 2014]. The paper [Obbard et al., 2014] also suggests that irregularly shaped particles and particles less dense than water are more effectively trapped in sea ice than silt and sand. Similar results were obtained in the laboratory of Helmholtz Center for Polar and Marine Research (Germany): mean MP concentrations of 2x106 particles/m3 in pack ice and 6x105 particles/m3 in landlocked ice were detected in ice cores from the Fram Strait (between Greenland and Svalbard).

This paper presents the results of:

(a) laboratory tests (samples of fresh and salt (35 ppm) water containing different types of floating and

sinking MP particles were frozen);

(b) ice cores taken from the Amur Bay (Sea of Japan).



We analyzed 13 sea ice cores collected during February 2020 in the Amur Bay (Sea of Japan) from three separate areas of the ice field; the total length is about 6 m.

Study area



Sampling

The cores were obtained with a ring drill (inner diameter 18 cm, outer diameter 22 cm) on flat (unhummocked) areas of the ice field. From January 21 to February 27 the ice thickness was measured 10-12 times with a measuring ruler (accuracy 0.5 cm), physical parameters in the water column were measured by CTD probe, the ice cores were taken 2-4 times for layer-by-layer (5 cm thick) determination of the integral salinity and dissolved organic



Each sea ice core removed from the ice field was placed on a metal tray to remove sea water and a part of brine (within 4-5 minutes). The core length (ice thickness) was measured and photographed. Then, two sheets of aluminum foil (44x80 cm) were spread on a wooden table (60x80 cm), the core was placed on these sheets and packed. At the next stage, plastic bags were used to pack the cores. The packed ice cores were placed in plastic boxes (20x38x56 cm) installed in the car's cabin. One box could store 2-3 ice cores (gasket material was used in addition).

Sea ice cores were delivered to the laboratory of the National Scientific Center for Marine Biology (NSCMB FEB RAS) by car and placed in a chest refrigerator (T-18C).



The work with ice samples was carried out under laboratory conditions. The cores were fragmented into sections of approx. 5 cm thick, placed in sterile zipp-bags, weighed (700-900 g) and melted at room temperature. Then, portions of water were passed through a vacuum filter unit. As a result, filters with a suspended matter were obtained.

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bubbles, i.e.

centers and

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Investigations are supported by the Russian Science Foundation, grant No. 19-17-00041

0.21 items/cm<sup>3</sup>

fibers

fragments

films





61

37

2





yellow

black

pink

orange

violet

«zebra»

blue



10

9

6

6

4

3

2



### Microplastic contamination in snow from Val d'Aosta (Italian Western Alps)

Parolini Marco, Fresta Jacopo, Gibellino Cristina, Borgogno Franco, Antonioli Diego, De Felice Beatrice, Canuto Susanna, Albonico Carlo, Concedi Donatella, Romani Alessandra, Rosio Emanuela, Gianotti Valentina, Bongioanni Maurizio, Laus Michele, Ambrosini Roberto, Cavallo Roberto.

A growing body of evidence has highlighted that plastic contamination affects both anthropic and natural ecosystems, including remote areas. Recent studies have highlighted the presence of microplastics (MPs) in soils or sediments collected in mountain and glacier environments. However, the information concerning the transport routes of MPs to mountain environments is still limited. Aerial transport and snow scavenging have been suggested as the main route of MPs deposition in mountain areas, but data supporting this hypothesis are scant. The present study aimed at exploring the presence of MPs in snow collected in four locations on the path of the Tor des Géants® (Val d'Aosta, Italian Westers Alps) with different characteristics in terms of accessibility and anthropic presence. Snow was sampled close to the Deffeves, the Miserin and the Cuney mountain huts, as well as at the col du Malatrà, in late summer 2019. Overall, 40 putative MPs were isolated from melted snow samples. Further µFTIR analyses confirmed that 45% of isolated items were MPs (length range 50 – 1,910 μm, average length 300 μm), 43% were cellulose fibres and 2% were wool fibres, while 10% were unidentifiable. The 39% of MPs were fibres, while the 61% were fragments. Polyethylene (39%) was the main polymer, followed by PET (17%), HDPE (17%), polyester (11%), LDPE (6%), polypropylene (5%) and polyurethane (5%). The concentration (MPs/L ± SE) of MPs in melted snow from Cuney ( $0.39 \pm 0.39$ ), Deffeyes ( $1.08 \pm 0.01$ ), Malatrà ( $1.45 \pm 1.45$ ) and Miserin  $(4.91 \pm 2.48)$  did not differ significantly among locations, but values reflected the accessibility and the anthropic presence of sampling sites. Our results demonstrated that MPs can be deposited to the ground by snow, which acts as a scavenger of these contaminants by the atmosphere and plays a crucial role in contamination of remote areas.

Keywords : Italian Alps , microplastics , mountain , snow











## MICRO2020 INTERNATIONAL CONFERENCE 23-27 NOVEMBER 2020 - LANZAROTE

## **MICROPLASTIC CONTAMINATION IN SNOW FROM VAL D'AOSTA** (ITALIAN WESTERN ALPS)

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AIM OF

Recent studies have documented the presence of microplastics (MPs) in remote areas, including soils or sediments collected in mountain and glacier environments. However, the information concerning the transport routes of MPs to mountain environments is still limited. The present study aimed at exploring the presence of MPs in snow collected in four locations of the Val d'Aosta (Italian Western Alps), with different characteristics in terms of accessibility and anthropic presence.

## **METHODS**

SNOW WAS COLLECTED BETWEEN 7TH AND 11TH OF SEPTEMBER 2019 OVER THE TOR DES GÉANTS® TRAIL RUNNING RACE PATH (VAL D'AOSTA WESTERN ITALY)

Snow was sampled in four locations close to the Cuney (2,652 m a.s.l; i.e., Cuney), the Miserin (2,582 m a.s.Imicr.; i.e., Miserin) and the Deffeyes (2,500 m a.s.l.; i.e., Deffeyes) refuge, as well as at Col du Malatrà (2,936 m a.s.l.; i.e., Malatrà).

Isolation and characterization of MPs was performed according to Coppock et al., (2017), with slight modifications.

Fourier Transformed Infrared (FTIR) microscopy (µ-FTIR) was performed using a Nicolet iN10 MX Infrared Imaging Microscope (Thermo Scientific) to characterize the polymeric composition of the isolated items.



THREE SNOW SAMPLES PER LOCATION WERE COLLECTED WITH A STAINLESS STEEL SPOON AND TRANSFERRED TO 2 L GLASS JARS.

## CONCLUSIONS

deposition, which acts as a scavenger of transport

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18 MPs (length range 50 – 1,910 µm, average length ~ 300 µm) with different polymeric composition were isolated from snow samples. 6%



The mean (±SE) MPs abundance in snow ranged between 0.39 ± 0.39 MPs/L and 4.91 ± 2.48 MPs/L but it did not differ among locations (Kruskal-Wallis test; x2 = 2.12, d.f. = 3, p-value = 0.54).



## Session 25.10\_Me. Chaired by Manuel Monge-Ganuzas, Urdaibai

**Bridging the gap: synthesis of microplastic research from field and laboratory studies, facilitating a better understanding of microplastic impacts on freshwater biota** *Kukkola Anna, Krause Stefan, Lynch Iseult, Sambrook-Smith Gregory, Nel Holly.* Paper number 334342

## Quantitative lipidome analysis of mouse serum exposed to microplastic using metabolic deuterium oxide labeling

*Kim Tae-Young, Park Jinyoung.* Paper number 334361

### Zero Plastic: joining efforts to fight plastic pollution in island and coastal Biosphere Reserves

Bethany Jorgensen, Aquilino Miguelez, Eva Cardona, Juan Baztan, Tony Gallardo, Pilar Pérez, Anastasia Barkusova, Katja Bonnevier, Mattias Holmquist, Jo Overty, Lázaro Márquez, Joseph Emmanuel Philippe, Julio C. Medrano,Kwang Sub Jang, Mercè Mariano, Nelson Davanadera, Fabien Boileau, Jean Jackes Barreau, Patrick Pouline, Darja Kranjc, Jorge Blanco, Manuel Monge-Ganuzas, and Nicolás García-Borreguero Paper number 339037

## Trends and extremes of microplastic pollution on North Atlantic coasts: key points of 14 years of observations 2007-2020

Baztan Juan, Carrasco Ana, Huck Thierry, Jorgensen Bethany, Miguelez Aquilino. Paper number 334861

**Volunteer beach cleanups: civic environmental stewardship combating global plastic pollution** *Jorgensen Bethany, Krasny Marianne, Baztan Juan.* Paper number 334855

# Bridging the gap: synthesis of microplastic research from field and laboratory studies, facilitating a better understanding of microplastic impacts on freshwater biota

Kukkola Anna, Krause Stefan, Lynch Iseult, Sambrook-Smith Gregory, Nel Holly.

Microplastics are known to pollute freshwater environments and interact with freshwater biota. However, the present understanding of their potential effects in the freshwater environment is limited, hindered by the mismatch between field and laboratory based studies. Our extensive review of published literature identified fibres as the major morphology found in organisms in the field, but these are currently lacking from laboratory-based toxicity studies which focus on spheres / granules. Certain plastic types are over-expressed in laboratory settings and don't match those identified in the field. To bridge these gaps, we recommend that field studies make more stringent efforts to report on plastics at the lower end of the size scale, and minimize conclusions based on visual identification only which miss large fractions of particles. For lab studies, expansion to more realistic plastic types / shapes should be prioritized. Both lab and field studies should improve reporting on the chemical composition of plastics, and include studies of leaching of cofounding factors, such as plasticizers and colorants, supplemented with chemical fingerprinting wherever possible. The plastics found under field conditions are typically larger than those used in toxicity testing, despite field evidence that bigger particles are capable of translocating in fish; the expansion of laboratory toxicity studies to bigger plastics sizes is encouraged. Finally, the endpoints used in toxicity testing varied greatly, making comparison between studies difficult. We recommend principal endpoints for three major trophic levels: for primary producers photosynthetic activity and growth should always be included, and as secondary endpoints gene regulation or stress responses can be explored. For primary consumers, principal endpoints should be growth and mortality with reproduction and multigenerational studies as secondary endpoints. For secondary consumers, growth, mortality and locomotion/behavioral changes should always be reported, supplemented with secondary sub-lethal endpoints such as AChE activity and oxidative stress.

Keywords : freshwater , Microplastics , principal endpoints , reporting criteria , reporting standards , toxicity endpoints

## Bridging the gap: synthesis of microplastic research from field and laboratory studies, facilitating a better understanding of microplastic impacts on freshwater biota

Anna Kukkola, Stefan Krause, Iseult Lynch, Gregory Sambrook-Smith, and Holly Nel School of Geography, Earth and Environmental Sciences, University of Birmingham, Edgbaston, Birmingham B15 2TT, United Kingdom

#### Objective

To map-out the current knowledge regarding micro,-and nanoplastic interactions in freshwater biota (Figure 1) with focus on challenges and solutions



Figure 1. Conceptual model showing the proposed direct and indirect microplastics in freshwater food-webs



Figure 2. Relative abundance of the different plastic size categories

### **Recommendations for field sampling**

- Collection of field samples in conjunction with biota samples
- Separation of organism to its constituents
- Usage of same digestion method
- Identification methods
- Recording physical characteristics of microplastics
- Reporting the smaller detection limit



Figure 4. potential principal and secondary toxicity endpoints to be included I future studies.

#### Methods

Literature search was conducted on ISI Web of Knowledge with the advanced key search equation. Full text analysis of obtained studies was subsequently conducted and screened for four compulsory elements: 1) Original study, 2) Fresh water study 3) Biota interaction and 4) Microplastics < 5 mm.

#### Findings

There is difference in size and morphology categories used/reported (Figure 2, Table 1) with cladocerans and fish over presented (Figure 3)

Type of study	Number of studies	Most common morphology	Most prevalent plastic type
Field	44	Fibres	PE, PP, PS, PET and PA
Laboratory	102	Granules/spheres	PS, PE, PP, 'others' and PET

Table 1. Table showing key findings between field and laboratory studies



Figure 3. Number of published microplastic interaction studies in freshwater biota groups.

#### **Recommendations for laboratory studies**

- Environmentally relevant media (i.e. river water)
- Food provision
- Setting baselines for:
- Effects of morphology
- Effects of chemical composition
- Effects of size

We recommend *principal endpoints* that should be

included in each microplastics effects study in the future (Figure 4)





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## Quantitative lipidome analysis of mouse serum exposed to microplastic using metabolic deuterium oxide labeling

Kim Tae-Young, Park Jinyoung.

The evaluation of the health risk of microplastics (MPs) on mammals has been limited compared to a number of studies on the toxicological effects of MPs on aquatic organisms. To bridge this knowledge gap, we use a mouse model to study the biological effects of long-term exposure to MPs at an environmentally relevant concentration based on the deuterium oxide (D2O) global omics relative quantification (DOLGOReQ) method recently developed by our group. In the experimental group, ICR mice were given normal drinking water containing MPs at a concentration of approximately 400 ng of polystyrene (50 nm) or polyethylene (1-4 µm) daily. Mice in the control group were fed drinking water rich in 5% D2O. After 5 or 24 weeks of the exposure to MPs, serum lipids were extracted at the expense of mice. Relative quantification of serum lipids by DOLGOReQ showed that 5 triacylglycerols were down-regulated in the polystyrene and polyethylene groups. In addition, 11 triacylglycerols were down-regulated and 3 sphingomyelins were up-regulated in the polyethylene group. Finally, we sought to interpret the changes in lipid species resulting from exposure to MP in the context of lipid metabolism.

Keywords : deuterium oxide , lipidomics , metabolic labeling , Microplastics , mouse

### Quantitative lipidome analysis of mouse serum exposed to microplastic using metabolic deuterium oxide labeling

Jin Young Park and Tae-Young Kim\*

dangwagi-ro, Buk-gu, Gwangju 61005, South Kore ol of Earth Scier nces and Envi





- > Study on the biological effects of long-term exposure (6 months) to microplastics (MPs) at an environmentally relevant concentration on a mouse model
- > Quantitative analysis of serum lipidome by deuterium oxide (D2O) global omics
- relative quantification (DOLGOReQ) method recently developed by our group

#### INTRODUCTION

- > The evaluation of the health risk of MPs on mammals has been limited compared to a number of studies on the toxicological effects of MPs on aquatic organisms
- > Four MP sources including drinking water, salts, shell fish, and fish were considered
- to calculate an environmentally realistic MP concentration > After 5 or 24 weeks of the exposure to MPs, serum lipids were extracted from mice
- and subjected to relative quantification with the control mice adminstrated with 5%  $D_2O$

#### **EXPERIMENTAL**



D2O physiological control group Control group Experimental group Eight-weeks-old male mice (ICR) were exposed to MPs for 5 and 24 weeks through drinking wate ncentration of MPs is approximately 400 ng/day of polystyrene (50 nm) or polyethylene (1-4 µm).

#### Concentration of MPs

#### > Source from Bottle water

Source	Most abundant MP	Reference size (µm)	ng /particle <sup>ª</sup>	MP intake (n/day)	MP intake (ng/day)
Single use bottle	PET(57%)	5~10(41%)	0.30	24.5	7.35
Returnable plastic bottle	PET(78%)	5~10(56%)	0.30	31.5	9.45
Glass bottle	PE (35%)	5~10(45%)	0.21	87.5	18.4
Beverage carton	PE (38%)	5~10(39%)	0.21	19.5	4.10
PET = Polyethyleneterephthalate; PP = Polypropylene; PE = Polyethylene					

≻ So	ource	from	Salt			≻ So	ource	from	Fish		
Source	Refer ence Size (µm)	Most Abund ant MP	ng /particle <sup>ª</sup>	MP Intake (n/day)	MP Intake (ng/day)	Source	Refere nce Size, μm	e Most Abund ant MP	i ng /particle <sup>®</sup>	MP Intake (n/day)	MP Intake (ng/day)
Salt	50	PET	89	0.44-0.54	39.16-48	sardine	50	PE	60	5.48	329.18
42.50											

#### Source from Shell fish

Source	Reference Size, μm	Most Abundant MP	ng /particle <sup>ª</sup>	MP Intake (n/day)	MP Intake (ng/day)
Mussel	35	PP	20	0.159	3.18
Manila Clam	50	PE	60	0.088	5.28
<sup>a</sup> Density of MPs (g/cm <sup>3</sup> ); PET = 1.36; PP = 0.91; PE = 0.93					Mean : 4.23

#### Total Concentration : 9.83 + 43.58 + 329.18 + 4.23 = 386.82 ng/day ≈ 400 ng/day

 $\sim$ 

Animal dose 12.3



#### LC-MS & MS/MS analysis

- > HPLC system: Agilent 1200 infinity series
- Column: Agilent Zorbax Extend-C18, 4.6 mm × 50 mm, 1.8 μm
- Solvent A: 10 mM ammonium formate in H<sub>2</sub>O/ACN (4:6, v/v)
- Solvent B: 10 mM ammonium formate IPA/ACN (9:1, v/v)
- Q-ToF: Agilent 6520 Quadrupole Time-of-Flight

#### Lipid identification by Lipid annotator

- ➤ Q score ≥ 30.0
- ➤ Mass deviation ≤ 10 ppm
- Fragment ion score ≥ 30
- ➤ Total score ≥ 60
- ➤ constituent level threshold ≥ 10%

#### Data processing for finding biomarkers

ILI. 111. Lipid Biomarker DOLGOReQ identification discovery



Figure 1. Lipid subclasses in each experimental group. Among 144-172 commonly identified lipids, 10 lipid subclasses were common in all experimental groups. TG, PC, and LPC were the top three mostly identified lipid subclasses.

#### Lipids significantly changed between control and experimental groups



Figure 2. Quantified lipids in each experimental group. The x-axis shows  $\log_2(fold change)$ and the y-axis shows  $-\log(p-value)$ .; |fold change| > 1.5; p value < 0.05; TG was found to be down-regulated in the H<sub>2</sub>O and polystyrene groups following 5 weeks of exposure, whereas SM was down-regulated in the polyethylene group. After 24 weeks of exposure, one LPC was downregulated in the polystyrene and polyethylene groups. In addition, 11 TGs were down-regulated and three SMs were up-regulated in the polyethylene group,

#### CONCLUSIONS

- > A long-time exposure to MPs at a low concentration up to 6 months resulted in quantitative changes in the lipidome of the mouse serum
- > Among the metabolism pathways that involved the significantly altered lipid classes, glycerolipid and glycerophospholipid metabolism were particularly notable.
- > We speculate that microbial changes in the intestine are factors causing these changes

#### ACKNOWLEDGEMENTS

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## Zero Plastic: joining efforts to fight plastic pollution in island and coastal Biosphere Reserves

Bethany Jorgensen, Aquilino Miguelez, Eva Cardona, Juan Baztan, Tony Gallardo, Pilar Pérez, Anastasia Barkusova, Katja Bonnevier, Mattias Holmquist, Jo Overty, Lázaro Márquez, Joseph Emmanuel Philippe, Julio C. Medrano, Kwang Sub Jang, Mercè Mariano, Nelson Davanadera, Fabien Boileau, Jean Jackes Barreau, Patrick Pouline, Darja Kranjc, Jorge Blanco, Manuel Monge-Ganuzas, and Nicolás García-Borreguero

Growing from the Zero Plastic campaign in Lanzarote, begun in 2009, the "Zero Plastic" working group was launched in May 2018 at the annual meeting for the World Network of Island and Coastal Biosphere Reserves (WNICBR) in Menorca. The Zero Plastic working group is an alliance between the WNICBR and the Marine Sciences For Society research network; connecting Biosphere Reserves, researchers and society as a whole in efforts to eradicate plastic pollution. Membership is open to any Biosphere Reserve willing to participate.

The aim of the group is:

- . Make it simple;
- . Take it easy;
- . Lead by example;
- . Turn off the tap;
- . Enough is enough.

These five working principles, +1, guide how we share efforts to fight plastic pollution, connect experiences, and create an open-access processes moving through knowledge and action. The first 19 Biosphere Reserves in the Zero Plastic working group are: Archipelago Sea Area, Blekinge Archipelago, Cat Ba Archipelago, Commander Islands, Fuerteventura, Gran Canaria, Islands of Iroise sea, Isle of Man, Jeju, Karst and Reka River Basin, La Hotte, Lanzarote, Marinas Corunesas e Terras do Mandeo, Menorca, Ometepe, Palawan, Peninsulade Guanahacabibes, Terres de l'Ebre and Urdaibai, representing 13 countries. Coastal and island Biosphere Reserves make visible the complex issue of plastic pollution. Plastic on our river beds and shores represents a serious symptom of larger problems; these sites serve as warning systems illuminating the limits of the planet. The working group experience echos Ostrom's call for policentric governance: Biosphere Reserves working with different social groups, organized or not, across scales and arrangements. The world's remote coasts indicate our plastic-based civilization is out of balance with the biosphere, but we find hope in efforts to learn from the past and experiment in the present for a plastic pollution-free future.

Keywords:Biosphere Reserves, working group, policentric governance, plastic

## Trends and extremes of microplastic pollution on North Atlantic coasts: key points of 14 years of observations 2007-2020

Baztan Juan, Carrasco Ana, Huck Thierry, Jorgensen Bethany, Miguelez Aquilino.

The supervision of a Master's thesis in 2007 made it possible to link observations from beaches in Brittany-France and the Spanish-Basque coast, launching formallythe microplastic sampling process presented in this paper. In 2008, a first series of observations from Lanzarote and New York beaches highlighted (i) the impact of microplastic pollution at the North Atlantic scale and its asymmetry; (ii) the importance of having a first diagnosis as a baseline; and (iii) the need for a long-term time series. Thousands of observations have accumulated over the past 14 years confirming the working hypothesis of seasonal variability of plastic pollution (Baztan et al., 2016) and the role of local shore conditions. The regular sampling launched on 3 beaches since 2014: Lostmarc'h (Finistère, France), Famara (Lanzarote, Spain) and Popham (Maine, USA), along with the collection of more data on all latitudes of North Atlantic shores ground-truths the trends and extremes of plastic pollution, key points in microplastic sampling methodologies and highlights the related participatory action research processes.

Keywords : beaching , extremes , microplastic , North Atlantic , plastic pollution , trends

## Volunteer beach cleanups: civic environmental stewardship combating global plastic pollution

Jorgensen Bethany, Krasny Marianne, Baztan Juan.

Marine litter, the majority of which is plastic, is one of the most pressing global environmental challenges impacting the planet. One way coastal communities respond to this challenge is through the environmental stewardship practice of vol- unteer beach cleanups. Bevond providing temporarily cleaner local beaches, how might these beach cleanups have broader impacts in the global struggle against plastic pollution? Using the lenses of environmental stewardship and civic ecology, we conducted a content analysis of primary source materials created by 50 groups involved in volunteer beach cleanups. We collected data on the scale at which groups coordinate volunteer beach cleanups, the roles they play in conducting these cleanups, how they interact with other volunteer beach cleanup groups, and the other forms of stewardship they conduct, if any. Within our sample, we identified groups coordinating volunteer beach cleanups at five geographic scales: local, subnational, national, multi-national, and global. Within the groups operating at each scale, we found groups conducting environmental stewardship in the forms of education, advocacy, research, and monitoring in addition to their conservation work through beach cleanups. Our findings demonstrate that groups branch out their impacts by combining different forms of environmental stewardship targeting plastic pollution, and they collaborate to scale up their actions in ways that contrib- ute to plastic pollution governance. Connecting these findings with the literature on the broader impacts of civic ecology practices allows us to theorize how volunteer beach cleanup groups branch out and scale up their efforts to weave a global net of ocean stewardship.

Keywords : Beach cleans , Civic ecology , Marine litter , Ocean stewardship

## Session 25.10\_Ma. Chaired by Melanie Pöhlmann, Bayreuth

**Cigarette butt litter reduction initiative with voting ballot box unit at Alexandra Beach, Sunshine Coast Australia** *Foley Alison.* Paper number 329634

### Plastics in organic fertilizers of waste management systems

Steiner Thomas, Zhang Yuanhu, Möller Julia, Löder Martin, Laforsch Christian, Agarwal Seema, Greiner Andreas, Freitag Ruth. Paper number 334152

## A sticky situation – quantifying interactions between microplastic particles and cells Wieland Simon, Groß Wolfgang, Spreng Sarah, Lehmann Moritz, Gekle Stephan, Laforsch Christian, Kress Holger.

Paper number 334257

## Cigarette butt litter reduction initiative with voting ballot box unit at Alexandra Beach, Sunshine Coast Australia

Foley Alison.

Cigarette butts are the world's most common type of marine debris. They have been documented to negatively impact biodiversity, ecosystems, human health, and the economy. In order to address this, we conducted a social experiment of cigarette butt littering behaviour modification through the employment of a Cigarette Butt Voting Ballot Box to encourage appropriate and safe disposal of cigarette butts in a targeted area at Alexandra Beach, Sunshine Coast Australia. Utilising the Honolulu Strategy and in keeping with the United Nations Sustainable Development Goal 14, the initiative was an opportunity to educate the public as to the environmental and punitive consequences of improper disposal and generate awareness across the Sunshine Coast Region. It was also an opportunity to demonstrate collaborative partnerships between government, non-profit, local citizens and private sector organisations in response to the prevalence of this form of waste through the employment of behaviour modification initiative to achieve a 70% reduction in cigarette butt litter at this location at the conclusion of the trial.

Keywords : behaviour modification , Cigarette Butts , Community Activation , Grassroots Action , Honolulu Strategy: DPSIR , Literature Review , littering behaviour , Sunshine Coast Australia , Ten Little Pieces , UNEP MOOC Marine Litter

### **Plastics in organic fertilizers of waste management systems**

Steiner Thomas, Zhang Yuanhu, Möller Julia, Löder Martin, Laforsch Christian, Agarwal Seema, Greiner Andreas, Freitag Ruth.

As part of the SFB 1357 – Microplastics and MiKoBo [1], we investigate the formation of macroand microplastics in technical systems of waste and water management. Necessary for a circular economy is the use of organic fertilizers from waste management plants like biogas and composting plants. Here, for example, organic waste from households, renewable resources, green waste and many more are used as substrate. With the produced organic fertilizers, the plants are an interface to the terrestrial environment. Substrates obtained from organic households and green waste are often contaminated with plastic waste like garbage bags, packages, and other plastics, its fragments entering the environment [2]. The technical systems are implementing different approaches for the preparation of the substrates and the sorting out of foreign materials. Our projects cooperate with fourteen technical plants in Germany, that use different process engineering, substrates and catchment areas. We studied the organic fertilizers from these different plants regarding their contamination with plastic particles. [1] MiKoBo – Mikrokunststoffe in Komposten und Gärprodukten aus Bioabfallverwertungsanlagen und deren Eintrag in Böden erfassen, bewerten, vermeiden (Ministerium für Umwelt, Klima und Energiewirtschaft Baden-Württemberg) [2] Weithmann, N., Möller, J. N., Löder, M. G., Piehl, S., Laforsch, C., & Freitag, R. (2018). Organic fertilizer as a vehicle for the entry of microplastic into the environment. Science Advances, 4(4), eaap8060.

Keywords : biogas , compost , microplastics , MiKoBo , organic fertilizer , plastics , SFB1357 , terrestrial environment , waste management systems

## A sticky situation – quantifying interactions between microplastic particles and cells

Wieland Simon, Groß Wolfgang, Spreng Sarah, Lehmann Moritz, Gekle Stephan, Laforsch Christian, Kress Holger.

During the last decades, the production, use, and disposal of plastic products have experienced an unprecedented increase. As a consequence, by now microplastic particles (MPPs) can be found in a multitude of ecosystems all around the world. Increasingly, reports about negative effects of MPPs on organisms and ecosystems are published. A major concern is the translocation of MPPs into cells and, therefore, the interactions between MPPs and cells have gained attention in recent years. However so far, little is known about the mechanisms and factors affecting the attachment of MPPs to cells, which is a key determinant for their interactions. Here, we introduce a microfluidic device that enables high-throughput quantification of MPP-cell interactions. With a Poiseuille flow in a rectangular channel, we can exert defined forces on particles attached to cells, which are adhered to the bottom of the channel. The forces were quantified by Lattice-Boltzmann simulations. Following automated detection of the particles by a cross-correlation algorithm and classification by an artificial intelligence, the numbers of ruptured particles at different hydrodynamic forces were evaluated. We measured attachment strengths between various mammalian cell lines and pristine and functionalized polystyrene MPPs, and also MPPs that were previously incubated in freshwater and saltwater. We found that MPP attachment to cells was increased for functionalized MPPs and MPPs with an ecocorona, compared to pristine MPPs. Conclusively, our research has the potential to pinpoint the critical factors determining MPP-cell interactions. This not only might support risk assessment of potential toxicity of MPPs for organisms and ecosystems, but also facilitate the development of potentially safer, less sticky polymers, enabling a more sustainable use of plastic products.

Keywords : attachment forces , cell particle interaction , ecocorona , microfluidics , microplastic beads

Day 4, Thursday 26th. November 2020						
9h-10h	26.1_O 26.1_Me 26.1_		26.1_Ma			
10h-10h15	26.1_Gaia: 3 sessions brief					
10h30-11h30	26.2_O 26.2_Me 26.2_M					
11h30-11h45	26.2	2_Gaia: 3 sessions	brief			
12h-13h	<b>26.3_O</b>	26.3_Me 26.3_Ma				
13h-13h15	26.3_Gaia: 3 sessions brief					
13h15-14h	26_Highlight, The Editor's perspective					
14h-15h	26.4_O 26.4_Me 26.4_Ma					
15h-15h15	26.4_Gaia: 3 sessions brief					
15h30-16h30	<b>26.5_O</b>	26.5_Me	26.5_Ma			
16h30-16h45	26.5_Gaia: 3 sessions brief					
17h-18h	26.6_O	26.6_Me	26.6_Ma			
18h-18h15	26.6_Gaia: 3 sessions brief					
18h30-19h	Poster.26.7_O	Poster.26.7_Me	Poster.26.7_Ma			
19h-19h30	Poster.26.8_O	Poster.26.8_Me	Poster.26.8_Ma			
19h30-20h	Poster.26.9_O	Poster.26.9_Me	Poster.26.9_Ma			

## Day 4/5, Thursday 26th November 2020

## Session 26.1\_O. Chaired by Cristina Panti, Siena

**More threats for the Mediterranean monk seal: presence of microplastics in their diet** *Hernandez-Milian Gema, Tsangaris Catherine, Anestis Anastasios, Panou Aliki.* Paper number 334383

### Microplastic particles in road de-icing salt

Rødland Elisabeth, Okoffo Elvis, Rauert Cassandra, Heier Lene Sørlie, Lind Ole Christian, Reid Malcom, Thomas Kevin, Meland Sondre. Paper number 334391

## Effects of polyethylene microplastics on benthic invertebrates and ecosystem function in lotic mesocosms

Silva Carlos, Machado Ana Luísa, Campos Diana, Rodrigues Andreia, Patrício Silva Ana L., Gravato Carlos, Pestana João L.t.. Paper number 334409

## Investigating the seasonal importance on the composition and concentration of microplastics in coastal seawater, beach sediments and the wild mussel (*Mytilus edulis*)

Paradinas Lola, James Neil, Quinn Brian, Dale Andrew, Narayanaswamy Bhavani E. Paper number 334448

## More threats for the Mediterranean monk seal: presence of microplastics in their diet

Hernandez-Milian Gema, Tsangaris Catherine, Anestis Anastasios, Panou Aliki.

For the very first time microplastics were systematically analysed in feaces of the endangered Mediterranean monk seal (Monachus monachus) in Greece, as a top marine predator. This study was carried out in a joint project by the Hellenic Centre for Marine Research (HCMR) and the Greek NGO Archipelagos – environment and development within the framework of the project "PLASTIC BUSTERS MPA". Twelve samples were collected in monk seal caves along the southwestern coasts of Zakynthos island, Ionian Sea, Greece, according to a special protocol elaborated for this purpose. The set of samples was analysed at the HCMR. A total of 208 particles were detected 42 of which (19.2%) were identified as organic material and were discarded from the analysis. The length of anthropogenic particles ranged from 45µm to 28,763µm, though 75% of the particles ranged from 100 to 3000µm. Interestingly, 34.2% of the particles were smaller than 1,000µm and only 10.9% were larger than 5,000µm. Most of the particles identified were filaments (n=140, 84,3%), followed by fragments (n=24, 14.5%). Transparent particles were the most abundant ones (39.8%), followed by blue ones (39.2%). Red particles occurred in 7.8%, while green and black occurred in 6.0% and 3.0% respectively. Most of the transparent and blue particles were filaments (92.5% and 89.2% respectively). The majority of the particles identified were polyamides (73.8%), followed by polycarbonate (14.8%), and polypropylene (6.6%). Because all faeces samples analysed contained microplastics the exposure of monk seals to this pollutant might be a cause of concern. Further investigations are necessary for drawing firm conclusions.

Keywords : Greece , microplastics , Monk seals , Tirrhenian Sea

### Microplastic particles in road de-icing salt

Rødland Elisabeth, Okoffo Elvis, Rauert Cassandra, Heier Lene Sørlie, Lind Ole Christian, Reid Malcom, Thomas Kevin, Meland Sondre.

The largest source of microplastic particles to environment is estimated to be roads, through the wear and tear of road surface and release of tire particles, road particles and road markings. During the winter in cold climates, the road surface may freeze and cause icing on the roads. For traffic safety reasons, de-icing of the road surface is important and road salt is applied for this purpose. Knowledge of microplastic (MP) contamination in road salt has, until now, been lacking. This is contrary to the increasing number of studies of microplastics in food-grade salt. In this study, road salt was investigated as a potential new source of road-related microplastic particles to the environment. Two analytical techniques were employed in this study; Fourier-Transform Infrared spectroscopy (FT-IR) and Pyrolysis gas chromatography mass spectrometry (GC-MS). Polymer content in four types of road salts, three sea salts and one rock salt, was investigated. The particle number of MP in sea salts (range 4-240 MP/kg, mean  $\pm$  s.d. = 35  $\pm$  60 MP/kg and rock salt (range 4-192 MP/kg, 424 ± 61 MP/kg, respectively) were similar, whereas, MP mass concentrations were higher in sea salts (range 0.1-7650  $\mu$ g/kg, 442 ± 1466  $\mu$ g/kg) than in rock salts (1-1100  $\mu$ g/kg, 322 ± 481 µg/kg). A total of 96% of the concentration (µg/kg) and 86 % of number of particles/kg came from black rubber-like particles, which appeared to be attributable to wear of conveyer belts used in the salt production. The contribution of MP from road salt in Norway was estimated to 0.15 tonnes/year (0.003% of total road MP release), 0.07 tonnes/year in Sweden (0.008%) and 0.03 tonnes/year in Denmark (0.0004-0.0008%). The study shows that road salt is a source of MP, however, it is a negligible source compared to other road-related sources.

Keywords : FTIR , GC , MS , Plastic pollution , Pyrolysis , road , salt

## Effects of polyethylene microplastics on benthic invertebrates and ecosystem function in lotic mesocosms

Silva Carlos, Machado Ana Luísa, Campos Diana, Rodrigues Andreia, Patrício Silva Ana L., Gravato Carlos, Pestana João L.t..

Given the ubiquitous presence of microplastics (MPs) in freshwaters and their concerning levels in riverbeds, it is of utmost importance to gather ecotoxicological data on macroinvertebrate community and ecosystem functioning. Using indoor artificial streams, this study evaluated the effects of polyethylene microplastics (PE-MPs, one of the most common polymers found in freshwaters; concentration range: 0.1 - 1 - 10 g/kg) on macroinvertebrate benthic community and ecosystem functioning (using primary production and leaf decomposition - as functional endpoints ) after 10 days of exposure. Plus, MP internal concentration was assessed for several invertebrates. The presence of PE-MPs in freshwater sediments altered the macroinvertebrate community structure, namely by the reduction in the abundance of grazers and fine sediment eaters such as Chironomus, Baetis and Ephemerella. Such reduction can be related to the ingestion of MPs, which was higher on these functional feeding groups (i.e., grazers and collectors). There were no alterations on the functional parameters analyzed, primary production and leaf decomposition. Results show that invertebrates' feeding strategies are the main drivers for MPs ingestion and effects. The absence of changes in primary production and leaf decomposition might be related to functional redundancy within invertebrate communities under short-exposure assays. Determination of MP after 10 days of exposure confirms ingestion by several benthic invertebrates highlighting the potential deleterious ecological effects of microplastics to freshwater and nearby terrestrial ecosystems. Thanks are due to FCT/MCTES for the financial support to CESAM (UIDP/50017/2020+UIDB/50017/2020, through national funds) and the research project ComPET (POCI-01-0145-FEDER-030361, funded by FEDER, through COMPETE2020 - Programa Operacional Competitividade e Internacionalização (POCI) and by national funds (OE)).

Keywords : benthic invertebrates , freshwaters , indoor artificial streams , plastic pollution

# Investigating the seasonal importance on the composition and concentration of microplastics in coastal seawater, beach sediments and the wild mussel (*Mytilus edulis*)

Paradinas Lola, James Neil, Quinn Brian, Dale Andrew, Narayanaswamy Bhavani E.

Microplastics are particles ranging in size from 1µm to 5mm, observed in natural ecosystems globally. While a better understanding of their distribution and effects on biota are increasing, the dynamic of microplastics in an active environment have not yet been extensively studied. It is hypothesised that the coastline is a transitional area where plastic particles are accumulating on the shore, in biota or released into the ocean and atmosphere. Microplastics have been studied temporally in seawater, fauna and sediments but the outputs have not proved to be convincing; some studies have highlighted a seasonal change whilst others the opposite. Here we present a quantification and characterisation of microplastics within mussels, seawater, and sediment in six sites located along the North and West coasts of Scotland. The combined effect of sites and seasons was investigated to explain the microplastics fluctuations along the shore. Despite the variations between sites, microplastics were recorded at all seasons. Of the digested mussels (n=240), 1030 particles were analysed with 15% of them being microplastics. Concentrations ranged from  $0.19 \pm$ 0.05 to  $1.38 \pm 0.64$  particles per g.wwt flesh (mean  $\pm$  SE). Fragments and fibres were the prevalent shapes observed, covering the main plastic families such as polyester, polystyrene, polyethylene and polyamide. In water samples, ¿70 microplastics were identified from 60L collected with concentrations ranging from 0.60  $\pm$  0.24 to 1.75  $\pm$  0.48 particles per L (mean + SE). Fibres, fragments and films were observed comprising mainly polyester and polypropylene. Over 36 glass jars (10 g each) of sediment were analysed, containing 19 microplastics. Concentrations were ranging from  $0.02 \pm 0.02$  to  $0.16 \pm 0.04$  particles per gram of dry sand. Future work will examine microplastics found within seawater and sediment during a lunar cycle to assess the effect of tides on the accumulation on the shore.

Keywords : biota , coastal area , concentrations , microplastics , seasonal variation , seawater , sediment

## Session 26.1\_Me. Chaired by Mateo Cordier, Guyancourt

**The dietary bioavailability of nanoplastics to salmon using an ex vivo gut sac technique** *Clark Nathaniel, Mitrano Denise, Khan Farhan, Boyle David, Thompson Richard.* Paper number 334189

**Effects of microplastics in marine organic matter production and transformation.** *Boldrini Amedeo, Loiselle Steven, Galgani Luisa.* Paper number 334216

## Abundance and microplastics ingestion by commercial shrimp *Pleoticus muelleri* at an impacted coastal environment (Southwestern Atlantic)

Fernandez Melisa, Forero López Ana Deisy, Colombo Carolina Victoria, Ardusso Maialen, Buzzi Natalia Sol, Rimondino Guido Noe. Paper number 334408

**Exposure of** *Sparus aurata* **to a microplastic enriched diet under laboratory conditions** Alomar Carme, Sanz Marina, Ripolles Vincent, Ríos Beatriz, Albertí Ines, Compa Montse, Álvarez Elvira, Deudero Salud. Paper number 334344

## Bioavailability of phenanthrene and pyrene onto microplastics in the aquatic environment and simulated digestive fluids

*Liu Yang, Breider Florian.* Paper number 334385

## The dietary bioavailability of nanoplastics to salmon using an ex vivo gut sac technique

Clark Nathaniel, Mitrano Denise, Khan Farhan, Boyle David, Thompson Richard.

The dietary bioaccumulation potential of nanoplastics to adult fish remains unclear due to analytical constraints. However, labelling of plastics with inorganic metals allows for quantitative assessments of nanoplastic uptake via inductively coupled plasma mass spectrometry (ICP-MS). By using palladium-doped nano-polystyrene particles (Pd NPs), the aim of this study was to determine if (1) nanoplastics in the fish gastrointestinal tract (GIT) and transported across the gut wall, and (2) identify the region of the GIT where uptake occurs. Salmon (n = 16) were sacrificed and the entire GIT was removed. The GIT was separated into four functional sections (stomach, anterior-intestine, mid-intestine and hind-intestine) and rinsed with physiological gut saline. Each compartment was filled with physiological gut saline containing a known amount of nanoplastics. Control gut sacs with no added Pd were also performed to validate the gut sac method. Gut sacs were closed, placed into physiological gut saline to start the experiment. After 4 h, the tissues were rinsed and the gut tissue was separated into the mucosa and muscularis. Tissues were dried, digested and analysed for total Pd concentrations by ICP-MS. In the control tissue samples, there was negligible background Pd ( $6 \pm 2$ ng/g). Nevertheless, gut sacs performed with Pd NPs contained Pd (indicating presence of nanoplastics) in all tissue layers and across the four sections. Most of the Pd, and presumably nanoplastic, was localised to the mucosa (80-98%), regardless of gut region. However, the underlying muscularis is more representative of active uptake. Within the hind-intestine a significant 1.9-3.2 fold higher concentration was found compared to the stomach and mid-intestine ( $104 \pm 19$ ,  $32 \pm 8$  and  $55 \pm 5$  ng/g, respectively). In summary, the presence of Pd in the muscularis suggests nanoplastics are taken up in fish GIT, and that the hind-intestine is the site for greatest transport.

Keywords : bioaccumulation , Fish , gastrointestinal tract , uptake

**Effects of microplastics in marine organic matter production and transformation.** Boldrini Amedeo, Loiselle Steven, Galgani Luisa.

Plastic pollution, in particular micro and nano plastics, has a range of impacts on aquatic and terrestrial environments. One area where new insights are needed is the effect on key microbial processes that control the carbon cycle (marine organic matter production and degradation). The present study explores plastic-related changes in organic matter in a controlled laboratory experiment with a non-axenic culture of the diatom species Chaetoceros socialis. The phytoplankton culture was grown in three different 2-L flasks: one served as control and contained the algae, heterotrophic bacteria, and the growing medium prepared with artificial seawater. The other two flasks were additioned with particles of similar concentration and size ( 30-µm): polystyrene microspheres in one, and SiO2 particles in the other, to compare the effects of microplastics and inorganic particles. Following the algal growth phase, the water from the three flasks was filtered to remove the algal cells and the particles. A 5-day dark experiment was performed on these new samples containing only bacteria and dissolved organic matter. Optical changes in CDOM (Chromophoric Dissolved Organic Matter) were monitored daily to examine CDOM quality and degradation in the three systems. CDOM absorbance (as a proxy for concentration) was higher in the control samples with respect to samples from both particles-enriched systems. This suggests different rates of CDOM production and degradation in the presence or absence of particles, with possible lower CDOM degradation in the control. The further comparison among CDOM indicators of microbial activity, such as spectral slope and slope ratio, showed that CDOM from the particle-enriched systems was more subject to microbial alteration, pointing to a higher microbial turnover of this material, and especially in the presence of inorganic particles (SiO2). These results indicate the potential role of particles, and among these, microplastics, to modify marine microbial organic matter dynamics.

Keywords : CDOM , marine organic matter , Microplastics

## Abundance and microplastics ingestion by commercial shrimp *Pleoticus muelleri* at an impacted coastal environment (Southwestern Atlantic)

Fernandez Melisa, Forero López Ana Deisy, Colombo Carolina Victoria, Ardusso Maialen, Buzzi Natalia Sol, Rimondino Guido Noe.

Microplastics (particles of size < 5 mm) are recognized as emerging pollutants that have been found in most of the world's aquatic environments. They are of particular concern due to their capability to be ingested, enabling bioaccumulation and/or bio-magnification through marine food webs. Focusing on Argentina's coastal wetlands, the Bahía Blanca Estuary (BBE) is considered one of the most important in this country due to its high economic impact and also for being habitat of important commercial species like macrocrustaceans and fishes. The shrimp *Pleoticus muelleri* is an important economic resource for artisanal fishing and highly consumed in Argentina. For this reason, we investigated the abundance and characteristic of MPs in the abdominal muscle and gastrointestinal tract of the shrimp P. muelleri, as well as in the surface waters from the inner zone of BBE. The results showed that the dominant shape of MPs were fibers both in surface waters and shrimps. The fibers' colors were mainly blue and black in surface waters, while in shrimps, they were transparent and black. The mean abundance of MPs in surface water and the shrimp's abdominal tissues were 31.03 ± 3.01 items.L-1 and 3.91 items. g-1 wet weight, respectively. Infrared Spectroscopy suggests that these fibers found in both environmental matrices correspond to semi-synthetic cellulose-based remains, poly(amide), polyethylene (PE), and polypropylene (PP). Therefore, the information of this study generates new knowledge about the materials of the sewage discharges, which should be considered for stakeholders in the management and conservation of this large coastal wetland.

Keywords : estuary , microplastics , seafood , shrimp

## **Exposure of** *Sparus aurata* **to a microplastic enriched diet under laboratory conditions**

Alomar Carme, Sanz Marina, Ripolles Vincent, Ríos Beatriz, Albertí Ines, Compa Montse, Álvarez Elvira, Deudero Salud.

During the last years, microplastic (MP) ingestion has been quantified in marine species both with an ecological and commercial interest at sea and under experimental conditions. Growing evidence indicates MP ingestion can produce both physical harm such as internal abrasion as well as chemical harm such as cancerinogenesis, endocrine disruption, amongst others, associated to chemicals that are added during manufacturing processes or sorbed to their surface once in the marine environment (Rochman et al., 2013). Moreover, enzymatic activation of Glutathione-S-Transferase (GST), which is used in the detoxification system of species, has been quantified in commercially important species exposed to plastic pollution (Alomar et al., 2017) confirming that there is a physiological effect related to MPs ingestion. Given the ubiquity of plastic polymers in the marine environment (Compa et al., 2018) and their susceptibility to sorbed contaminants with a potential of being transferred along the food chain, including humans, it is important to assess MPs ingestion in a commercially important species. For the purpose of this research, Sparus aurata (gilt-head (sea) bream) was exposed to an enriched diet with virgin LDPE and marinanted LDPE over the course of three months. After this feeding period, gilt-head (sea) bream were exposed to a detoxification period of one month to investigate if Sparus aurata is able to recover after being fed with MPs. Preliminary results demonstrate that Sparus aurata are ingesting both virgin and marinated MPs but with no apparent effect in their body condition, however physical damage is observed due to this ingestion. Acknowledgments: Laboratorio de Investigaciones Marinas y Acuicultura and J.M. Valencia. Research project ACUIPLASTIC, ref CTM2017-88332-R funded by the Programa Estatal de Investigación, Desarrollo e Innovación, Spanish Government. References: Alomar, C. et al., 2017. Environmental Research, 159: 135–142. DOI: https://doi.org/10.1016/j.envres.2017.07.043. Rochman, C.M., et al., 2013. Sci. Rep. 3 (3263). http://doi.org/10.1038/srep03263

Keywords : exposure , LDPE , physiological effects
# Bioavailability of phenanthrene and pyrene onto microplastics in the aquatic environment and simulated digestive fluids

Liu Yang, Breider Florian.

The adsorption and desorption behaviors of phenanthrene and pyrene onto polyethylene (PE), polypropylene (PP), polystyrene (PS), and poly (methyl methacrylate) (PMMA) microplastics in the simulated freshwater and digestive fluids (i.e. simulated gastric fluid (SGF), simulated intestinal fluid (SIF)) were studied. The second-order kinetic model described the sorption kinetics of polycyclic aromatic hydrocarbons (PAHs), and the Langmuir model fitted the sorption isotherms of PAHs better, which indicated the surface sorption as the main mechanism. The sorption capacity was in the order of PMMA ¿ PS ¿ PE ¿ PP, which may due to the specific surface area and particle size of microplastics. Various environmental conditions were considered, including pH, salinity and dissolved organic matters (DOM). Although higher salinity would bring negative effects on the adsorption of PAHs onto microplastics, the adsorption behaviors were not significantly influenced by pH and DOM. The desorption rate of PAHs from microplastics in SGF was lower than that in the SIF, with the desorption equilibrium time of 12 h in SIF. This study compares for the first time the desorption PAHs from microplastics in freshwater and two kinds of digestive fluids. The results of our work suggest that microplastics to could contribute significantly to the exposure of aquatic organisms to PAHs when these particles go through the digestive tract. Further studies are needed to quantify the toxicity threshold of aquatic organisms through ingestion of microplastics combined with persistent organic pollutions.

Keywords : Adsorption , Desorption , Digestive fluids , Microplastics , PAHs

### Session 26.1\_Ma. Chaired by Juan Baztan, Crozon

**Transport of polyamide microplastics at the sediment-water interface – First results from mesocosm studies** Nal Holly, Schneidewind Llwe, Kukkola Anna, Sambrook Smith Createry, Lunch Icault, Krause Sta

*Nel Holly, Schneidewind Uwe, Kukkola Anna, Sambrook-Smith Gregory, Lynch Iseult, Krause Stefan.* Paper number 334435

**Degradation and half-life of biodegradable plastic films on different marine beach sediments** *Eich Andreas, Lott Christian, Weber Miriam.* Paper number 334538

**Abundance and distribution of microplastics in the German bight – Do ships leave skid marks?** *Dibke Christopher, Fischer Marten, Scholz-Böttcher Barbara.* Paper number 334566

Characterizing microplastics formed from photodegraded plastics placed in simulated moving water conditions

*Maurer-Jones Melissa, Hebner Tayler.* Paper number 334572

## **Transport of polyamide microplastics at the sediment-water interface – First results from mesocosm studies**

Nel Holly, Schneidewind Uwe, Kukkola Anna, Sambrook-Smith Gregory, Lynch Iseult, Krause Stefan.

Microplastic particles are increasingly found in freshwater environments in a range of sizes, shapes, amounts and compositions. While recent studies have focused on the detection and quantification of microplastics in various river and estuarine environments, researchers have just begun to unravel the underlying transport and retention processes for the different microplastics particles. Of special interest in perennial rivers is microplastic transport and retention in the hyporheic zone, i.e. the biogeochemically active upper part of the riverbed and banks. To better understand the fate of polyamide fragments and fibers in riverine sediments, we conducted mesocosm studies at the Environmental Change Outdoor Laboratory (ECOLAB) facility of the University of Birmingham, UK. Twelve recirculating flumes (dimensions are 200 by 42 by 15 cm) were filled with precharacterized gravel or sand sediment and 45L of water and flow velocities were recorded in each flume with a flowmeter. During one-day-flow experiments, the flumes were either exposed to polyamide fragments (sizes: 150-250 um and 400-600 um, pre-stained with Nile Red), red nylon fibers (500 um, 1.7 dtex) or a mix of both. Samples (20 mL) were taken over a course of up to 24 hours at three locations per flume at predefined intervals. After filtration on 0.45 um GF/D filters they were analyzed using a Zeiss Stemi 2000 stereomicroscope. Results showed that both fibers and fragments settled out of the water column over the course of the experiments and formed distinct depositional pattern on the sediment. While larger fragments mostly settled out within the first 30 minutes, smaller particles of 150 um or less as well as fibers remained in the water column significantly longer. Results will serve as input to further studies of these specific microplastics on resuspension and long-term behaviour under various environmental conditions.

Keywords : deposition , mesocosm , recirculating flume , transport

# Degradation and half-life of biodegradable plastic films on different marine beach sediments

Eich Andreas, Lott Christian, Weber Miriam.

The substitution of conventional materials by biodegradable plastic is a possible countermeasure against the accumulation of plastic in the environment. Biodegradability of a material should be proven as part of a risk assessment. The actual degradation speed of these materials depends on the environmental settings, which include the concentration of enzymes, microorganisms, temperature, pH, humidity and oxygen and light availability. Most of the seafloor is covered with sediment. Sediment properties as permeability, oxygenation and nutrient content directly or indirectly depend on the grain size, as do the microbial community and the biogeochemical processes linked to it. To gain more insight into the effects of different sediments on the degradation rates of biodegradable plastic, we performed two iterative tank experiments. In a pilot study, marine sediment from one location was divided into four grain size fractions. In a follow-up experiment, sediment was collected from four different locations. Plastic disintegration was tested for a commercially available biodegradable plastic film (Mater-Bi HF03V), LDPE as a negative control and polyhydroxyalkanoate copolymer (PHA) as a positive control. In the pilot experiment, it was shown that disintegration rates of biodegradable plastic films were inversely correlated with grain size. Fastest rates were measured for mud, followed by fine to medium-fine sand (63 - 500 µm) and lastly the coarse.

Keywords : Bi , Biodegradation , environmental conditions , grain size , half life , Mater , PHA , sediment , tank experiment

## Abundance and distribution of microplastics in the German bight – Do ships leave skid marks?

Dibke Christopher, Fischer Marten, Scholz-Böttcher Barbara.

Microplastics (MP, particles from 1 µm to 5 mm) are a complex class of contaminants. They are highly diverse in polymer type with a multitude of potential sources and are ubiquitous pollutants in the marine environment. Here, abundance and distribution of MP are determined by its sources of entry and subsequent complex physical, chemical, and biological interactions. Mass related MP data provided by thermal methods enable polymer specific geospatial and/or temporal studies independent of any particle appearance or size. Data acquisition in defined regions gives insight into general horizontal and vertical MP composition and changes. Potential sources can be tracked and located within the area of interest. In this first mass related survey of MP in surface waters (2.5 m depth) of the German Bight, North Sea the spatial MP load, its potential sources and temporal variation were comprehensively examined. For this purpose the most relevant 10 consumer plastics were analyzed using Py-GCMS/thermochemolysis as an excellent method for qualitative and trace level polymer specific mass quantitative analysis [1, 2]. The overall MP concentration in surface waters of the German Bight ranges between 2 and 1400 µg m-<sup>3</sup>. Neither its concentration nor type distribution was homogenous. Besides meteorological and oceanographic conditions of substantial influence on spatial and temporal mass loads, this variability was attributed to the particulate nature of MP as well. While detected polymer distribution patterns of some areas were closely related to packaging waste others attracted attention by polymers closely related to marine (antifouling) coatings. First estimations of their abundance indicate that they as marine sourced MP invert the widely cited 80% terrestrial - to 20% marine based debris ratio for MP at least for the studied area. [1] Fischer M, Scholz-Böttcher BM. 2017. ES&T, 51, 5052–5060. [2] Fischer M, Scholz-Böttcher BM. 2019. Anal Meth, 11, 2489-2497

Keywords : geospacial distribution , marine coatings , mass quantitative data , Microplastics , North Sea , source tracking

# Characterizing microplastics formed from photodegraded plastics placed in simulated moving water conditions

Maurer-Jones Melissa, Hebner Tayler.

Aquatic plastic debris experiences environmental stressors that lead to breakdown into smaller micro-sized plastic particles. This work quantified microplastic formation with the environmental stressors of UV irradiation followed by mechanical strain induced by movement of water with an emphasis on connecting our results to changes in the materials chemical/physical properties. Polypropylene, polyethylene, and polyethylene terephthalate thin films and polypropylene injectionmolded sheets were irradiated with 254 nm UV light, placed into aquatic microcosms, collected through sieving, and counted under a microscope. Results showed increasingly more particles in smaller size classes, the smallest being 74-177 µm. Mechanical strain from the turbulent water caused 2.3-3x more microplastics to be formed for the thinnest ( 25 µm) film and 1.4-2x more for thicker films and sheets. The most common morphology of microplastics was fibers, particularly in thicker polypropylene samples, which was attributed to absorbance of the photons and the changes observed in the crystallinity and glass transition as measured with differential scanning calorimetry (DSC). When irradiated for 24, 48, or 72 h, longer irradiation resulted in more microplastics formed by polypropylene films, which correlated with changes in the glass transition temperature as measured by DSC and the extent of oxidation as measured with FTIR. Irradiation at 300 nm produced fewer microplastics due to slower kinetics of phototransformations. Overall, this work evaluates the impact of combined photodegradation and water motion toward microplastic particles formed. It provides quantitative evidence that mechanical strain of water movement exacerbates photo-induced formation of microplastics and shows that the existence of fibers in natural systems can be the result of photodegradation.

Keywords : aquatic plastic debris , fibers , microplastic , photodegradation

### Session 26.2\_O. Chaired by Arnaud Huvet, Plouzané

### Exposure to polyethylene microplastics with or without herbicide significantly affect valve activity and growth of oyster, *Crassostrea gigas*

Bringer Arno, Tran Damien, Dubillot Emmanuel, Le Floch Stéphane, Receveur Justine, Cachot Jérome, Thomas Hélène.

Paper number 334309

### Occurrence of microplastics in pacific oysters from aquaculture areas in Santa Catarina Island, Brazil: preliminary results

brocardo giulia, saldaña-serrano miguel, nogueira diego, vieira khauê, bainy afonso. Paper number 334366

#### Biodiversity and structure of plastisphere in lentic ecosystems

Di Pippo Francesca, Venezia Cristina, Sighicelli Maria, Pietrelli Loris, Di Vito Stefania, Nuglio Simone, Rossetti Simona. Paper number 334550

### A global study reveals rare and unexplored bacteria that are specific to the plastic biofilm and reoccur across habitats

Scales Brittan S., Cable Rachel N., Duhaime Melissa B., Gerdts Gunnar, Kreikemeyer Bernd, Pedrotti Maria-Luiza, Gorsky Gaby, Oberbeckmann Sonja, Labrenz Matthias. Paper number 334694

## Exposure to polyethylene microplastics with or without herbicide significantly affect valve activity and growth of oyster, *Crassostrea gigas*

Bringer Arno, Tran Damien, Dubillot Emmanuel, Le Floch Stéphane, Receveur Justine, Cachot Jérome, Thomas Hélène.

Microplastics and pesticides are found in coastal waters around the world today. Their high uses and production make them emerging pollutions. These pollutants are likely to have an impact on the physiology of marine organisms, such as the oyster (Crassostrea gigas). In this present study, we monitored the valve activity of oysters, C.gigas, exposed for 24 days, following a 7-day acclimatization period, to polyethylene (HDPE 20-25 µm) microplastics (MPs, 10 µg MP/L) alone or combined with the herbicide, chlortoluron (30 µg/L). The valve activity of juvenile oysters (12month-old) was determined using four parameters. HFNI technology (high frequency and noninvasive valvometer) allows us to study the daily growth of the oyster shell thanks to the electrodes attached to the two oyster valves. In addition, the HFNI system makes it possible to measure the number of micro-closures (stress factor in bivalves), the Valve, Opening Duration (VOD) and Opening Amplitude (VOA). We observed a significantly higher frequency of micro-closures in oysters exposed only to MPs after six days and until the end of the exposure. No significant effect was observed on the VOA for ovsters exposed to MPs alone. VOD was significantly reduced for oysters exposed only to MPs. For the cocktail conditions (MPs + chlortoluron), a significant effect was observed with the increase in the number of micro-closures and a decrease in the VOA. Finally, the growth of the oyster shell was slowed down in the presence of MPs alone with a significantly reduction of 50.4 % and for the cocktail condition with 42.7 % of growth reduction. This study provides evidence for the impact of MPs on valve behaviour and oyster shell growth, using high but realistic MPs concentrations. Moreover, additional analyses will be carried out on toxicity biomarkers, oxidative stress, metabolites and energy reserves potentially impacted by the MP exposure.

Keywords : experimental biomonitoring , growth , microplastic , Oyster , pesticide , valve behaviour

## Occurrence of microplastics in pacific oysters from aquaculture areas in Santa Catarina Island, Brazil: preliminary results

Brocardo Giulia, Saldaña-Serrano Miguel, Nogueira Diego, Vieira Khauê, Bainy Afonso.

Microplastics (MPs) are particles smaller than 5 mm that do not degrade rapidly and can be ingested by marine organisms and can also be transferred to higher trophic levels, including humans. Therefore, it is of great importance to monitor the occurrence of microplastics in marine bivalves. This study aimed to evaluate the presence, shape, size and chemical composition of microplastics in the oyster Crassostrea gigas in an aquaculture area in Santo Antônio de Lisboa, located in the Santa Catarina North Bay, Brazil. Fifteen oysters were harvested and only five were analyzed, their soft tissues were extracted, digested with KOH at 10% (1:3) for 72 hours, and then a density separation was performed with NaCl (1.2 g/cm3). Subsequently, the samples were filtered through a membrane (8 µm) and analyzed under the stereoscopic fluorescence microscope and the transmission electron microscope coupled to an energy dispersive spectrometer (SEM/EDS). Forty-two microparticles with sizes from 261.54 to 3066.6 µm were found, the majority being fibers (97% of the total) and the remaining fragments. Five different colors were found among the microparticles, blue being the color that was most present especially in the fibers. Fourteen microparticles were analyzed under the SEM/EDS, and all presented a high peak in the carbon molecule (C), suggesting it as a polymer. Consequently, the number of MPs ranged from 2 to 4 MPs/oyster. The chemical composition found in the microparticles was compared with other studies and four possible types of polymers were found: PVC, PET, PP and PS. Furthermore, with SEM/ EDS analyzes it was possible to visualize cracks and grooves on the surface of the particles, which are most likely due to environmental exposure. These results allowed us to show the presence of microplastics in oysters C. gigas from an aquaculture farm.

Keywords : Crassostrea gigas , Florianópolis , Marine Pollution , Ostreiculture , Polymer

#### **Biodiversity and structure of plastisphere in lentic ecosystems**

Di Pippo Francesca, Venezia Cristina, Sighicelli Maria, Pietrelli Loris, Di Vito Stefania, Nuglio Simone, Rossetti Simona.

During transport by water flow, microplastics (MPs) can be colonised by planktonic microorganisms able to adhere to plastic surfaces with subsequent formation of biofilms. Studies investigating marine MP-associated communities reported the existence of plastic-specific microbial communities, constituting a peculiar micro-ecosystem defined 'plastisphere'. Despite the well-documented widespread presence of MPs also in freshwater ecosystems, studies on plastisphere in these environments are still rare. This study investigated for the first time community composition and structure of microbial biofilms attached to MPs sampled from lentic ecosystems, by using 16S rRNA gene high throughput sequencing and Fluorescence In Situ Hybridization (FISH) combined with confocal laser scanning microscopy (CLSM). To address the lack of information on plastisphere microbiome composition, and to evaluate the eventual selection and recruitment of specific taxa from the planktonic community, this study analysed plastisphere and associated water communities in the main Italian lakes. Clear differences in microbial community composition among plastisphere and the corresponding planktonic populations were found. Freshwater biofilms shared a core microbiome constituted by known biofilm formers. Species composition of plastisphere did not substantially differ between the diverse polymers, while a clear link with the MP exposure time was found by Fourier Transform Infrared spectroscopy (FT-IR) and Scanning Electron Microscopy (SEM) analysis. It is here suggested that MPs may select, from water column, microorganisms able to adhere to these substrata (i.e. generalist taxa as members of Sphingomonadaceae and Rhodobacteraceae families) that likely act as pioneers, thus allowing the further attachment of other microbial colonizers (i.e. members of Hyphomonadaceae, Rhizobiaceae, Rhodocyclaceae, Xanthomonadaceae and of cyanobacterial families). Interestingly, several bacterial genera previously reported to be able of degrading PE were found in plastisphere samples. This finding deserves further attention and efforts to evaluate if these micro-consortia may be good candidates for MP degradation in freshwater environments.

Keywords : bacterial biodiversity , biofilm , plastisphere

### A global study reveals rare and unexplored bacteria that are specific to the plastic biofilm and reoccur across habitats

Scales Brittan S., Cable Rachel N., Duhaime Melissa B., Gerdts Gunnar, Kreikemeyer Bernd, Pedrotti Maria-Luiza, Gorsky Gaby, Oberbeckmann Sonja, Labrenz Matthias.

Plastic pollution is pervasive in marine systems in the Anthropocene era, and this pollution is known to carry complex biofilm-forming microbial communities. However, it is unknown whether these biofilm communities are specific to plastic or if they merely reflect those naturally occurring in the environment. To address this question, floating microplastics and non-plastic particles were sampled from the surface water of three marine ecosystems: the Baltic, Sargasso and Mediterranean Seas; for each location we characterized both free-living and particle-associated water community composition using 16S rRNA sequencing. We found that plastic and non-plastic biofilm communities were similar to each other across this large geographical range but found no measurable effect of the plastic polymer type. We identified plastic-specific OTUs that were not found either on non-plastic particles nor in the surrounding waters. Twenty-six of the plastic-specific OTUs were globally ubiquitous; this provides novel evidence that plastic repeatedly enriches the same bacteria from the surrounding water in diverse locations instead of simply mirroring natural microbiota. That the majority of the ubiquitous plastic-specific OTUs were assigned to the Rhodobacteraceae family, known to contain numerous hydrocarbonoclastic bacteria, and were also highly similar to bacteria previously sequenced in relation to oil-spills, suggests that properties of plastic, such as the absorption and leaching of hydrocarbons, could be an important part of the enrichment of bacteria to plastic biofilms. These plastic-specific bacteria that are repeatedly enriched from the surrounding waters, and not detectable on non-plastic particles, therefore have the potential to shift the aquatic microbial communities in which they are found. If plastic pollution continues to increase, selection for plastic-specific bacteria across the global oceans has potential to affect food web and ecosystem dynamics.

Keywords : Baltic Sea , biofilms , Mediterranean Sea , plastic biofilms , rare biosphere , Rhodobacteraceae , Sargasso Sea , the Anthropocene

### Session 26.2\_Me. Chaired by Eva Cardona, Menorca

**Microplastics budget for Norway's largest lake utilizing a multidisciplinary approach** *Clayer Francois, Buenaventura Nina, Jartun Morten, Guerrero Jose-Luis, Lusher Amy.* Paper number 334567

**Primary and secondary microplastics in rural and urban beaches in southern Brazil** *Schneider Ingrid, Fernandino Gerson, Maffessoni Daiana, Barbosa Eduardo.* Paper number 334596

## The Ocean Race Science: An innovative collaboration between science and extreme offshore sailing

*Gutekunst Sören, O'donovan Mairéad, Turner Anne-Cécile.* Paper number 334681

### The amount of plastic in soil from Mausund and Froan Nature reserve, Norway. – A Comparison with concentrations in laboratory exposure studies

*Cyvin Jakob Bonnevie, Ervik Hilde, Kveberg Anne Aasen.* Paper number 334819

# Microplastics budget for Norway's largest lake utilizing a multidisciplinary approach

Clayer Francois, Buenaventura Nina, Jartun Morten, Guerrero Jose-Luis, Lusher Amy.

Microplastic research initially focused on marine environments leaving freshwater ecosystems mainly unexplored, despite their hypothesized importance as suppliers of microplastics to the ocean. In addition, relevant approaches for unravelling microplastic sources and transport pathways at the catchment level are still missing to enable efficient actions towards mitigation and remediation. Here, we investigate microplastic stocks and fluxes in a Nordic lake ecosystem, Lake Miøsa. We utilised a multidisciplinary approach for identifying sources of microplastic pollution and draw a plastic budget for the catchment of L. Mjøsa. The microplastics spatial and vertical distribution in lake sediments is compared to socio-economic modelling of plastic fluxes as well as spatial datasets on land use and potential plastic sources in the catchment and sub-catchments. We quantified the different plastic morphologies within sediment cores from 20 sites in the lake and explain their spatial distribution with an innovative spatially comprehensive dataset. We estimate that 32.5 tonnes (5.8-117.1 t) of plastics are released into the lake annually of which 90% is transported downstream and only 10% is deposited in the sediment. Synthetic textiles and packaging are likely the main sources of sediment contamination in L. Miøsa probably through agricultural sludge application and urban waste mismanagement and littering. Most of the spatial variability of sediment microplastic is explained by the distance to the nearest town. The 10 most polluted sites, so called urban sites, contain polymers of higher diversity than natural sites and show increasing microplastic concentrations, diversity and richness over 1980-2018. This interdisciplinary approach to understand microplastics in Lake Mjøsa utilising catchment processes takes microplastic research further than simply presence, absence and abundance. By corroborating sediment inventories with top-down estimates of plastics emissions, modelled on the socio-economic activities in the catchment we have been able to provide a microplastic budget for the whole lake catchment.

Keywords : catchment , lake sediment , land use , microplastics , plastic sources

### Primary and secondary microplastics in rural and urban beaches in southern Brazil

Schneider Ingrid, Fernandino Gerson, Maffessoni Daiana, Barbosa Eduardo.

Primary and secondary microplastics (MPs) are found in virtually any marine and oceanic sedimentary environment, regardless of distance from urban areas. The objective of this study was to classify the degree of pollution of beaches using the Pellet Pollution Index (PPI). This methodology was adapted, and fragments were included. MPs were sampled in an urbanized (Capão da Canoa) and a rural (Cidreira-Cabras) beach, located in the northern coast of the state of Rio Grande do Sul. Sampling occurred during austral summer and winter 2018 in triplicates (50 x 50 cm squares) along the hightide strandline and the backshore, adding up 0.75 m<sup>2</sup> of superficial sediment sampled. During summer, more MPs were found in Cabras beach – strandline: 343 MPs [pellets np = 210 (61.2%); fragments nf = 133 (38.8%); PPI = 1.52 "moderate"]; backshore: 13 MPs [np = 4 (30.8%); nf = 9 (69.2%); PPI = 0.06 "very low"] – than in Capão da Canoa beach – strandline [ nf = 26 (100%); PPI = 0.12 "very low"]; backshore: 21 MPs [np = 6 (28.57%); nf = 15 (71.43%); PPI = 0.09 "very low"]. During winter, Cabras beach also presented the highest number of MPs - strandline:194 MPs [np = 173 (89.2%); nf = 21 (10.8%); PPI = 0.9 "low"]; backshore: 93 MPs [np =73 (78.5%); nf = 20 (21.5%); PPI = 0.41 "very low"]. Capão da Canoa - strandline: 29 MPs (np= 0 (0%); nf = 29 (100%); PPI = 0.13 "very low"]; backshore: 78 MPs [np = 4 (5.13%); nf = 74 (94.87%); PPI = 0.35 "very low"]. Although cities are important sources of marine litter, particles are carried along the coast and end up stranding on a beach far from its origin. Therefore, as observed in the present study, rural areas are not free from MP pollution.

Keywords : beaches , fragments , Microplastics , pellets

## The Ocean Race Science: An innovative collaboration between science and extreme offshore sailing

Gutekunst Sören, O'donovan Mairéad, Turner Anne-Cécile.

The Ocean Race is a professional round-the-world sailing race, which has shown its potential in previous editions as a platform for ocean and weather observations, particularly from remote areas outside of routine shipping routes. Deployables (drifters), microplastic sampling, oceanographic insitu (fCO2, salinity, sea surface temperature), and meteorological measurements (wind speed, wind direction, barometric pressure) were recorded as part of the 2017-18 edition of the race. Over five months during 2019 the VO65 race yacht of team AkzoNobel continued this contribution to scientific data collection during The Ocean Race - European Tour. A flow-through filter system installed onboard was used to sample for microplastic particles. The samples were collected using a flowthrough system that operated 24/7. Filters were changed at 24 h frequency. A total of 21 positions were collected and returned to a laboratory in Kiel for analysis using Raman. The route sampled started in Lisbon/PO and consisted of samples from the North Atlantic, North Sea, Baltic Sea and Mediterranean and finally finished in Alicante/SP. The Ocean Race fleet of competing yachts will be involved in data collection, with a science programme tailored for each class of boat - IMOCA 60 and VO65. Strong collaborations and partnerships are being developed to optimise sustained observations both during and outside of the race period with a particular value recognised for remote transects through rarely observed regions of the Southern Ocean and South Pacific. The next start of the Ocean Race will be in 2022 next to a possible European Race happening in 2021.

Keywords : communication , microplastic , race yacht , raman , real , sailing , sampling , time

### The amount of plastic in soil from Mausund and Froan Nature reserve, Norway. A comparison with concentrations in laboratory exposure studies

Cyvin Jakob Bonnevie, Ervik Hilde, Kveberg Anne Aasen.

On the Norwegian coast, on uninhabited islands of Mausund, there are by visual analysis found high concentrations of macro and microplastic. When turning the grassy soil close-to-shore upside down, there are multiple places found a layer of macro and micro plastic. Plastics is so far found to be quite harmless in environmental realistic concentrations. At the other side there are challenges in comparing different studies when different methodologies, size-classes and terminology are used; and in that manner of fact it is also difficult to conclude on witch concentrations there actually are in the environment. In laboratory studies is consumption of high concentrations of microplastic, found to affect the dietary function, trigging inflammations, change egg production and endocrine system of different species of low-trophic organisms. In this presentation, we are presenting data and methodology for upper soil-layer analysis of macro- and microplastic. We are presenting the proportion of plastic, compared to natural materials in soil, and we are comparing these results to the concentrations used in laboratory studies looking at ecotoxicological effects of microplastic exposure to biota.

Keywords : biota, ecotoxicological effects, macro and microplastic, plastic in soil

### Session 26.2\_Ma. Chaired by Ana Liria, Gran Canaria

### Concentration of marine litter on the beaches of the Galapagos Islands and main factors that influence its distribution

*Sánchez-García Natalia, Sánz-Lazaro Carlos.* Paper number 334510

#### Ecotoxicology of microplastics – knowledge gaps and possible steps forward

Wendt-Potthoff Katrin, Laforsch Christian, Hoess Sebastian, Fueser Hendrik, Rauchschwalbe Marie-Theres, Traunspurger Walter, Haegerbaeumer Arne, Hennig Ariane, Beggel Sebastian, Imhof Hannes, Geist Jürgen, Schulte-Oehlmann Ulrike. Paper number 334549

### A snapshot of global actions to reduce the flow of microplastic to the ocean from the UNEP stock-taking exercise 2020

*Pahl Sabine, Goodhew Julie, Bacosa Hernando, Creencia Lota, Thompson Richard.* Paper number 334862

### Assessment of multiple impacts of marine litter in the Adriatic Sea: from fishing for litter to fish species

Panti Cristina, Baini Matteo, Galli Matteo, Giani Dario, Limonta Giacomo, Concato Margherita, Pasanisi Eugenia, Galasso Gabriele, Delaney Eugenia, Pojana Giulio, Fossi Maria Cristina. Paper number 334920

## **Concentration of marine litter on the beaches of the Galapagos Islands and main factors that influence its distribution**

Sánchez-García Natalia, Sánz-Lazaro Carlos.

Plastic pollution is a growing environmental concern that affects all areas on earth even remote islands such as Galapagos Islands. In this study, macrodebris and meso- and microplastics were sampled in several beaches of the Galapagos Islands to estimate their concentration. Environmental parameters, such as orientation, degree of use and slope of each beach and distance from the population center were measured to study test if they played a relevant role in the accumulation of marine debris. For that purpose, protocols for the characterization, extraction and quantification of plastics in the study area were development and Raman spectroscopy was used to identify the type of polymer of the microplastics. Here, we present preliminary data of this study. Plastics constitute a 98.8% of the total number of macrodebris. The abundance of plastics in the sampled beaches in Galapagos Islands is  $9.87 \pm 2.15$  macroplastics/m2 and  $1489 \pm 183$  mesoplastics/m2, despite the differences among beaches is notable. Polyethylene (PE) is the most common type of microplastics. Marine currents and the accumulation of macroplastics may be the main factors affecting the distribution of plastics of a more reduced size through their degradation. Fibers, fragments and films are the most abundant types of plastics, especially those derived from fishing activities. Galapagos Islands could serve as an indicator of the plastic pollution from the surrounding continents and of the activities developed around them. However, it is necessary to carry out complementary studies to confirm this.

Keywords : Galapagos islands , marine environment , plastics , pollution , Raman spectroscopy , remote areas

#### Ecotoxicology of microplastics – knowledge gaps and possible steps forward

Wendt-Potthoff Katrin, Laforsch Christian, Hoess Sebastian, Fueser Hendrik, Rauchschwalbe Marie-Theres, Traunspurger Walter, Haegerbaeumer Arne, Hennig Ariane, Beggel Sebastian, Imhof Hannes, Geist Jürgen, Schulte-Oehlmann Ulrike.

The ubiquitous contamination of the environment with microplastics (MP), the associated risks to ecosystems and ultimately to human health has recently attracted great public and scientific attention. However, possible risks associated with MP cannot be generalized for several reasons: (1) MP is a collective term for a very heterogeneous class of particles with respect to their size (; 1 mm - 5 mm), polymer type, chemical and physical properties as well as surface characteristics. (2) Wellestablished tools and guidelines for the risk assessment of solutes cannot be transferred to MP as polymers are composed of various chemical compounds, and research on effects of sparingly soluble particulates in the environment is generally scarce. (3) In contrast to macroplastics (¿ 5 mm), there is almost no mechanistic understanding of observed effects vet. (4) Some fundamental deviations between field and laboratory studies (e.g., types and concentrations of MP, selection of exposed organisms, time scales) hamper the understanding of MP effects at the community or ecosystem level. (5) There is a bias in scientific literature towards studies that focus on adverse effects, but for a sound risk assessment, a more balanced view is essential. (6) The interpretation of observed effects of microplastics in the context of measured environmental concentrations and naturally occurring microparticles is often missing. We suggest possible advancements through a careful design of relevant exposure scenarios, use of appropriate reference materials, systematic research on mode of action, and establishing model systems. Furthermore, we emphasize the need for a detailed documentation and communication of experimental parameters for a more balanced and substantial valuation of both effect and non-effect studies.

Keywords : ecotoxicology , exposure , freshwater , microplastic

### A snapshot of global actions to reduce the flow of microplastic to the ocean from the UNEP stock-taking exercise 2020

Pahl Sabine, Goodhew Julie, Bacosa Hernando, Creencia Lota, Thompson Richard.

The ad hoc open-ended expert group (AHEG) on marine litter and microplastics was established through United Nations Environment Assembly (UNEA). One of its mandates is to take stock of currently existing actions to reduce marine litter and microplastics. A dedicated online survey tool was designed to conduct the stock-taking, and invitations were sent to all UN Member States, major groups and other stakeholders by the AHEG Chair. Here we report on the results of the stock-taking specifically for microplastic actions. Of the 220 usable submissions on actions we received, the majority were on macroplastics, with microplastics a smaller category, followed by additives. Analysis is still ongoing of the data received in August 2020, but we will report on the nature (e.g., technical, capacity-building, monitoring) and geographic spread of microplastic actions, and where in the lifecycle phase and environmental zone they are targeted. We will summarise the barriers and facilitators reported and review funding and partnerships supporting these actions as well as evaluation and reporting practices. Finally, we will discuss these insights in the context of actions mentioning biodegradable materials and discuss limitations and practical issues of the UNEP stock-taking exercise.

Keywords : evaluation , overview , policy , solutions , United Nations Environment Programme

## Assessment of multiple impacts of marine litter in the Adriatic Sea: from fishing for litter to fish species

Panti Cristina, Baini Matteo, Galli Matteo, Giani Dario, Limonta Giacomo, Concato Margherita, Pasanisi Eugenia, Galasso Gabriele, Delaney Eugenia, Pojana Giulio, Fossi Maria Cristina.

The AdriCleanFish project aimed to study the impact of marine litter in the fishing system and how this can affect fish species and fishing resources. The study was carried out through the removal of marine litter by fishing vessels, the analysis of collected litter, the analysis of floating marine litter and microplastic ingestion and effects on fish species of commercial interest for human consumption. The areas of interest were located in the Adriatic Sea (Chioggia and Civitanova Marche). The collected material from the sea bottom was characterized to assess weight, volume, number and composition. In addition, the presence of macro-, meso- and microplastics in fish species (European anchovy, hake, European pilchard, sole, horse mackerel and red mullet) was determined. The results show an average concentration of floating microplastics and marine litter on the surface of study area in line with the average values of the Mediterranean. Form marine litter collected by the "fishing for litter" activities from the bottom, the data confirm that plastic materials are the most frequently found (more than 70%) and the most abundant in term of weight. The plastic objects analyzed are mainly disposable and packaging items (eg. bags and bottles), having a land-based origin and they are also related to maritime activities, to fishing and aquaculture. In all the fish species analyzed, the presence of plastic in the gastro-intestinal tracts was detected. On average, 2 fish out of 10 had ingested from one to five microplastics. The pelagic species (anchovy and pilchard) have the highest percentage of occurrence of ingested microplastics. PCBs and PBDEs were also measured in the muscle of fish species as well as biomarker responses. The results show negligible effects related to plastic ingestion by fish as well as low levels of contaminant accumulation in fish with microplastics in their gastro-intestinal tract.

Keywords : effects , fish species , fishing for litter , microplastics , seafloor litter

### Session 26.3\_O. Chaired by Martin Wagner, Trondheim

### Toxicity of microplastics and nanoplastics for aquatic organisms along the freshwater-marine water continuum

Latchere Oihana, Metais Isabelle, Baudrimont Magalie, Feurtet-Mazel Agnès, Perrein-Ettajani Hanane, Mouloud Mohammed, Gonzalez Patrice, Gigault Julien, Châtel Amélie. Paper number 334375

### Assessment of polystyrene microparticles effects by a set of integrated biomarkers in the marine bivalve species *Scrobicularia plana*

Gonçalves Ana M. M., Knobelspieß Sara, Mesquita Andreia Filipa, Sahadevan Seena, Gonçalves Fernando J. M., Marques João C.. Paper number 334451

**Immunotoxicity screening to understand the impacts of microplastics on human health** Beijer Nick, Carlier Maxim, Wolter Helen, Mengelers Marcel, Dehaut Alexandre, Duflos Guillaume, Niemann Helge, Amaral-Zettler Linda, Staal Yvonne. Paper number 334513

### Is immune response of freshwater insects triggered by microplastics ingestion? A case study using the aquatic midge *Chironomus riparius*

Silva Carlos, Beleza Fernandes Sónia, Campos Diana, Soares Amadeu, Silva Ana Luisa, Pestana João, Gravato Carlos.

Paper number 334517

### Toxicity of microplastics and nanoplastics for aquatic organisms along the freshwater-marine water continuum

Latchere Oihana, Metais Isabelle, Baudrimont Magalie, Feurtet-Mazel Agnès, Perrein-Ettajani Hanane, Mouloud Mohammed, Gonzalez Patrice, Gigault Julien, Châtel Amélie.

Pollution by plastic particles is a major environmental concern in aquatic environments. Numerous studies have been carried out to assess the toxicity of microplastics (MPs), and more recently nanoplastics (NPs), for aquatic organisms. However, some aspects have so far been little studied. The majority of studies focus on the effects of plastic particles in the marine environment while estuarine and freshwater environments are little studied despite their important role in the transfer of plastics to the environment. In addition, the potential transfer of MPs and NPs into aquatic trophic chains, for example, is currently poorly understood. This led us to develop a multidisciplinary approach to better understand the effects of MPs and NPs, representative of those found in an estuary, on key species from a food chain living in the freshwater-marine water continuum. The food chain includes phytoplanktonic algae Scenedesmus subspicatus and diatoms Thalassiosira weissflogii (first trophic levels), endobenthic bivalve molluscs Corbicula fluminea and Scrobicularia plana (second trophic levels) and the European eel Anguilla anguilla (third trophic level). The impacts of MPs and NPs on aquatic species are assessed by both direct route and trophic route and are characterized using a multi-marker approach ranging from effects at the individual level (physiological, behavioral) to the sub-individual level (molecular, biochemical, microscopic). This talk will focus on the preliminary results of the direct exposures of the mollusc bivalves to plastic particles.

Keywords : environmental representativeness , estuaries , microplastics , nanoplastics , trophic transfer

### Assessment of polystyrene microparticles effects by a set of integrated biomarkers in the marine bivalve species *Scrobicularia plana*

Gonçalves Ana M. M., Knobelspieß Sara, Mesquita Andreia Filipa, Sahadevan Seena, Gonçalves Fernando J. M., Marques João C..

Microplastics are emerging contaminants in marine environments and represent a main concern to the scientific community. Estuarine ecosystems are hotspots of microplastic pollution, with possible impacts to its communities. Microplastic ingestion has been reported for several marine species, however the toxicity to aquatic biota and the effects on the ecosystems are still poorly studied. Scrobicularia plana has a large pollutants filtration ability and plays a key role in structure and functioning of estuarine communities, therefore it was selected for this work. In this study two size classes of S. plana organisms were exposed to environmentally relevant polystyrene microparticles  $(1 \,\mu\text{m})$  concentrations (0.00  $\mu\text{g/L}$  to 25  $\mu\text{g/L}$ ) by 96h. At the end, the mortality and condition index were determined and performed biochemical analysis to assess the effects on antioxidant defence system. Based on the results, no direct lethal effects or index condition changes were observed, as consequence of the polystyrene exposure. Still it was observed alterations on the antioxidant defence mechanisms and consequently an increase of oxidative damage. Big size class showed dosedependent responses, with activities induction of total glutathione peroxidase and glutathione reductase, superoxide dismutase activity was inhibited with the exposure, with this inhibition decreasing with the concentration increase, resulted in significantly increase of lipid peroxidation (quantified as TBARS levels), whereas catalase and glutathione-S-transferase activities were not altered. Organisms of the small size class did not show clear alterations of antioxidant biomarkers, with the exception to superoxide dismutase activity that showed the same trend of big size class. This study highlights the adverse effects of polystyrene microparticles on the estuarine clams and suggests S. plana adult organisms as good bioindicator for the presence of polystyrene pollution in aquatic environments, being tGPx, GRed, SOD and TBARS appropriate biomarkers to assess polystyrene microplastic toxicity, with emphasis to GRed and SOD.

Keywords : Antioxidant Defence Mechanisms , Bivalve species , Estuarine ecosystems , Microplastics

## Immunotoxicity screening to understand the impacts of microplastics on human health

Beijer Nick, Carlier Maxim, Wolter Helen, Mengelers Marcel, Dehaut Alexandre, Duflos Guillaume, Niemann Helge, Amaral-Zettler Linda, Staal Yvonne.

There is growing awareness of the quantity of minute plastic particles found in the food we consume, raising concerns on potential adverse effects. Macrophages play an important role in scavenging of particles, which may lead to induction of an immune response. To investigate the role of macrophages, we assessed the effects on macrophages of microplastics of different size and origin. Environmentally-weathered macroplastic samples were collected from the open-ocean South Atlantic Subtropical Gyre (Garbage Patch) and from the French coastal environment. Plastics were cryomilled to obtain sizes below 300 µm. Coastal plastic samples were identified as Poly-ethylene (PE). polypropylene (PP), polystyrene (PS) and polyethylene terephthalate (PET) and subsequently sizefractionated to: 20-50 µm, 50-100 µm, and 100-200 µm size classes. We used (micro)Ramanspectrometry, FT-IR and Py-GC/MS, and phase-contrast microscopy to further characterize their physico-chemical properties. Differentiated macrophages (THP-1 cells) were grown in 96-well plates, and exposed to sinking particles directly or were placed on the basolateral side of inserts, to expose them to floating particles. After 48 hours of exposure, we assessed cell viability and cytokine response. Immunotoxicity response showed distinct patterns for each size class, but smaller particles were also detected in larger size fractions, making it difficult to interpret results. Macrophages also responded differentially to different plastic resin compositions. PET exposure produced the largest changes, including a dose-related response in cytokine production (IL-1 $\beta$ , IL-6, and TNF- $\alpha$ ). Smaller particles (indicated size range) induced cytokine production at lower concentrations. We intend to use a combination of physiochemical and biological data for in-depth multi-dimensional data analysis to further explore the potency of particles properties in relation to their potential immunotoxicological effects. This research contributes to our understanding of the potential hazards of environmentally-sourced plastics on human health.

Keywords : environmental microplastics , human health , Immunetoxicity , macrophages

## Is immune response of freshwater insects triggered by microplastics ingestion? A case study using the aquatic midge *Chironomus riparius*

Silva Carlos, Beleza Fernandes Sónia, Campos Diana, Soares Amadeu, Silva Ana Luisa, Pestana João, Gravato Carlos.

The activation of insects' immune system due to ingestion of microplastics (MPs) has been evidenced by the upregulation of specific genes. Yet, the pathway through which MP elicit such responses is poorly understood. The activation of phenoloxidase (PO) is one of the main responses involved in the innate immunity of insects when facing parasites and pathogens, and similar process may occur in the presence of MPs. Therefore, this study aims to address the activity of PO and total PO (PO+proPO) activities in larvae of Chironomus riparius, allowing to estimate if de novo synthesis of prophenoloxidase (proPO) occurs. Larvae were exposed 48 h to irregularly-shaped (size range 32-63 µm) polyethylene microplastics (PE-MPs) present in sediments (1.25; 5; to 20 g kg-1). The ingestion of PE-MPs by larvae triggered a significant increase of PO activity (+25 and +28% in larvae exposed to 5 and to 20 g kg-1, respectively), whereas total PO activity increased +48% in larvae exposed to 20 g kg-1. Results also showed an increased activity of PO showing that particles can induce the innate immune system of larvae. Moreover, the induction of total PO activity on larvae exposed to 20 g kg-1 PE-MPs suggests de novo synthesis of the inactive form of the enzyme (proPO). Since the size of the PE-MPs ingested by larvae are too large to cross biological barriers, the activation of the immune response is probably linked to an inflammatory response due to their accumulation and retention inside the gut. This research work provides the first evidence on the direct activation of innate immune system of an insect after ingestion of MPs. Thanks are due to FCT/MCTES for the financial support to CESAM (UIDP/50017/2020+UIDB/50017/2020, through national funds) and the research project ComPET (POCI-01-0145-FEDER-030361, funded by FEDER, through COMPETE2020-POCI and by national funds (OE)).

Keywords : biomarkers , innate immune system , invertebrates , phenoloxidase , plastic pollution

### Session 26.3\_Me. Chaired by Bruno Tassin, Marne la Vallée

**A review on the impacts of microplastics on freshwater wild biota** *Alessandra Cera, Scalici Massimiliano.* Paper number 334194

**Goldilocks and the three surface water sampling methods** *Markley Laura, Driscoll Charles, Costello Staniec Andria, Huth Ellenora.* Paper number 334562

### Improvement of the microplastics separation methodology in freshwater systems based on reused zinc chloride

Rodrigues Mariana Oliveira, Gonçalves Ana Marta Mendes, Gonçalves Fernando José Mendes, Abrantes Nelson. Paper number 334577

#### A review on the impacts of microplastics on freshwater wild biota

Alessandra Cera, Scalici Massimiliano.

Microplastics pollution exerts pressure on freshwater ecosystems globally, due to their abundance and widespread distribution. The impacts on biota are usually assessed by indoor experiments, however, a link between laboratory and natural observations is critical for better risk assessment and for prioritising research aims. We provide a review of the scientific literature on observations conducted on wild biota exposed to microplastics in nature since 2012. Fish and macroinvertebrates are the main taxa investigated, but amphibians, birds, and microorganisms are also observed. Especially for fish, the evaluation of microplastic ingestion is a common research topic. The results show contamination of the gastrointestinal tract of fish, but promising results are also emerging from liver investigations. The factors that cause fish to ingest microplastics are different, however, microplastics bioavailability and the act of feeding are thought to increase the ingestion. The environmental contamination of microplastics is linked to its proximity to sources, such as areas with high population density. Indeed, research on microplastics colonisation by microorganisms revealed a close relationship with urban areas, specifically to wastewater treatment plants effluents. Monitoring the occurrence and quantity of microplastics is increasingly required and fish and invertebrate species are emerging as suitable tools. In this regard, we support the publication of standardised protocols for the assessment of the concentration of microplastics in biota and for the identification of suitable indicator species for biological monitoring. Future research is suggested to include under-studied taxa, such as mammals, reptiles, amphibians and assessing the vulnerability of chicks to adult birds. We contributed with a collection of natural observation on the impacts of microplastics on wild biota for providing insight to researchers working in the laboratory experiments and in nature. This interdisciplinary approach wants to support the overall knowledge on the phenomena of microplastics pollution for a better assessment of environmental safety.

Keywords : bivalve , freshwater ecosystem , ingestion , microplastic , monitoring , plastisphere

#### **Goldilocks and the three surface water sampling methods**

Markley Laura, Driscoll Charles, Costello Staniec Andria, Huth Ellenora.

While one method's sampling volume is too low, others present challenges with contamination and sample processing. Which is just right? Sampling microplastics in fresh surface waters can be difficult given the variable accessibility and quality of bodies of water. Choosing the right sampling method is integral to collecting accurate data, while also keeping costs and potential challenges to a minimum. We employed three distinct surface water sampling methods on freshwater lakes and streams in central New York: trawling with a 300 micron microplastic sampling net, bulk sampling with 1L mason jars, and bucket-to-bucket sampling through variable sieve sizes. This study reflects on the applicability of these three sampling methods and potential variations between them based on our ongoing work and those reported by other studies. While bulk sampling was the easiest method, it also obtained the least quantifiable microplastics using visual ID. By contrast, trawling was more time-intensive, contamination prone, and presented the most challenges for organics processing because of the high inputs of leafy debris. However, trawling sampled the highest volume and had the most visually identifiable microplastics. Depending on sample location, bucket-to-bucket sampling had characteristics of both bulk and trawl sampling but was the most flexible as far as cost and accessibility. This work further reflects on the context in which each method is useful and how processing and counting methodology could impact sampling method decision making.

Keywords : freshwater , methods , sampling , surface water

## Improvement of the microplastics separation methodology in freshwater systems based on reused zinc chloride

Rodrigues Mariana Oliveira, Gonçalves Ana Marta Mendes, Gonçalves Fernando José Mendes, Abrantes Nelson.

Microplastics (plastic particles with ; 5 mm; MPs) are considered emerging aquatic pollutants since they can derive from a variety of sources (primary and/or secondary) reaching high densities, they are ubiquitous and persistent and they can interact negatively with wildlife and human beings, depending on their shape and polymer type/specific density. These properties have led to an increasing societal and scientific concern. Nevertheless, the scientific community has not yet stablished a standard method/protocol for separating MPs from aquatic systems resulting in data that cannot be compared. Density separation is one of the most adopted methodologies using high concentrated or saturated salt solutions such as sodium chloride, zinc chloride (ZnCl2) or sodium iodide (NaI) to increase the water density and to make MPs float. Currently, ZnCl2 is reported as the most cost-efficiency method (density of 1.6 - 1.8 g/cm3) for separating MPs of low and high density, however it also has disadvantages such as being very hazardous, corrosive and requiring large amounts of product. Moreover, there is an immense lack of information relative to this substance when compared to others (e.g. NaI). Hence, the present study aimed to overcome these disadvantages by presenting an improved methodology based on the reuse of the ZnCl2 solution while keeping its efficiency. For that, artificial samples were prepared and subjected to the ZnCl2 methodology up to 5 times. These samples included MPs of polyethylene terephthalate and polyvinyl chloride, two of the densest polymers found in freshwater systems. Vacuum filtration and visual inspection, using a stereomicroscope, followed the density separation. This study showed that the ZnCl2 solution can be reused at least five times maintaining an efficiency above 95%.

Keywords : Aquatic systems , Density separation methodology , Microplastics , Monitoring , Zinc chloride

### Session 26.3\_Ma. Chaired by Matthew Cole and Chris Walkinshaw, Plymouth

**Microplastics influence sediment microbial communities and biogeochemical cycling** *Seeley Meredith, Song Bongkeun, Hale Robert.* Paper number 334140

Ingestion and retention of plastic fibres of different lengths by the green shore crab (*Carcinus maenas*) *Carreras-Colom Ester, Carrassón Maite, Welden Natalie A.*. Paper number 334284

### Taking a step back: Using lockdown to review what we know about interactions between benthic fauna and microplastics in the environment

Porter Adam, Lewis Ceri, Galloway Tamara. Paper number 334340

#### **Evaluation of the potential interaction between microplastic distribution and biodiversity richness along the coast of the Western Ionian Sea**

Galli Matteo, Baini Matteo, Panti Cristina, Rosso Massimiliano, Tepsich Paola, Fossi Maria Cristina.

Paper number 334557

# Microplastics influence sediment microbial communities and biogeochemical cycling

Seeley Meredith, Song Bongkeun, Hale Robert.

Microplastics are increasingly found in freshwater, estuarine and marine sediments. The bacterial communities within these sediments regulate key biogeochemical cycles, such as nitrogen and carbon. Previous studies have reported that polymer type can influence the composition of microbial communities thriving on floating plastics. Here, we show that microplastics of diverse polymer types differentially influenced sediment bacterial communities, as well as the biogeochemical cycles they mediate. With the two-fold goal to address microbial community structure and nitrogen cycling, we established a microcosm experiment with microplastics (53-300 mm) of four different polymer types: polyethylene (PE), polyvinyl chloride (PVC), polyurethane foam (PUF) and polylactic acid (PLA; a bio-polymer). After 7 and 16 days of incubation, sediment aliquots were sampled for 16S rRNA gene sequencing with Illumina MiSeq platform and qPCR of the genes responsible for nitrification (amoA) and denitrification (nirS and nirK). Nitrogen cycling processes were examined by measuring dissolved inorganic nitrogen concentrations in the overlying water and potential denitrification rates using a sediment slurry experiment with 15NO3- tracer. We observed that bacterial community compositions differed significantly between treatments with the most divergent in PVC-amended sediment. Bacterial amoA gene abundances and dissolved inorganic nitrogen fluxes revealed that nitrification was lowest in PVC treatments, and highest in PLA and PUF. Denitrification rates were correspondingly suppressed in PVC, but enhanced in PLA and PUF compared to the non-amended control. This suggests that both PLA and PUF may be serving as a carbon source to support denitrifiers in sediments. These results illustrate that microplastic interactions with bacteria extends beyond just the biofilm on the surface of the plastic, and can influence adjacent sediment bacterial communities mediating the critical biogeochemical cvcles.

Keywords : bacteria , microbiome , microplastic , nitrogen , sediment

## Ingestion and retention of plastic fibres of different lengths by the green shore crab (*Carcinus maenas*)

Carreras-Colom Ester, Carrassón Maite, Welden Natalie A.

Plastic ingestion has been reported in wide range of terrestrial and marine organisms and it is considered a major threat due to its potential accumulation, which has been associated to false satiation effects or the release of potentially toxic compounds (e.g. plastic additives). However, data on the retention and potential accumulation/egestion of plastics after a prolonged exposure is scarce. In this study, we analysed the ingestion and retention time of nylon threads of varied lengths in the green shore crab Carcinus maenas as well as the potential effect of a prolonged exposure to nylon fibres through diet. Adult male individuals (n=16) were divided into groups and exposed to four different diets: control diet, diet with short fibres (0.5 mm), diet with long fibres (2-4 mm) and diet with a mix of short and long fibres (0.5, 2 and 4 mm). Crabs were fed 3 times a week (0.5 g of blendered mussels and 0.15% of added fibres) for a period of 21 days. Faecal pellet production was analysed throughout the study (weight was recorded and images were taken in order to analyse their dimensions) and the haemolymph protein content (Bradford assay) and body condition indices (condition factor and hepatosomatic and gonadosomatic index) were recorded and compared at the end of the study. Nylon fibres of all lengths were encapsulated and successfully egested through faecal pellets. Faecal pellet production did not seem disrupted by fibre ingestion, yet those with longer fibres were more easily fragmented. As other authors have already suggested, the gastric mill might play a key role on the accumulation of fibres, especially longer ones, as they took the longest to be egested and presented deformations (e.g. bended, molten-like defects) after egestion. No clear significant differences were observed on the total protein content or condition indices between diets.

Keywords : Carcinus maenas , crustacean , experimental exposure , ingestion , Nylon fibre , retention

# Taking a step back: Using lockdown to review what we know about interactions between benthic fauna and microplastics in the environment

Porter Adam, Lewis Ceri, Galloway Tamara.

The pollution of the global environment by plastics is one of the great societal challenges of the 21st century. Almost all plastic pollution in the environment is predicted to sink to the benthic realm and the current levels are predicted to increase 50 fold by 2100. Benthic samples have historically been under represented in the body of literature to date due, to methodological constraints and the relatively difficulty in processing samples however as more research is gathered a general picture of widespread contamination is emerging. With so much still unknown in plastic pollution research, we believe it prudent to develop tools to identify sensitive species and ecosystems to elucidate the theoretical risk posed by plastic pollution. We believe that a fundamental understanding of organismal traits could help us to create a likelihood of uptake framework within which key species for investigation can be identified; guiding further research and enabling us to couple laboratory exposure experiments with field collected organisms in a more cohesive manner by investigating "at risk" species. We therefore present the early results collected from undertaking a meta-analysis of benthic microplastic ingestion and discuss pathways towards a risk assessment for benthic species. The aim of this study was to determine to what extent species traits (or other factors) confer vulnerability to microplastic ingestion, focusing on marine benthic species. We undertook an investigation of 1471 primary research articles returning 74 articles for final analysis after using a stringent inclusion and exclusion methodology. In doing so we identify 285 benthic associated species and over 17000 individuals who have been recorded to ingest microplastics and we present some of the early data highlighting trends found. Early results will be presented and the results discussed in relation to the risks microplastics pose to benthic ecosystems and the organisms that inhabit them.

Keywords : benthic , risk , traits , uptake

## **Evaluation of the potential interaction between microplastic distribution and biodiversity richness along the coast of the Western Ionian Sea**

Galli Matteo, Baini Matteo, Panti Cristina, Rosso Massimiliano, Tepsich Paola, Fossi Maria Cristina.

The presence of microplastic particles (MPs) in the Mediterranean Sea has been widely reported, but their ecological risk in the marine ecosystem have not been completely addressed. With this purpose, the MPs distribution in the coastline waters of the Western Ionian Sea were investigated for the first time. Considered a hot spot of biodiversity and important resting and nesting sites for birds, this area is heavily interested by anthropogenic pressure. Simultaneous visual census of several marine species and manta trawl sampling were performed during a cruise carried out in 2017, to provide an assessment of biodiversity richness and MPs presence. Species richness, Shannon index, and evenness were computed to determine the biodiversity of the studied area. MPs collected were isolated and characterized by shape, size, color, and chemical composition. All data from field surveys were analyzed in Quantum GIS platform. Kernel density estimation was overlapped with marine litter densities to obtain a preliminary risk assessment. In total, 380 sightings were recorded for a total of 17 species: five species of Odontocetes and twelve species belonged to different taxa such as birds and marine turtles. The highest values of biodiversity in terms of species richness and Shannon index were observed in the Gulf of Noto and the Gulf of Augusta, located in the southern part of the studied area. Taking into account the MPs distribution, the highest concentrations of MPs were found in the waters facing Syracuse and the Gulf of Augusta, with an average abundance of  $0.10 \pm 0.06$  items/m<sup>2</sup>. The overlap between the hotspots of biodiversity and MPs occurrence represents a preliminary contribution to the risk assessment of marine organisms' exposure to microplastics and may contribute to the identification of sensitive areas where mitigation actions to tackling the plastic pollution are needed.

Keywords : Biodiversity richness , Microplastic , Plastic distribution , Risk assessment

### The Editor's Perspective. Chaired by Andy Booth, Trondheim

Have you ever wondered what life is like as an editor of a scientific journal that publishes manuscripts related to plastic pollution? Want to know what it takes to publish in these journals what the editors are looking for?

Well, attend this session and meet Yong Sik Ok and Pavani Dissanayake from **Environmental Pollution,** Francois Galgani from **Marine Pollution Bulletin** and Bart Koelmans from **the new journal Microplastics and Nanoplastics**.

We encourage MICRO2020 participants to join us and contribute to a lively panel debate hosted by **Andy Booth** on 'plastic publishing'!

Environmental Pollution **Yong Sik Ok and Pavani Dissanayake**: The journal publishes high quality research papers and review articles about all aspects of environmental pollution and its effects on ecosystems and human health. It welcomes high-quality process-oriented and hypothesis-based submissions that report results from original and novel research and contribute new knowledge to help address problems related to environmental pollution at a regional or global scale.

Marine Pollution Bulletin **Francois Galgani**: The journal is concerned with the rational use of maritime and marine resources in estuaries, the seas and oceans, as well as with documenting marine pollution and introducing new forms of measurement and analysis. A wide range of topics are discussed as news, comment, reviews and research reports, not only on effluent disposal and pollution control, but also on the management, economic aspects and protection of the marine environment in general.

Microplastics and Nanoplastics **Bart Koelmans**: Microplastics and Nanoplastics is an inclusive, **interdisciplinary forum for worldwide efforts to solve the plastics pollution challenge**. The journal aims to publish innovative high quality science providing a quantitative and mechanistic understanding of the factors that drive the emissions, fate, effects, risks and societal responses to the presence of plastic debris in nature and society, as well as novel remediation and risk mitigation options and technologies.

We are pleased to welcome the new Springer journal 'Microplastics and Nanoplastics' into the world. To mark this occasion, we are very happy to announce that the journal is offering 4 full fee waivers for articles published in the new journal. In addition, three special posters will be chosen to receive a voucher for a Springer book. The selection will be made by the MICRO2020 scientific committee together with the editors of the new journal. Selection criteria will be scientific quality and quality of presentation. Finally there will also be a topical collection at the new journal.
### Session 26.4\_O. Chaired by Richard Thompson and Florence Parker-Jurd, Plymouth

Lost at Sea – where are all the tyre particles?

*Thompson Richard, Napper Imogen, Parker-Jurd Florence.* Paper number 334706

### The Travelling Particles: Investigating microplastics as possible transport vectors for multidrug-resistant *E. coli* in the Weser Estuary (Germany)

Song Jessica, Jongmans Elanor, Mauder Norman, Imirzalioglu Can, Wichels Antje, Gerdts Gunnar. Paper number 333929

### Drinking plastics? – Quantification and qualification of microplastics in drinking water distribution systems by µFTIR and Py-GCMS

Kirstein Inga, Hensel Fides, Gomiero Alessio, Vianello Alvise, Wittgren Hans Bertil, Iordachescu Lucian, Vollertsen Jes. Paper number 334462

### Lifetime accumulation of microplastic in children and adults

Nor Nur Hazimah Mohamed, Koelmans Albert, Kooi Merel, Diepens Noël. Paper number 334521

### Lost at Sea – where are all the tyre particles?

Thompson Richard, Napper Imogen, Parker-Jurd Florence.

Billions of road miles are driven every year, generating countless particles of synthetic rubber and reportedly one of the largest sources of microplastics to the environment. However, such estimates are in disagreement with environmental data where polyethylene, polypropylene, polystyrene and PET are the main types of microplastics and on a global scale only around 1% of studies report finding tyre particles; posing the question where are all the tyre particles? Here we summarise recent empirical evidence demonstrating the transfer of tyre particles to the environment by storm water, water, effluent form wastewater treatment and deposition from the atmosphere. Tyres contain a range of potentially hazardous chemicals, if their accumulation in the environment has been underestimated how concerning might this be, what mitigation measures might be appropriate and what lessons can be learned from previous work on other sources of microplastic contamination.

Keywords : tyre tire microplastics detection pathway

# The Travelling Particles: Investigating microplastics as possible transport vectors for multidrug-resistant *E. coli* in the Weser Estuary (Germany)

Song Jessica, Jongmans Elanor, Mauder Norman, Imirzalioglu Can, Wichels Antje, Gerdts Gunnar.

The prevalence of multidrug-resistant Gram-negative bacteria in aquatic environments has been a long withstanding health concern, namely extended-spectrumbeta-lactamase (ESBL) producing Escherichia coli, Given increasing reports on microplastic (MP) pollution in these environments, it has become crucial to better understand the role of MP particles as transport vectors for such multidrug-resistant bacteria. In this study, an incubation experiment was designed where particles of both synthetic and natural material (HDPE, tyre wear, and wood) were sequentially incubated atmultiple sites along a salinity gradient from the Lower Weser estuary (Germany) to the offshore island Helgoland (German Bight, North Sea). Following each incubation period, particle biofilms andwater samples were assessed for ESBL-producing E. coli, first by the enrichment and detection of E. coli using Fluorocult® LMX Broth followed by cultivation on CHROMAgar<sup>™</sup> ESBL media to select for ESBLproducers. Results showed that general E. coli populations were present on the surfaces of wood particles across all sites but nonewere found to produce ESBLs. Additionally, neither HDPE nor tyrewear particles were found to harbour any E. coli. Conversely, ESBLproducing E. coli were present in surrounding waters from all sites, 64% of which conferred resistances against up to 3 other antibiotic groups, additional to the beta-lactam resistances intrinsic to ESBL-producers. This study provides a first look into the potential of MP to harbour and transport multidrug-resistant E. coli across different environments and the approach serves as an important precursor to further studies on other potentially harmful MP-colonizing species.

Keywords : Antibiotic resistance , aquatic environments , Escherischia coli , extended , lactamases , spectrum beta , synthetic polymers

# Drinking plastics? – Quantification and qualification of microplastics in drinking water distribution systems by µFTIR and Py-GCMS

Kirstein Inga, Hensel Fides, Gomiero Alessio, Vianello Alvise, Wittgren Hans Bertil, Iordachescu Lucian, Vollertsen Jes.

While it seems indisputable that drinking water contains microplastics (MP), the actual concentrations are much debated and reported numbers vary many orders of magnitude. It is difficult to pinpoint the cause of these differences, but it might be variation between source waters, variation between quantification methods, and variation in analytical standards. Despite the urgent need to understand human exposure to MP mediated by drinking water, there is a lack of trustable methods generating reliable data. Essentially, proper MP assessment requires that quality assurance is in place and demonstrated, that an adequate volume of drinking water is assessed, and that differences in analytical methods are understood. This study presents a systematic and robust approach where MP down to 6.6 µm were assessed in potable water distribution systems in terms of quantity, size, shape, and material. For the first time, sub-samples of drinking water were analysed using two of the most validated and complementary analytical techniques: µFTIR imaging and Py-GCMS. Both methods successfully determined low contents in drinking water. However, µFTIR and Py-GCMS identified different polymer types in samples with overall low MP content. With increasing concentration of a given polymer type, the values determined by the techniques became more comparable. Most detected MPs were smaller than 150 µm, and 32 % were smaller than 20 µm. Our results indicate a potential annual uptake of less than one MP per person, suggesting that drinking potable water produced at a high-performance drinking water treatment plant represents a low risk for human health.

Keywords : drinking water treatment plants , human health , microplastic mass quantification , small microplastic

### Lifetime accumulation of microplastic in children and adults

Nor Nur Hazimah Mohamed, Koelmans Albert, Kooi Merel, Diepens Noël.

A probabilistic lifetime human exposure assessment for microplastics is essential for quantifying the magnitude, diversity and uncertainty of microplastic exposure. We review 134 studies on microplastic concentrations in nine intake sources, constituting 20 % of the human diet by mass. We simulate microplastic exposure in children and adults using probabilistic models for the gut passage that account uptake via dietary sources and inhalation, intestinal absorption and biliary excretion. To evaluate the potential chemical risk, we perform physiologically based pharmacokinetic modelling with and without chemical intake via microplastics. Microplastic median intake rates range from 553 to 883 #/capita/day and can accumulate up to 50100 #/capita ( 0.041 µg/capita) in the body at the end of 70 years. The contribution of microplastics to total chemical intake is negligible. In conclusion, there is a large variation in exposure and accumulation, and these estimates are subject to increase due to other unknown sources.

Keywords : bioaccumulation , chemical , probabilistic assessment

### Session 26.4\_Me. Chaired by Thierry Huck, Plouzané

Riverine plastic pollution from fisheries: insights from the Ganges River system

Nelms Sarah, Duncan Emily, Patel Surshti, Badola Ruchi, Bhola Sun, Chakma Surfarsha, Wahidunnessa Chowdhury Gawsia, Godley Brendan, Hague Alifa, Johnson Jeyaraj Antony, Khatoon Hina, Kumar Sumit, Napper Imogen, Niloy Nazmul Hasan, Akter Tanjila, Badola Srishti, Dev Aditi, Rawat Sunita, Santillo David, Sharma Ekta, Koldewey Heather. Paper number 332331

### A review of microplastics categorization schemes to facilitate source apportionment: Implications for North American freshwaters

*Yu Jasmine T., Helm Paul A., Diamond Miriam L..* Paper number 333588

### **Hyporheic exchange processes increase retention of microplastics in rivers** *Drummond Jennifer D., Nel Holly A., Packman Aaron I., Krause Stefan.* Paper number 334215

Seasonal variation in relative abundance and composition of microplastic particles and

### zooplankton in a pelagic environment (NE Atlantic)

Sambolino Annalisa, Herrera Inma, Álvarez Soledad, Alves Filipe, Canning-Clode João, Dinis Ana, Kaufmann Manfred, Cordeiro Nereida. Paper number 334482

### **Riverine plastic pollution from fisheries: insights from the Ganges River system**

Nelms Sarah, Duncan Emily, Patel Surshti, Badola Ruchi, Bhola Sun, Chakma Surfarsha, Wahidunnessa Chowdhury Gawsia, Godley Brendan, Hague Alifa, Johnson Jeyaraj Antony, Khatoon Hina, Kumar Sumit, Napper Imogen, Niloy Nazmul Hasan, Akter Tanjila, Badola Srishti, Dev Aditi, Rawat Sunita, Santillo David, Sharma Ekta, Koldewey Heather.

Abandoned, lost or otherwise discarded fishing gear represents a substantial proportion of global marine plastic pollution and can cause significant environmental and socio-economic impacts. Yet little is known about its presence in, and implications for, freshwater ecosystems, or its downstream contribution to plastic pollution in the ocean. This study documents fishing gear related debris in one of the world's largest plastic pollution contributing river catchments, the Ganges. Riverbank surveys conducted along the length of the river, from the coast in Bangladesh to the Himalaya in India, show that derelict fishing gear density increases with proximity to the sea. Fishing nets were the main gear type by volume and all samples examined for polymer type were plastic. Illegal gear types and restricted net mesh sizes were also recorded. Socio-economic surveys of fisher communities explored the behavioural drivers of plastic waste input from one of the world's largest inland fisheries and revealed short gear lifespans and high turnover rates, lack of appropriate end-of-life gear disposal methods and ineffective fisheries regulations. A biodiversity threat assessment identified the air-breathing aquatic vertebrate species most at risk of entanglement in, and impacts from, derelict fishing gear; namely species of threatened freshwater turtle and otter, and the endangered Ganges river dolphin. This research demonstrates a need for targeted and practical interventions to limit the input of fisheries related plastic pollution to this major river system and ultimately, the global ocean. The approach used in this study could be replicated to examine the inputs, socio-economic drivers and ecological impacts of this previously uncharacterised but important source of plastic pollution in other major rivers, worldwide.

Keywords : ALDFG , Behavioural drivers , Entanglement , Fishers' knowledge , Inland fisheries , River catchments

### A review of microplastics categorization schemes to facilitate source apportionment: Implications for North American freshwaters

Yu Jasmine T., Helm Paul A., Diamond Miriam L..

Microplastics, a widespread pollutant in freshwater systems, have numerous sources which are not well understood. Part of source reductions efforts requires knowledge of sources which is based on confident categorization and characterization. A literature review was conducted to compare the categories used for reporting types of microplastics found in North American freshwater environments. Analysis showed that categorization schemes for grouping microplastic particles are highly variable, with up to 17 different categories used across 32 studies. In some studies, pellets and microbeads are used interchangeably and grouped in a single category or assigned separate categories. Similar observations were seen with the fiber and line categories. Fragments are commonly a 'catch-all' category to describe irregularly shaped particles. Although uncommon, some studies have included source-specific categories such as tire wear, commercial fragments, paint, and irregular microbeads. While there is no universally accepted categorization framework for characterizing microplastics, harmonization would help with source identification. The broad range of categories across studies to report microplastic types creates ambiguity in determining key sources and their contributing load in freshwater environments. Source apportionment efforts would benefit from using particle morphology to assign microplastics to source-specific categories. This will help facilitate cross-study comparisons of microplastic types and help target management and reduction strategies in North America.

Keywords : categorization , freshwater , harmonization , microplastic , sources

### **Hyporheic exchange processes increase retention of microplastics in rivers** Drummond Jennifer D., Nel Holly A., Packman Aaron I., Krause Stefan.

Microplastics are abundantly found in streambed sediments, including both small and low-density particles of neutral and positive buoyancy. Hyporheic exchange, the two-way movement between the overlying water and the streambed sediments, is driven by both turbulence in the near bed region and pressure variations at the streambed surface that force water and suspended particles (e.g., microplastics, fine sediments, microbes) into and out of the sediment porewater. In fact, hyporheic exchange is occurring to some extent in most river systems, which leads to the accumulation of even very small particles with very low settling rates within the hyporheic region. However, the relative importance of hyporheic exchange compared to other mechanisms that lead to streambed retention of microplastics has not yet been assessed. Here we evaluate the effects of hyporheic exchange on microplastics by calculating and comparing the rates of microplastic delivery to streambed sediments by hyporheic exchange and gravitational settling for combinations of particle size and density most commonly found in streams. In a field stream study, we found that 23% of all microplastic combinations have a higher hyporheic exchange rate than settling rate. This fraction was as high as 42% for microplastics composed of low-density polymers, such as polyethylene. We then expand these findings to consider a wide range of hydrodynamic conditions in rivers and demonstrate that hyporheic exchange is important for the transport and fate of particles smaller than 100 µm diameter, irrespective of polymer type. Hyporheic exchange can substantially increase the retention of microplastics in rivers and facilitate modification of these particles prior to reaching the oceans (e.g., through the combination of increased retention and riverbed processes such as fragmentation, biofouling, and aggregation). Models that do not include hyporheic exchange are therefore likely to substantially underestimate the deposition, retention, and long-term accumulation of microplastics in streambed sediments.

Keywords : accumulation , freshwater , hyporheic exchange flow , microplastics , streams and rivers

# Seasonal variation in relative abundance and composition of microplastic particles and zooplankton in a pelagic environment (NE Atlantic)

Sambolino Annalisa, Herrera Inma, Álvarez Soledad, Alves Filipe, Canning-Clode João, Dinis Ana, Kaufmann Manfred, Cordeiro Nereida.

The occurrence of marine litter, mostly plastic items, is on a widespread constant increase. The ingestion of plastic by marine organisms depends on its availability in the environment and its physical properties. Zooplankton represents a crucial food source for many secondary consumers, thus, the relative abundance and similarities between plastic particles and planktonic organisms define the probability for these particles to enter the food web and transfer up the trophic levels. The temporal overlap between the consumers' occurrence, which mainly depends on the presence of the food source, and the debris accumulation also represents a critical factor. In this context, the present study describes the seasonal variation in the abundance, types and sizes of microplastics and zooplankton in a pelagic environment. Monthly samplings of surface waters were carried out off the South coast of Madeira Island (bathymetry ¿ 1000 m) during 2019. The plastic particles collected were counted and classified into different categories (fragment, line, film, foam, fibre) and size classes.

Keywords : Macaronesian waters , marine litter , pollution , sustainable management , trophic web

### Session 26.4\_Ma. Chaired by Patricia Ostiategui, Gran Canaria

#### Exposure to textile microplastic fibers impairs epithelial growth

Van Dijk Fransien, Van Eck Gail, Cole Matthew, Salvati Anna, Bos Sophie, Gosens Reinoud, Melgert Barbro.

Paper number 331805

### The fate of microplastics along salinity gradient and tidal cycles in a well-mixed estuary: a case study of the Seine Estuary

Gasperi Johnny, Dris Rachid, Alligant Soline, Tramoy Romain, Halm Marie-Pierre, Gangnery Aline, Simon Benjamin, Maheux Frank, Cachot Jérome, Tassin Bruno. Paper number 333668

#### Seasonal variation of microplastics in Southampton water, UK

Stead Jessica, Cundy Andrew, Hudson Malcolm, Thompson Charlie, Williams Ian, Russell Andrea. Paper number 334373

### Microplastics in the Biosphere Reserves of Islands and Coastal Zones of Spain: Activities that generate them and ongoing mitigation initiatives

*Estevez Adan Virginia*. Paper number 334582

### Microplastic concentrations in the intertidal sediment and benthic organisms in the Bay of Fundy

*Beardy Krista, Hunt Heather.* Paper number 334641

### Exposure to textile microplastic fibers impairs epithelial growth

Van Dijk Fransien, Van Eck Gail, Cole Matthew, Salvati Anna, Bos Sophie, Gosens Reinoud, Melgert Barbro.

Microplastics are a pressing global concern. Inhalation of microplastic fibers has been associated with interstitial lung disease related to alveolar epithelial damage in nylon flock workers. However, the means by which fibers affect lung tissue and epithelial growth remains unknown. Our aim was to assess the effects of nylon and polyester textile microplastic fibers on epithelial growth and differentiation using airway and alveolar lung organoids cultured from epithelial cell progenitors, isolated both from murine lungs and human lung tissue obtained from COPD patients. Exposure to nylon (11x30 µm) or polyester (15x53 µm) microfibers resulted in significantly fewer and smaller human and murine airway organoids after 14 days of culture, the effect being most profound with nylon. Alveolar organoids were not affected by these fibers. Incubation with nylon- or polyesterconditioned medium also resulted in fewer airway organoids. Effects were mainly observed in developing airway organoids; exposure of developed organoids from day 14 to day 21 to fibers or fiber-conditioned medium had no significant effect on organoid number or size. In conclusion, airway organoid formation is negatively impacted by the presence of textile microplastic fibers and this effect appears to be mediated by leaching additives. Our results suggest that microplastic fibers may especially harm the developing airways or airways undergoing repair. Further studies will focus on identifying these additives and the mechanism behind their effect. Importantly, wider investigations into the presence of microplastic fibers in human lung tissue are urgently needed to determine the actual risk of these fibers to human health.

Keywords : epithelial cells , indoor air pollution , inhalation , lungs , microplastic fibers , textile

# The fate of microplastics along salinity gradient and tidal cycles in a well-mixed estuary: a case study of the Seine Estuary

Gasperi Johnny, Dris Rachid, Alligant Soline, Tramoy Romain, Halm Marie-Pierre, Gangnery Aline, Simon Benjamin, Maheux Frank, Cachot Jérome, Tassin Bruno.

While most of the studies focused on microplastic (MP) pollution in both marine and continental environments, estuaries are poorly documented and their role as an important pathway and a physical filter are still unclear. In this study, the fate of MP was investigated along the salinity gradient of the Seine well-mixed estuary and over two tidal cycles. Four sampling surveys were conducted at three stations during the same day (La Bouille-upper estuary, Vieux-Port-middle estuary and La Roquelower estuary) along the salinity gradient. For each survey, surface water (first 15 cm including the sea surface microlayer - SML) and sub-surface water (first 50 cm excluding SML) were collected using a 300-µm mesh size manta net during the ebb tide. Fragments and microbeads were considered while microfiber pollution was excluded. Concentrations in surface waters were 5 to 6 times higher than those in subsurface waters, due to the significant accumulation of MP in SML. Differences were observed along the salinity gradient with the highest concentrations measured at Vieux-Port located upstream the maximum turbidity front (MTF) and lower concentrations at La Bouille and La Roque stations. This could be explained by i) the salt intrusion and ii) the accumulation of macrolitters in the vicinity of Vieux-Port. Two tidal cycles were also investigated in La Roque, which is characterized by the presence of the MTF. For both cycles, the vertical distribution was examined by sampling sub-surface water, middle-water column and bottom layer during flood and ebb tides. Results clearly demonstrated a vertical gradient between water collected at the bottom exhibiting the highest concentrations while lowest values were observed in surface water. This gradient can be explained by both the resuspension of bottom sediments, rich in MP, during the flood tide and sedimentation during the long water high period and the ebb tide.

Keywords : estuary , Microplastic , tide cycle , vertical distribution

### Seasonal variation of microplastics in Southampton water, UK

Stead Jessica, Cundy Andrew, Hudson Malcolm, Thompson Charlie, Williams Ian, Russell Andrea.

Recorded abundances of microplastics in the environment vary by several orders of magnitude, with a range of environmental factors being argued to have an impact (or no impact) on these abundances. In order to develop a more detailed understanding of how environmental factors might influence abundances of microplastics in estuaries, surface microlayer (SML) and bulk water sampling was employed to assess daily and seasonal variation in microplastics abundance at two locations in Southampton Water, U.K. High variation in suspected microplastic concentration was observed with both sampling methods, and at both locations. Suspected microplastic abundance in the SML ranged between 1 and 356 suspected microplastics/L. Observed concentrations were seasonally higher at the more sheltered River Itchen site, and more variation was seen at this site, as compared to the more exposed estuarine site at Mayflower Park. The wide range in suspected microplastic abundance suggests that repeated sampling of locations over a period of time under a range of meteorological and oceanographic (i.e. tidal) conditions is necessary to reflect the variation of abundances at any given site, and to enable an accurate assessment of microplastic abundance and fluxes in the estuarine system. While a variety of meteorological and oceanographic conditions were recorded over the four months sampled, no one variable appeared to be a driving factor determining microplastic abundance in Southampton Water. This supports previous (non microplastic-related) studies in Southampton Water which define it as a well-mixed system. A significant difference was found between suspected microplastic abundances sampled using SML and bulk water sampling, with the SML-specific method (glass plate sampling) more clearly highlighting inter-site differences. This suggests that the SML glass plate method is a more sensitive method for sampling microplastics from estuarine waters.

Keywords : estuary , sea surface microlayer , seasonal variation

### Microplastics in the Biosphere Reserves of Islands and Coastal Zones of Spain: Activities that generate them and ongoing mitigation initiatives

Estevez Adan Virginia.

As stated in Directive 2008/56 /EC of the European Parliament and the Council of June 17, 2008, the marine environment is a precious heritage that must be protected, preserved and, where practicable, restored. In Spain Law 41/2010, of December 29, on the protection of the marine environment, regulates it. The National Parks Autonomous Agency (OAPN) is also committed to this goal, since one of its responsibilities is to coordinate the MaB Program, and to promote and support the Spanish Network of Biosphere Reserves, which currently encompasses 52 territories, and 15 of them have the marine environment as part of it. All of them face common challenges, as the increasingly abundant presence of marine litter in the seas and oceans. While the vast majority of it, it is generated on land, a significant part ends up within the marine environment. Plastics, and in particular microplastics, are a typology of marine litter. Their small size facilitates their ingestion by marine fauna, which poses a risk for human health. This communication focuses on the human activities that are taking place in the Spanish Island and Coastal Biosphere Reserves, and on the potential plastic waste that they can generate. The responsible of these territories are aware of the danger that this constitutes for the maintenance of marine ecosystems. For this reason, diverse research and collaboration initiatives are ongoing. The objective pursued with them is three-fold: (i) learning about the effects of microplastics on the marine environment, and on human health; (ii) raising awareness between all productive sectors, which are potential generators of plastics; (iii) understanding the importance of taking measures to reduce them, while promoting circular economy. These initiatives are described within this communication, so that they provide an overview of the work being carried out, and the main results achieved so far.

Keywords : Island and Coastal Biosphere Reserves , MaB Program , National Parks Autonomous Agency (OAPN) , plastic waste

# Microplastic concentrations in the intertidal sediment and benthic organisms in the Bay of Fundy

Beardy Krista, Hunt Heather.

Microplastics (<5mm) in marine and coastal habitats continue to be a contaminant of emerging concern in Atlantic Canada. Originating primarily from the degradation of larger plastic debris deposited in the marine environment, microplastics become widely distributed and can accumulate in marine sediments where they can leach toxins introduced in the original production processes (i.e. bisphenol A, phthalates). Microplastics can also adsorb hydrophobic background contaminants such as persistent organic pollutants (POPs) which has the potential to bioaccumulate through the marine food web as microplastics become an unintended food source. Microplastic accumulation affects all marine and coastal wildlife, increasing mortality and reducing the availability of healthy coastal habitat. This study builds upon work conducted in the southwestern New Brunswick portion of the Bay of Fundy and continues to quantify regional microplastic distribution in intertidal marine sediments and the potential for biological uptake in bivalve communities. Bivalves and adjacent intertidal sediment were harvested from various sites along the Bay of Fundy coastline. Microplastics were separated from bivalve tissue using a potassium hydroxide digestion. For sediment samples, microplastics were removed by density separation in a concentrated salt solution. Both bivalve and sediment samples were filtered through a 0.22 µm membrane and inspected under a dissecting microscope (40x). Data analysis will assess the relationship between microplastic concentrations in intertidal sediments and bivalve communities and will consider proximity to potential sources of this contamination as well as potential hot spots for coastal microplastic production.

Keywords : Atlantic Canada , Bay of Fundy , Bivalves , Microplastics , Sediment

### Session 26.5\_O. Chaired by Jesús Gago, Vigo

Effects of nanopolystyrene beads on oyster early life stages (gametes and embryos) and investigations of long-term repercussions Tallec Kevin, Paul-Pont Ika, Huvet Arnaud. Paper number 332893

### Monitoring of microplastics in UK coastal and marine sediments using a rapid-screening approach based on fluorescent tagging with Nile Red

Bakir Adil, Wilkinson Tim, Doran Denise, Maes Thomas. Paper number 334206

### The occurrence of paraffin waxes floating in the southern North Sea

Lorenz Claudia, Schafberg Michaela, Roscher Lisa, Meyer Melanie S., Primpke Sebastian, Kraus Uta R., Gerdts Gunnar. Paper number 334531

### Microplastic and artificial cellulose microfibers ingestion by reef fishes in the southwestern Atlantic

Macieira Raphael, Oliveira Leticia Aparecida Da Silva De, Cardozo-Ferreira Gabriel C., Pimentel Caio Ribeiro, and rades Ryan, Gasparini João Luiz R., Sarti Francesco, Chelazzi David, Cincinelli Alessandra, Gomes Levy Carvalho, Giarrizzo Tommaso. Paper number 334579

# Effects of nanopolystyrene beads on oyster early life stages (gametes and embryos) and investigations of long-term repercussions

Tallec Kevin, Paul-Pont Ika, Huvet Arnaud.

Over a 3-years PhD project we have investigated the acute toxicity of nanopolystyrene beads (50 nm) on oyster gametes (behavior, fertilization success) and embryos (developmental success), as well as subsequent repercussions of an early exposure at a sub-lethal dose on the larval and adult stages over two generations of oysters. In the acute toxicity experiments, dose-response effects were observed from 10, 1 and 0.1 µg.mL-1 on spermatozoa mobility, fertilization success and embryolarval development success, respectively. Furthermore, the toxicity was dependent on the surface functionalization, amino-nanopolystyrene beads exhibiting the highest toxicity, compared to plain or carboxylic-nanopolystyrene beads, due to their high stability in seawater (aggregates ; 100 nm) and their positive charge increasing interactions with cell membranes. An early embryonic exposure at a sublethal dose reduced significantly the oyster larval growth (by 9 to 21%) leading to a delay in larval settlement but no long term consequences were observed on juvenile and adult stages in terms of growth, reproductive outputs or ecophysiological parameters (e.g. respiration and filtration rates). On the second generation of larvae, no significant effects were detected on the selected endpoints whether they had been exposed to early embryonic exposures at the first or at both generations, suggesting no stress-memory mechanism. An intermediate but non-significant growth response of oyster larvae exposed at both generations opens a question of further interest on the occurrence of adaptive response allowing higher tolerance. Overall, our results suggested a limited risk of nanopolystyrene beads for oyster early life stages according to the LOECs from acute toxicity experiments and data provided by our long-term experiment. Nevertheless, because the features and the current environmental concentrations of nanoplastics remained unknown, this assumption must be confronted in the future when methods will be available to quantify nanoplastics at sea providing accurate risk scenarios.

Keywords : Early life stages , Nanoplastics , oyster , toxicity

### Monitoring of microplastics in UK coastal and marine sediments using a rapidscreening approach based on fluorescent tagging with Nile Red

Bakir Adil, Wilkinson Tim, Doran Denise, Maes Thomas.

Microplastics in the marine environment are of global concern due to their abundance, widespread distribution, and unknown environmental effects. Different methods are available for their quantification in the marine environment and their monitoring has been mainly focusing on the water column, biota, and sediments. Currently, there are no universally accepted standardised methods for any of these matrices. This study presents the findings and recommendations from a Research & Development study on the development of a rapid screening approach for the detection and quantification of microplastics in sediment based on a fluorescent tagging method using Nile Red coupled with an automated recognition software. Coastal and marine sediment samples were collected as part of the Clean Seas Environment Monitoring Programme (CSEMP). Coastal and offshore stations were also selected to investigate any gradient in the abundance of microplastics from coastal to offshore areas. Other factors, including the sediment type (particle size) and temporal variations were also considered. Microplastic particles were detected in almost all the sediment samples from all the stations selected around the UK. Spatial differences were also characterised for the different stations under investigation. This method was found to be in alignment with previous recommendations that the Nile red method is a promising approach for the largescale mapping of microplastics in a monitoring context. Addressing the knowledge gaps in monitoring data would allow the creation of "exposure risk maps" for the area with the mapping of accumulation zones for microplastics and more sensitive areas (e.g. marine protected areas, fishing zones). Exposure risk maps would be the first step in developing robust and reliable risk assessment frameworks based on environmental relevant conditions such as concentrations, size, and type of plastics items in the natural environment.

Keywords : marine litter , Microplastics , Nile Red , Particle Size Analysis , UK sediments

### The occurrence of paraffin waxes floating in the southern North Sea

Lorenz Claudia, Schafberg Michaela, Roscher Lisa, Meyer Melanie S., Primpke Sebastian, Kraus Uta R., Gerdts Gunnar.

Marine pollution occurs in many different forms. One more recently addressed emerging pollutant is paraffin wax (PW) as petroleum-based semi-solid. As such, PWs have been included in several monitoring guidelines and are regularly recorded washed-up on beaches. However, knowledge on their occurrence, particularly as persistent floaters of small size (To investigate this matter, samples collected by net-sampling (100µm-mesh) at 24 stations in the southern North Sea, were screened for putative PWs. Particles (500–5000 um) of certain wax-like appearance were detected at 14 of these stations. From six of these stations with the highest abundances, putative PWs with a similar appearance were pooled per station and analyzed by ATR-FTIR (Attenuated total reflectance Fouriertransform infrared spectroscopy) and thermoanalytical methods, namely gas chromatography coupled with a flame ionization detector and coupled with a mass spectrometer (GC-FID and GC-MS). The pooled samples contained mainly PW particles, being partly accompanied by substances like fatty acids and fatty alcohols. Utilizing both analytical techniques provided a reliable detection of PWs and a more detailed understanding of their chemical composition. Furthermore, the presence of PWs of smaller size (20–500  $\mu$ m) was proven exemplarily on the sample with the highest concentration of PWs by µFTIR imaging. This study gives valuable insights into PW pollution in the North Sea, further emphasizing the need for harmonized sampling and detection methods, ideally accompanying regular monitoring of microplastics.

Keywords : ATR , FID , FTIR , GC , marine pollution , MS , Petroleum waxes , sea based sources
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# Microplastic and artificial cellulose microfibers ingestion by reef fishes in the southwestern Atlantic

Macieira Raphael, Oliveira Leticia Aparecida Da Silva De, Cardozo-Ferreira Gabriel C., Pimentel Caio Ribeiro, and rades Ryan, Gasparini João Luiz R., Sarti Francesco, Chelazzi David, Cincinelli Alessandra, Gomes Levy Carvalho, Giarrizzo Tommaso.

This study evaluated the ingestion of microplastic and artificial cellulose in 103 fishes belonging to 21 reef fish species from the southwestern Atlantic. Nine species ingested particles- seven species both types and two only one kind. Fishes ingested more man-made cellulose fibers than microplastics, and these particles were mainly found in the stomach. Haemulon aurolineatum ingested more microplastic and artificial cellulose particles than other species. Overall, transparent particles were dominant in our data. Polyamide was the commonest particle of microplastic, being associated with cellulose microfibers from washing clothes thought wastewater systems. Household sewage, fishery activity and marine navigation might be main sources of artificial particles for these reef fishes. This work provides a background of microplastic and artificial cellulose contamination. These information are important to understand the anthropogenic impact over a poor-examined marine group, the tropical reef species, as well as guidelines to mitigate plastic pollution on reefs.

Keywords : based fibers , cellulose , marine fish , Marine pollution , plastic , polymers

### Session 26.5\_Me. Chaired by Patricia Ostiategui, Gran Canaria

**The seasonal changes of plastic in Flesh Footed-Shearwaters (***Ardenna carneipes***)** *Paterson Harriet, Ford Benjamin, Woltjen Jaynie.* Paper number 334249

### Assessment of micro- and nanoplastics release from food packaging under simulated use conditions: challenges and analytical strategies

Cella Claudia, La Spina Rita, Mehn Dora, Valsesia Andrea, Fumagalli Francesco Sirio, Gilliland Douglas. Paper number 334324

Paper number 334324

### Occurrence of microplastics in household laundry water: quantification from a developing country

praveena sarva mangala, asmawi melati syahira, chyi josephine liew ying. Paper number 334372

### Microplastic spatial distribution patterns within the intertidal of beaches and estuaries: the influence of local variability and environmental conditions

*Vermeiren Peter, Munoz Cynthia.* Paper number 334384

### **The seasonal changes of plastic in Flesh Footed-Shearwaters (***Ardenna carneipes***)** Paterson Harriet, Ford Benjamin, Woltjen Jaynie.

The south coast of Western Australia is isolated from the major population centres of the world, but it is still subject to the impact of marine plastic. The many islands of the coast are home to a variety of sea bird species, including the Flesh Footed-Shearwaters (Ardenna carneipes). This species over winters in the Bay of Bengal and the North Pacific and then breeds on the Australian islands during the austral summer. During the breeding season these birds can be caught in nets as bycatch by the local pilchard industry where they compete for the fish during the last stages of chick rearing in April/May. However this provides the opportunity to routinely estimate the plastics load of this species. In October 2017 there was an unusual wreck of this species allowing us to compare the plastic load at the beginning and end of the breeding seasons in 2017. The Frequency of Occurrence of plastic for October was 60% compared to 20% in April/May. The processes that affect the plastic loads of birds may differ at the beginning and end of the breeding season. The birds may have imported plastic during their migration or consumed large amounts on arrival, while at the end of the season the local oceanography may reduce the availability of plastic. This research indicated that plastic – bird phenology is required to fully understand the interaction between fauna and plastic.

Keywords : ingestion , Marine Birds , Migration , South West Australia

# Assessment of micro- and nanoplastics release from food packaging under simulated use conditions: challenges and analytical strategies

Cella Claudia, La Spina Rita, Mehn Dora, Valsesia Andrea, Fumagalli Francesco Sirio, Gilliland Douglas.

The anthropogenic release of micro- and nanoplastics is an emerging issue since these particles constitute a ubiguitous and growing pollutant which not only threatens the environment but may have potential consequences for human health. In particular, there is concern about the release of secondary micro and nanoplastics from the degradation of plastic consumer products. The phenomenon is well documented in relation to plastic waste in the environment but more recently there have been reports of microplastic generated during the normal use of plastic food contact materials such as water bottles, tea bags and containers. Unfortunately, understanding these events is hindered by the lack of harmonized analytical methods in the field. In this study, we demonstrate that microplastics breakdown occurs during the normal use of polyethylene rice cooking bags and icecube bags, as well as from nylon teabags. Methods developed for the analysis of microplastics in environmental samples were explored as the starting point for identification and quantification of micro and nanoplastics coming from food contact materials. Our method includes testing particle release under conditions that mimic their intended use, by applying heat, microwaves, freeze-thaw or immersion in boiling water. A critical step is sample preparation - by lyophilisation or filtration - to concentrate particles on functionalized surfaces enabling the chemical recognition. Raman and Fourier-Transformation InfraRed (FT-IR) microscope technologies were applied to identify plastics as well as any other material coming from the packaging, such as dyes or food residues. Finally, mixtures of potassium bromide (KBr) and plastic particles were prepared to create pellets and perform FT-IR measurements. Integration of specific peaks in the FT-IR spectra allowed to build calibration curves and to quantify the mass of micro- and nanoplastics release from the packaging materials.

Keywords : contaminants , food contact material , method development , microplastics , nanoplastics , particle release , plastic quantification

# Occurrence of microplastics in household laundry water: quantification from a developing country

Praveena Sarva Mangala, Asmawi Melati Syahira, Chyi Josephine Liew Ying.

Microplastics emission via household washing machine has been reported as one of the microplastics sources. However, contribution of washing machine use to microplastics emission at the household level has been understood mostly in developed countries and still not completely understood in developing countries. In Malaysia, microplastics studies have reported involving sea water and river water despite the absence of studies relating to microplastics in grey water from household washing machine. This study aims to identify microplastics occurrence in household laundry water along with along with association between washing machine usage at household level. This cross sectional study was conducted involving 99 households from Klang Valley (Malaysia). Microplastics in laundry water samples were extracted via filtration method and identified in terms of size and morphology. Microplastics were found between 6.9E-3 g/m3 and 0.183 g/m3 in laundry water at household level. Microplastics shapes of fibre and fragment consist of polyester, nylon and acrylic with average length of 2258.59 µm were also identified in these laundry water samples. Morphology microplastics in grey water sample has indicated that the highest percentage was fibers (71%), followed by fibers and fragments(20%), mixture of fibers, fragments and pellet (9%). Questionnaire survey findings demonstrated fabric properties and washing parameters both likely contribute to microplastic emissions in laundry water and, ultimately, wastewater treatment plant influent. The impact of fabric properties and washing parameter factors on microplastics emission in laundry water at the household level merits further investigation. The findings of this study demonstrated the potential of laundry water as a microplastic source at the household level within a developing country.

Keywords : emission , household , laundry water , microplastics

# **Microplastic spatial distribution patterns within the intertidal of beaches and estuaries: the influence of local variability and environmental conditions** Vermeiren Peter, Munoz Cynthia.

Microplastic contamination in the intertidal zone of beaches and estuaries has been reported worldwide. Nonetheless, evidence of patterns in the small-scale distribution of microplastics within the intertidal zone is often contradictory. When such conflicting evidence is used to inform sampling campaigns, it increases uncertainty in resulting data. Moreover, the conflicting patterns hamper efforts in spatially explicit risk characterization of microplastic pollution to intertidal organisms. This study aimed to analyze spatial patterns in microplastic concentrations within the intertidal of beaches and estuaries. A laboratory protocol for cost-effective identification and quantification of microplastics from sediments was developed and validated. This protocol was used to sample microplastic concentrations in sediments at intermediate (60 m transects) and local (1 m2 quadrats) scales across sites with varying levels of urbanization. Large (5.0 mm – 0.5 mm) and small (0.5 mm -0.125 mm) microplastics were quantified using the protocol, and smaller microplastics ( $\geq 0.033$ mm) could be detected. Polymers with densities higher and lower than seawater were encountered in roughly equal proportions in both ecosystems. Our results suggest that sufficient sample replication is needed to account for the high variability in microplastic concentrations at local scale. Moreover, despite the drift zone often being targeted during microplastic monitoring, microplastic concentrations varied across beach zones without evidence of accumulation at the drift zone. Alternatively, local environmental factors such as sediment grain size and structural complexity of estuarine habitats, and the presence of larger plastics as a local source of microplastics, related to measured concentrations in microplastics. We recommend that samples should be spread across the intertidal to account for small scale variability, and that local environmental variables should be considered as confounding factors when comparing contamination levels among beaches or estuaries.

Keywords : beach , estuary , laboratory protocol , sediment , spatial distribution

### Session 26.5\_Ma. Chaired by Mateo Cordier, Guyancourt

**Rapid fragmentation of microplastics by the freshwater amphipod Gammarus duebeni (Lillj.)** *Mateos Cárdenas Alicia, O'halloran John, Van Pelt Frank, Jansen Marcel.* Paper number 333642

### Impacts to larval fathead minnows vary between pre-consumer and environmental microplastics

*Bucci Kennedy, Bikker Jacqueline, Stevack Kathleen, Watson-Leung Trudy, Rochman Chelsea.* Paper number 333722

### Environmental and ecological determinants of microplastic pollution and ingestion by freshwater organisms in the Garonne River (France)

Carvalho Aline, Garcia Flavien, Riem-Galliano Louna, Tudesque Loic, Albignac Magali, Ter Halle Alexandra, Cucherousset Julien. Paper number 334417

### Contribution and factors of influence of outdoor apparel to microfibre release

Cocca Mariacristina, De Falco Francesca, Di Pace Emilia, Avella Maurizio, Scholz Bernadette, Fox Ruediger, Mayershofer Martin. Paper number 334466

# Rapid fragmentation of microplastics by the freshwater amphipod *Gammarus duebeni* (Lillj.)

Mateos Cárdenas Alicia, O'halloran John, Van Pelt Frank, Jansen Marcel.

Microplastics have become ubiquitous in all environments. Yet, their environmental fate is still largely unknown. Plastic fragmentation is a key component of plastic degradation, which is mostly caused by abiotic processes over prolonged time scales. Here, it is shown that the freshwater amphipod Gammarus duebeni can rapidly fragment polyethylene microplastics, resulting in the formation of differently shaped and sized plastic fragments, including nanoplastics. Fragments comprised 65.7% of all observed microplastic particles accumulated in digestive tracts. Higher numbers of fragments were found in response to longer exposure times and/or higher microplastic oncentrations. Furthermore, the proportion of smaller plastic fragments was highest when food was present during the depuration process. It is concluded that G. duebeni can rapidly fragment polyethylene microplastics and that this is closely associated with the feeding process. These results highlight the crucial role, currently understudied, that biota may play in determining the fate of microplastics in aquatic ecosystems. DOI https://doi.org/10.1038/s41598-020-69635-2

Keywords : crustacean , fragmentation , freshwater , microplastics

### Impacts to larval fathead minnows vary between pre-consumer and environmental microplastics

Bucci Kennedy, Bikker Jacqueline, Stevack Kathleen, Watson-Leung Trudy, Rochman Chelsea.

Microplastics are a complex suite of contaminants varying in size, shape, polymer, and associated chemicals, and are sometimes referred to as a multiple stressor. Still, the majority of studies testing hypotheses about their effects use commercially bought microplastics of a uniform size, shape, and type. In this study, we investigated the effects of pre-consumer polyethylene (PE), pre-consumer polypropylene (PP), and a mixture of PE and PP collected from the shoreline of Lake Ontario, where they likely sorbed a complex cocktail of contaminants. Embryo-stage fathead minnows were exposed to physical plastic particles or chemical leachates alone at an environmentally relevant (280 particles/L) or high (2,800 particles/L) concentration for 14 days. We have shown that the effects of microplastics differ by polymer type and presence of environmental contaminants, and that effects can be driven by the physical particles and/or the chemical leachates alone. Larvae exposed to preconsumer PE experienced a decrease in survival, length, and weight, while pre-consumer PP caused an increase in weight. Environmental microplastics caused a more drastic increase in length and weight, and almost 6x more deformities than either of the pre-consumer microplastics. While preconsumer microplastics caused effects in the physical treatment only, the environmental microplastics caused effects in both the physical and chemical treatments, suggesting that the mechanism of impacts is context dependent. This study provides further support for treating microplastics as a multiple stressor and begs further research on the effects of environmentally relevant concentrations of microplastics on the long-term growth, survival, and reproductive output of aquatic organisms.

Keywords : ecotoxicology , effects testing , fathead minnows , freshwater , microplastic , pimephales promelas , Plastic pollution

# Environmental and ecological determinants of microplastic pollution and ingestion by freshwater organisms in the Garonne River (France)

Carvalho Aline, Garcia Flavien, Riem-Galliano Louna, Tudesque Loic, Albignac Magali, Ter Halle Alexandra, Cucherousset Julien.

Microplastic (MP) pollution and the ingestion of MP by organisms is now considered as a ubiquitous phenomenon in freshwater ecosystems. However, our understanding of the dynamics of MP pollution and the drivers modulating MP ingestion by macroinvertebrates and fish is still limited. In this study, we first quantified the spatial and temporal variability of MP (700  $\mu$ m – 5 mm) pollution in surface water in 14 sites located across the Garonne river catchment (France). We then quantified the level of MP contamination in macroinvertebrates and fish and used stable isotope analyses ( $\delta$ 13C and  $\delta$ 15N) to quantify trophic niches. Results revealed that spatial variation in MP concentration was high and driven by urbanisation. Temporal variation in MP concentration and size was driven by hydrological variations, with higher concentrations and smaller particles sizes observed in warm seasons with low discharge. The proportion of the main MP polymers (polyethylene, polystyrene, and polypropylene) did not significantly differ between sampled sites and MP colour differed between polymer types, with a high proportion of white polystyrene particles. Overall, fish had a significantly higher abundance of ingested microplastics than macroinvertebrates. MP ingestion by macroinvertebrates and fish was not significantly related to MP pollution in surface water and sediment. Interestingly, MP characteristics (shape, colour, size, and polymer composition) significantly differ between the abiotic (surface water and sediment) and biotic (macroinvertebrates and fish) compartments. The MP abundance increased with the size of organisms for macroinvertebrates and fish. Stable isotope analyses revealed that MP abundance tended to increase with trophic position in macroinvertebrates only. The origin of the resource consumed (allochthonous vs. autochthonous) significantly affected the abundance of microplastic ingested by fish. Altogether, these results highlight the complex dynamic of MP pollution in freshwater ecosystems and suggest the absence of microplastic bioaccumulation in freshwater food webs.

Keywords : hydrology , sediments , surface water , trophic niche , trophic position , urbanisation

Contribution and factors of influence of outdoor apparel to microfibre release

Cocca Mariacristina, De Falco Francesca, Di Pace Emilia, Avella Maurizio, Scholz Bernadette, Fox Ruediger, Mayershofer Martin.

Laundry processes of synthetic clothes are considered one of the major source of microplastics pollution of aquatic ecosystems. The production and purchasing of synthetic fibres have been constantly increasing in the last decade, and they are mainly used in the apparel industry. Outdoor garments represents a particular case in garment production since their production mostly relies on the usage of high-tech functional fabrics. Such textiles are characterized by properties like waterproofness, breathability and climate-regulating performances, that are achieved through the combination of high performance materials in a laminate structure. Up to know, laminated fabrics have been not investigated yet to evaluate their release of microfibres during washing. Several types of laminated fabrics were selected to cover a wide range of different raw materials, textile structures and characteristics. For each type of laminates, samples coming from different steps of its production were tested. These steps includes raw materials and laminates with or without durable water repellent (DWR) treatments. The release of microfibres was evaluated by testing the samples using a laboratory simulator of washing processes, filtering the washing water and analysing the obtained filters by scanning electron microscopy (SEM). Results showed for the first time the quantities of microfibres that can be released by washing of laminated fabrics. The data obtained were statistically analysed, highlighting which textile characteristics and processing steps have a decreasing or increasing effect on the release of microfibres. These outcomes could provide useful information to laminated fabrics producers on how to reduce the microfibre emission from their products.

Keywords : laminated fabrics , microfibres , microplastics , outdoor apparel , textile production

### Session 26.6\_O. Chaired by Bethany Jorgensen, Ithaca

### Sources, contamination, and fate of microplastics and other anthropogenic microdebris in an urban bay: A case study in San Francisco Bay

Zhu Xia, Munno Keenan, Grbic Jelena, Werbowski Larissa Meghan, Bikker Jacqueline, Ho Annissa, Guo Edie, Sedlak Meg, Sutton Rebecca, Box Carolynn, Lin Diana, Gilbreath Alicia, R.c. Holleman, Fortin Marie-Josee, Rochman Chelsea. Paper number 331977

#### Plastic pellet pollution on florida's space coast – a citizen science approach

Sluka Robert, Paterson Ann, Mcmann Stephen, Ederer Brittany, Hickman Caleb, Johnson Paige, Mclemore Kyrsten. Paper number 332355

### Optimization of the citizen protocol for microplastic sampling in surface water with Babylegs net

*Villanueva Marilou, Durantou Lise, Lanceleur Laurent, Monperrus Mathilde, Schaal Alexandre.* Paper number 333953

# Sources, contamination, and fate of microplastics and other anthropogenic microdebris in an urban bay: A case study in San Francisco Bay

Zhu Xia, Munno Keenan, Grbic Jelena, Werbowski Larissa Meghan, Bikker Jacqueline, Ho Annissa, Guo Edie, Sedlak Meg, Sutton Rebecca, Box Carolynn, Lin Diana, Gilbreath Alicia, R.c. Holleman, Fortin Marie-Josee, Rochman Chelsea.

Plastic particles less than 5 mm in diameter, also known as microplastics, have an enhanced ability to interact intimately with a greater diversity of organisms due to their smaller size. To mitigate risk, there is a need to trace microplastics found in nature back to their original sources to inform prevention. The physical and chemical properties of microplastics and their environmental distributions provide clues as to their sources and inform their fate. Here, we present a case study of local monitoring in San Francisco Bay, USA. Surface water, fish, sediment, stormwater runoff, and treated wastewater were sampled across the Bay and adjacent National Marine Sanctuaries (NMS). Appropriate clean-up and quantification procedures were applied to samples, and Raman and FTIR spectroscopically confirmed material types. We found microplastics and other anthropogenic microdebris in all sample types. Concentrations were greater in the Bay than in the NMS, and, within the Bay, greater during the wet season than the dry season. Different morphologies dominated different sample matrices, indicating that the fate of microdebris varied depending on their morphologies and densities: fibers were common in fish, black rubbery fragments in sediment, and buoyant fragments and fibers in surface waters. Stormwater contained predominantly black rubbery fragments and was a significant pathway for microplastics and other anthropogenic microdebris, with concentrations 140 times higher than wastewater, which was dominated by fibers. Thus, potential sources of microplastics and other anthropogenic microdebris to the Bay include rubber from car tires and fibers from textiles. Overall, we demonstrate the value of multi-matrix regional studies to evaluate sources and fate of microplastics, which can inform effective mitigation. We propose strategies to help mitigate microdebris in the Bay including the installation of rain gardens and stormwater catch basin filters as well as behavioural campaigns to reduce the littering of plastic items.

Keywords : contaminants , estuaries , fate , plastic pollution , regional monitoring , San Francisco Bay , sources

**Plastic pellet pollution on florida's space coast – a citizen science approach** Sluka Robert, Paterson Ann, Mcmann Stephen, Ederer Brittany, Hickman Caleb, Johnson Paige, Mclemore Kyrsten.

Plastic pellets, also known as nurdles, are primary microplastics forming the basis of the plastic production economy. Nurdles are lost to the environment through the transport and manufacturing process ending in the sea and ultimately on beaches globally. Little is known about the sources and sinks of plastic pellets in Florida. Research was conducted to understand the spatial and temporal distribution of plastic pellet pollution on Space Coast beaches using standardized "nurdle hunts." This is a simple method developed for implementation by citizen scientists, but which can also be used by experienced scientists to study spatial and temporal trends. Standardized 15-minute hunts were conducted primarily along Cape Canaveral National Seashore and south of Cape Canaveral for comparison in more developed regions of Brevard County. Citizen scientists ranged from elementary, middle and high school students to adults with little to extensive research experience. Temporal trends indicated the importance of storm events in bringing nurdles to shore. Spatial trends indicated similar numbers of nurdles found north and south of Cape Canaveral, with comparison to other areas of Florida also indicating ecologically similar abundance. Education was an important component of this study and this tool was useful for engaging students and the general public. All results are contributed to The Great Nurdle Hunt database which maps the results of nurdle hunts globally, allowing the research to contribute to wider use by the international research community. Future research will continue to expand work throughout Florida and identify land-based sources of plastic pellet pollution.

Keywords : Florida USA , nurdle , plastic pellet , sandy beach

# Optimization of the citizen protocol for microplastic sampling in surface water with Babylegs net

Villanueva Marilou, Durantou Lise, Lanceleur Laurent, Monperrus Mathilde, Schaal Alexandre.

Microplastic (MP) pollution in fresh and marine waters is actually widely assessed by the scientific community. Both the estimation of the volume of filtered water and the size of the net mesh are crucial information when comparing datasets based on quantity, concentration and flux of MP. Frequently, sampling is proceeding with a Manta net of 300 µm mesh size and a duration of 30 min filtration (Viršek et al. 2016). The challenge for citizen science is to develop simple and affordable tools that could be comparable to standardised methods. The Babylegs net, a device consisting of a plastic bottle cut on each end for the opening, extended with baby's tights as a net. The improvement of this handicraft system, which is inexpensive and easy to use, will make the MP collection and the sharing of usable results accessible to all. The aim of this project is to compare for the first time, a scientific tool (the Manta net) and the Babylegs net in the sampling of MP in freshwater. A sampling campaign of 14 samples was performed at three sites in the Adour River catchment area (France). These samples were analysed according to the National Oceanic Atmospheric Administration method. Each MP was then measured using the CoolingTech software. The average concentration obtained is  $1.7 \pm 0.5$  items/m<sup>3</sup> for the Manta net and  $1.3 \pm 0.3$  items/m<sup>3</sup> for the Babylegs net. No significant difference in concentration between the two sampling systems was observed. However, small differences in the particle size distribution were measured between the two devices, with fewer fragments smaller than 0.6 mm sampled by Babylegs, certainly lost by the widening of the tights mesh. These results validate the use of Babylegs device for the MP monitoring and led to recommendations for the use and the optimization of this method.

Keywords : Adour catchment area , citizen science , continental water , methodology , microplastics , surface sampling

### Session 26.6\_Me. Chaired by Alex Cortada, Menorca

Plastic litter in the Patos Lagoon estuary and adjacent coast

Soares Leonardo, Guedes Gisele, Lacerda Ana Luzia De Figueiredo, Proietti Maíra. Paper number 334544

### Monitoreo de microplásticos para la implementación de políticas públicas en la RB Isla Cozumel Hernández Brenda, Arista Itzel, Fuentes Karen. Paper number 334546

### First characterization of marine litter collected on Fort Dauphin and Sainte Marie Island, East Coast of Madagascar

Thibault Margot, Ramanampamonjy Aina, Saloma Anjara. Paper number 334603

# Citizen science for microplastic detection and analysis -A comparison with other fields of science – research limits and possibilities in the field of microplastic

*Cyvin Jakob, Lacerda Ana Luzia.* Paper number 330575
### **Plastic litter in the Patos Lagoon estuary and adjacent coast**

Soares Leonardo, Guedes Gisele, Lacerda Ana Luzia De Figueiredo, Proietti Maíra.

Plastics are the main type of litter in marine environments, but its fate and impacts are still being unraveled. Plastics at sea have both ocean and land-based sources, and although rivers and estuaries are great contributors of land-originated plastics, this kind of pollution is poorly documented in these environments. We investigated the abundance and characteristics of floating litter collected in surface waters of the Patos Lagoon estuary and its adjacent coastal region. Patos Lagoon is one of the largest coastal lagoons in the world, harboring the second largest Brazilian port in its estuarine portion, and being an important fishing site. Litter was collected using a fishing net (9 m x 1.80 m, mesh sizes: 13 mm in the wings and 5 mm in the center) at five stations within the estuary and two stations in the adjacent coast. Sampling was carried out monthly between August 2019 and February 2020. The collected items were counted, weighed and classified according to type, color and probable source (UNEP, 2016). A total of 2,784 items (weighing 4496 g) were collected, with plastic representing 99% of the materials. Most plastics were fragments (78%) and lines (19%). Transparent (51%), white (22%) and blue (17%) were the dominant colors of items. From the total, 311 items were sampled in estuarine waters, while 2,443 were from the coastal region immediately adjacent to the access jetty of the estuary. This suggests that the jetty could act as a barrier accumulating litter at this portion. The dominance of fragments in transparent/white colors suggests the breakdown of larger items in the environment, and the high amounts of nylon lines indicate contamination caused by fishing activities. Monitoring the types and sources of plastics is essential for mitigating its impacts.

Keywords : coastal lagoon , floating plastic , plastic pollution , southwest Atlantic

# Monitoreo de microplásticos para la implementación de políticas públicas en la RB Isla Cozumel

Hernández Brenda, Arista Itzel, Fuentes Karen.

Dentro de la Reserva de la Biósfera Isla Cozumel, se encuentra el Parque Nacional Arrecifes de Cozumel (PNAC), sus ecosistemas arrecifales pertenecen a la segunda barrera arrecifal más grande del mundo, el Sistema Arrecifal Mesoamericano. El PNAC alberga 120 especies de corales, sin embargo, esta riqueza se vió amenazada por una enfermedad reportada en octubre de 2018, el Síndrome Blancö, el cual mató una colonia completa de corales duros en cuestión de semanas. Aunque aún no se tiene certeza del patógeno ni vía de contagio de dicha enfermedad, los especialistas mencionan al estrés como principal factor para ser susceptible de contagio. Respecto al vector que ocasiona está enfermedad, se ha mencionado a un grupo de peces denominados "mariposa", como vector biológico, o inclusive restos de plásticos que viajan a gran distancia y que pudieran llevar consigo colonias de bacterias de una región a otra. En la región del caribe mexicano, el estudio del impacto por residuos sólidos es cada vez más estudiada, no así el de los microplásticos la cual es incipiente. La generación de información a través de monitoreos estandarizados es una herramienta efectiva para el manejo de áreas protegidas. En 2019 se colabora y facilita el estudio de presencia de microplásticos ejecutado por Manta Caribbean Project, quienes tienen por objetivo la protección del hábitat de la Manta Gigante (Mobula birostris) a través de la colecta de muestras mediante arrastres con maya (luz de 335 µm), las cuales fueron procesadas posteriormente en laboratorio. Se presentaron tres tipos de partículas microplásticas: fragmentos, fibras y espuma. Los fragmentos fueron los más abundantes, seguido por las fibras y espuma. Se considera continuar el muestreo en 2021. La coordinación entre gobierno, organizaciones civiles y academia, como en este caso, ayudarán a implementar políticas públicas para la conservación basadas en monitoreos científicos.

Keywords : arrecifes , microplasticos , salud arrecifal

# First characterization of marine litter collected on Fort Dauphin and Sainte Marie Island, East Coast of Madagascar

Thibault Margot, Ramanampamonjy Aina, Saloma Anjara.

Madagascar has a high degree of endemism. This biodiversity is threatened by pollution, especially by marinelitter along the coasts. So far, no research has been carried out on marine pollution in this island. This study focuses on the first characterization of marinelitter on the east-coast of Madagascar in order to describe the quantity of marine and land-based inputs. Data were collected in October 2019. An accumulation survey of macro-litter (¿ 2.5 cm) was carried out for 10 consecutive days on inhabited and uninhabited sites in Ford-Dauphin (FD) and Sainte-Marie Island (SMI). The abundance of the marine litter per category was noted according to the SST manual and an origin determination was undertaken. A total of 4 005 pieces of marine debris were collected at FD (uninhabited site: 0.0173 item.m2.d1; inhabited site 0.0893 item.m2.d1) and 15 666 at SMI (uninhabited site: 0.225 item.m2. d1; inhabited site: 0.0699 item.m2. d1). Globally, the frequency of daily occurrence of stranded marine litter decreased for each site, however, the quantity varied inter day. Hard plastic was the main category found at the uninhabited sites for 50% (n=310) at FD and 76% (n=6,202) at SMI, mainly plastic fragments and bottles. A total of 111 brands were identified for SMI with the largest number being 54% of Indonesian origin (n=368) for the uninhabited site and 11% (n=85) of Malagasy origin for the inhabited site. This first study confirms that the majority of the marine waste stranded on the east coast of Madagascar comes from the East Indian Ocean via the securrent and to a lesser extent from Madagascar's land-based input. Light and durable, hard plastic can resist degradation and travel the ocean. To preserve Madagascar's biodiversity and human health, the implementation of a waste management program is more than a necessity.

Keywords : accumulation survey , beaches , Madagascar , Marine litter , sources

# Citizen science for microplastic detection and analysis -A comparison with other fields of science – research limits and possibilities in the field of microplastic

Cyvin Jakob, Lacerda Ana Luzia.

Citizen science is an important and powerfull way of gathering scientific data. This method found to contribute to the democratisation of academia, and awake the communities to environmental values, increasing their wish to take care of nature. There are another fields, such as ornitology, botany, ecotoxicology, and geology, among others, where citizen science is used not only to crowd source data, but also to let people do the first step of analysis, before reporting to their responsible science contact. Regarding to studies of macroplastics, beach surveys are common envolving citizens worldwide, and crowd sourcing of data is quite normal; this method is also used for microplastics, but to a limited degree, due the small size of these particles. In this study, by literature reviewing, we examined the possibilities of using citizen science for survey and analysis of microplastic compared to studies in another fields of science. We seek to learn from the ornitologists and ecotoxicologists. Their experiences are important when we conduct citizen science studies on microplastics. What kind of studies are so far conducted for microplastic analysis, and how to move this method of research forward regarding to microplastic surveying?

Keywords : citizen science , microplastic , research limits

# Session 26.6\_Ma. Chaired by Sabine Pahl, Plymouth

**Students' conceptions on the subject of microplastics** *Raab Patricia, Bogner Franz X.*. Paper number 334319

**Enabling individuals to understand, act on and reduce their microplastic footprint** *Winton Debbie, Loiselle Steven, Marazzi Luca.* Paper number 334363

**Public risk perception of microplastics** *Kramm Johanna, Völker Carolin, Werschmöller Simon.* Paper number 334481

**Plastic pollution: Overall knowledge, perceived impacts, and pro-environmental behaviours** *Soares Joana, Miguel Isabel, Venâncio Cátia, Lopes Isabel, Oliveira Miguel.* Paper number 334592

### Students' conceptions on the subject of microplastics

Raab Patricia, Bogner Franz X.

Microplastics are ubiquitous. Therefore, it is a highly topical research field due to the different impacts on ecosystems, which arise from them. Despite its enormous fundamental importance and the extensive media presence, microplastics are still too little present in the school context. Therefore, the BMBF project PLAWES surveyed university students on the topic of microplastics. In this study, we gained insights into students' conceptions on the term microplastics, possible sources in the household, risk assessment, potential consequences, and their sources of knowledge on the topic. For this purpose, 267 university students (56.6 % female) with an average age of 20.3 (SD =  $\pm$ 2.56) years responded to open and closed questions. These were subsequently analyzed in utilizing a qualitative content analysis. 78 % of the respondents classified the term microplastics with reference to small plastic particles. As sources of microplastics in the household, plastic packaging was mentioned by 43 %, followed by cosmetic products (28 %) and diverse plastic objects (19 %). Furthermore, in the context of microplastics, students showed a great threat awareness: 36 % classified microplastics as very dangerous, and 55 % as dangerous, but 19 % could not reason their assessment. Media is the most crucial information source: 51 % of the respondents cited different media channels (e.g., TV, internet, print media). More reliable sources of information such as schools (named by 21 % of the respondents) and projects (4 %) have less importance in the procurement of information. It is precisely this frequency pattern that shows the great need for action in schools to form students' conceptions in a well-founded way and to close knowledge gaps on the subject of microplastics.

Keywords : awareness of danger , conceptions , knowledge sources , microplastic sources household , microplastics , university students

# Enabling individuals to understand, act on and reduce their microplastic footprint

Winton Debbie, Loiselle Steven, Marazzi Luca.

The concept of a plastic footprint is one that has been relatively well developed in recent years, with plastic calculators now available to allow the public to understand their personal plastic consumption. Such calculators inform respondents about their annual plastic usage, their largest areas of consumption, how this compares to a national average, and the types of actions they can take to reduce their footprint. The Earthwatch Plastic Footprint Calculator also questions people about barriers to action, asking which of a pre-determined list of barriers have prevented them from reducing their use of that particular plastic item any further. The resulting data present interesting insights into how consumers can be supported to further reduce their plastic consumption, and who should help such efforts; businesses, government and at a community level. Our Microplastic Footprint Calculator is a new concept incorporating consumer microplastic consumption, whether intentional or unintentional. This will inform individuals of their plastic usage at the microplastic level, raise awareness of potential actions, and help us to understand barriers. There are many areas of daily life where consumers contribute to microplastic release into the environment, both within and outside of the home. Enough data already exist on microplastic outputs from activities such as driving, clothes washing and artificial pitch use to create such a calculator. Likewise, there are a multitude of actions available to reduce personal emissions from these sources, with new innovations being created every year. However, these actions do not tend to be common knowledge, highlighting both a need for awareness raising and for increased understanding of potential barriers to their use. This presentation will describe lessons learned from the macro Plastic Footprint Calculator and explore the requirements for an equivalent Microplastic Footprint Calculator.

Keywords : action , barriers , consumer , footprint calculator , microplastic , plastic footprint , reduction

### **Public risk perception of microplastics**

Kramm Johanna, Völker Carolin, Werschmöller Simon.

Research on the social dimension of plastics use, disposal and awareness is increasing. However, social research on microplastics perception is rare. While there are first exploratory surveys on how the public perceives microplastics (e.g. the Special Eurobarometer of the European Commission or the study of the German Federal Institute for Risk Assessment, BfR 2019) an in-depth study on the public risk perception of microplastics is still lacking. Thus, this paper takes up this research gap and explores the public risk perception of microplastics differentiated by socio-economic factors. The objectives of this article are (i) to investigate public risk perception and misconceptions of microplastics in order to inform future risk communication efforts on microplastics and (ii) to contribute to risk perception research of new and unknown risks. The paper is based on an online survey of 1.027 participants. The paper covers the following aspects: public knowledge about microplastics, public concerns about the effects of microplastics on the environment as well as on human health, perceptions on sources of microplastics and actions on microplastics.

Keywords : public understanding of microplastics , risk perceptions , survey

# Plastic pollution: Overall knowledge, perceived impacts, and pro-environmental behaviours

Soares Joana, Miguel Isabel, Venâncio Cátia, Lopes Isabel, Oliveira Miguel.

Plastic pollution is recognized as a global problem and one of the most challenging tasks for communities and governments. This environmental problem, that results from the vast applications of plastics in industrial and domestic products is associated with poor product management, from manufacturing processes to products' end life. In this perspective, human consumption and management of daily used plastic materials can play a determinant role to control of this environmental issue. In this perspective, understanding public perceptions about plastic pollution may be a valuable resource to engage society in solutions to reduce its environmental release. In this study, perceptions about plastic pollution, its impacts as well as sociodemographic and psychological factors predicting individuals' pro-environmental behaviours were analysed in Portugal. Overall, results showed knowledge that plastics degrade in the environment. Participants of the study perceived the bio-ecological impacts of plastics as a greater threat than the socioeconomic impacts. A hierarchical regression analyses revealed that sociodemographic and variables related to knowledge and perceived impacts about plastic pollution can predict participants' pro-environmental behaviours. Awareness about the impacts of plastic pollution (socioeconomic, health impacts and bio-ecological impacts), were highly associated to pro-environmental behaviour. This study data can help to understand how to enhance pro-environmental behaviours and contribute to decrease the presence of micro(nano)plastics in the environment.

Keywords : Ecological behaviour , Environmental awareness , Plastic pollution , Public perception

**Poster session 26.7** 

# Session 26.7\_O. Chaired by Maria Murcia, Menorca

# Effects of different microplastic polymers on the terrestrial woodlouse *Porcellio scaber* and the associated gut microbiome

Hink Linda, Holzinger Anja, Bernstein Alina, Sandfeld Jensen Tobias, Schramm Andreas, Feldhaar Heike, Horn Marcus A.. Paper number 334529

### MALDI mass spectrometry imaging in aquatic model systems

Schirmer Elisabeth, Treu Axel, Ritschar Sven, Schuster Stefan, Laforsch Christian, Römpp Andreas. Paper number 334287

# Contamination and effects of microplastics in the Seine Estuary's food web. First results from the Plastic-Seine research project

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# A European assessment of plastic ingestion in the Norway lobster (*Nephrops norvegicus*)

Carrassón Maite, Carreras-Colom Ester, Cartes Joan E., Rodríguez-Romeu Oriol, Constenla María, Welden Natalie, Soler-Membrives Anna.

Paper number 334327

### Ingestion of microplastics by fishes of an estuarine trophic chain in the Western Atlantic

Justino Anne, Lenoble Veronique, Pelage Latifa, Ferreira Guilherme, Passarone Rafaela, Frédou Thierry, Frédou Flávia.

Paper number 334367

# Microplastics in marine macrophytes on the underwater slope of the Sambian Peninsula (the Baltic Sea)

*Esiukova Elena, Lobchuk Olga, Volodina Aleksandra, Kupriyanova Anastasia, Chubarenko Irina.* Paper number 334304

# Effects of different microplastic polymers on the terrestrial woodlouse *Porcellio scaber* and the associated gut microbiome

Hink Linda, Holzinger Anja, Bernstein Alina, Sandfeld Jensen Tobias, Schramm Andreas, Feldhaar Heike, Horn Marcus A.

Slowly degrading microplastic (MP) is a gradually increasing pollutant in various ecosystems. To date, a wealth of information is available for aquatic ecosystems, while the attention to MP in terrestrial ecosystems is just increasing. Ubiquitous MP may enter food webs via unintentional ingestion by the soil macrofauna, but consequences are not well known. The effects on the macrofauna could be direct or indirect through changes of the gut microbiome altering the nutrient supply of the host. In this study, we assessed the effects of common MP polymers (PET, PLA and PS) on the fitness of the soil-dwelling woodlouse Porcellio scaber and the associated gut microbiome. During an eight-weeks feeding experiment, 10 adult woodlice were exposed to food pellets containing no, 2.5% or 5% MP. No significant effects on P. scaber's fitness (survival, reproduction, growth, weight and locomotive activity) were observed, but molecular analysis of the gut microbiota revealed significantly higher bacterial abundance for individuals fed with 5%-PLAfood. Radial microsensor pH, oxygen and hydrogen concentration profiles were recorded from guts of woodlice fed with 5%-MP-food. Anoxia and acidic pH were measured for all guts. Maximum gut hydrogen (produced during fermentation) concentrations indicated significantly more production in woodlice fed with PLA-food than without MP and lowest production in woodlice fed with PET- and PS-food. These results demonstrate that woodlice are a source of hydrogen in oxic environments, suggest a positive effect of PLA on the activity of the gut microbiome of P. scaber likely due to hydrolysis and fermentation of lactic acid, while PET and PS show negative effects on the fermentation. Future analysis of the short-chain organic acid composition, the microbial diversity and active taxa in the gut will allow a better understanding. Despite the effects on the microbiota, P. scaber is widely unaffected reflecting the resilience of adult woodlice.

Keywords : bacteria , feeding experiment , fermentation , fitness , microbial activity , microsensor profiles , PET , PLA , PS , soil macrofauna

# Effects of different microplastic polymers on the terrestrial woodlouse Porcellio scaber and the associated gut microbiome

[334529]

Linda Hink<sup>1</sup>, Anja Holzinger<sup>2</sup>, Alina Bernstein<sup>2</sup>, Tobias Sandfeld Jensen<sup>3</sup>, Andreas Schramm<sup>3</sup>, Heike Feldhaar<sup>2</sup> and Marcus A. Horn<sup>1</sup>

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### Introduction

- P. scaber is a widespread, important decomposer associated with nutrient cycling in soil
- Microplastic (MP) is an ubiquitous pollutant in terrestrial and many other ecosystems
- MP might enter food webs via unintentional ingestion and affects gut microbiomes (important for digestion)
- Assessment of MP effects on P. scaber warranted

### Hypothesis

Biodegradable (PLA) and non-biodegradable (PET and PS) MP have contrasting effects on P. scaber and its gut microbiome

### Aim of the study

To assess the effects of common MP polymers (PET, PLA and PS) on the fitness of the soil-dwelling woodlouse P. scaber and the associated gut microbiome

### Results





# Avoidance of PS





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### Summary and conclusions

- P. scaber is widely unaffected by MP reflecting resilience of adult woodlice
- Food containing PS is less preferable
- Woodlice are a source of hydrogen in oxic environments
- Positive effect of PLA on the activity of the gut microbiome likely due to hydrolysis and fermentation of lactic acid
- PET and PS ingestion leads to negative effects on fermentation

# **Microbial growth in the gut**



# Microbial activity in the gut

Microsensor measurements - Representative radial hydrogen profiles



nosition	max. hydrogen concentration (µM)				Gut embedded in
position	Control	5% PET	5% PLA	5% PS	agarose
anterior	4.6 (1.6) <sup>ab</sup>	3.4 (1.0) <sup>ab</sup>	6.7 (3.4) <sup>ab</sup>	4.0 (1.0) <sup>ab</sup>	Same and
median	7.9 (1.8) <sup>bc</sup>	2.6 (0.9) <sup>a</sup>	30.5 (3.7) <sup>d</sup>	5.0 (0.7) <sup>ab</sup>	ALL OF THE OWNER
posterior	17.2 (3.8) <sup>cd</sup>	5.1 (2.3) <sup>ab</sup>	24.0 (11.6) <sup>d</sup>	4.3 (2.7) <sup>ab</sup>	РМА

Further microsensor measurements: oxygen and pH

Anoxia and acidic pH (~5.6) determined for all guts

# **Coming soon...**

- Quantification of short chain fatty acids in the gut
- · Analysis of microbial community composition and transcriptionally active taxa in the gut



Funded by the Deutsche Forschungsgemeinschaft (DFG, German Research Foundation) Project Number 391977956 - SFB 1357



### MALDI mass spectrometry imaging in aquatic model systems

Schirmer Elisabeth, Treu Axel, Ritschar Sven, Schuster Stefan, Laforsch Christian, Römpp Andreas.

The pollution not only in marine but also in freshwater ecosystems rises due to the increasing amount of chemicals in use today. Many substances, such as bisphenol A, are lipid soluble and can thereby affect the lipid content and distribution in tissue. Because of severe consequences for animal and human health, there is a need to develop a method to investigate the influence of environmental contaminants on the lipid distribution of different organs. Here we show a MALDI mass spectrometry (MS) imaging method including a sample preparation protocol encompassing embedding medium and cryosectioning for two model organisms, zebrafish and daphnia. Due to using a high resolution MALDI imaging method the distribution of lipids in different brain regions as well as in non-neuronal tissues of the zebrafish could be acquired in both ionization polarities. Furthermore sample preparation for MS imaging of daphnia cryosections was developed for the first time. Several matrix compounds and solvents were compared to optimize ionization efficiency which resulted in improved signal intensities. In conclusion, the observed phospholipid signatures are an ideal basis for the analysis of changes in lipid pattern caused by (environmental) stress factors. This is the first step to investigate molecular effects of environmental pollutants in aquatic organisms.

Keywords : daphnia , lipid pattern , MALDI mass spectrometry imaging , zebrafish



# MALDI mass spectrometry imaging in aquatic model systems [334287]

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#### INTRODUCTION

The pollution not only in marine but also in freshwater ecosystems rises due to the increasing amount of chemicals in use today. Many substances, such as bisphenol A, are lipid soluble and can thereby affect the lipid content and distribution in tissue. Because of severe consequences for animal and human health, there is a need to develop a method to investigate the influence of environmental contaminants on the lipid distribution of different organs. MALDI imaging can provide detailed information on molecular changes in biological samples. For this reason, we develop a MALDI-MS imaging method for the aquatic model organisms zebrafish (*Danio rerio*) and waterflea (*Daphnia magna*). This is the first step for further studies on molecular effects of environmental pollutants in aquatic organisms.



#### **CONCLUSIONS AND OUTLOOK**

- Sample preparation workflow for MALDI imaging of zebrafish established.
- · First results for MS imaging of Daphnia magna.
- Detailed phospholipid signatures in wide variety of organs, specifically neural tissue
- Basis for the analysis of changes in lipid pattern caused by environmental contaminants.



Deutsche Forschungsgemeinschaft German Research Foundation

Funded by the Deutsche Forschungsgemeinschaft (DFG, German Research Foundation) – Project Number 391977956 – SFB 1357



# **Contamination and effects of microplastics in the Seine estuary's food web. First results from the Plastic-Seine research project**

J. Cachot, C. Clérandeau, F. Le Bihanic, C. Vignet, P. Pannetier, F. Misurale, Q. Pedriat, B. Morin, M. Revel, C. Mouneyrac, M. Mouloud, A. Châtel, H. Perrein-Ettajani, M. Bruneau, M.P. Halm, A. Gangnery, M.L. Bégout, X. Cousin, C. Dreanno, M. El Rakwe, J. Thery, C. Bialais, S. Souissi, M. Kazour, R. Amara, M.A. Dutertre, R. Coulaud, T. Monsinjon, B. Xuereb, L. Boudahmane, C. Partibane, B. Grassl, S. Lecomte, J. Gasperi.

The Plastic-Seine project (Seine-Aval, 2017-2020) aims to analyse the occurrence, levels of contamination and ecotoxicological effects of microplastics (MPs) in seven species (fishes, worms, copepods and mussels) of the Seine Estuary's ecosystem. MPs were recorded in the digestive tract and/or in the soft tissues of all the species sampled but the level of contamination was very variable depending on the species and the individual. Synthetic (mainly PET) or natural (cellulose) fibres constitute the vast majority of MPs found in biota. The kinetic of MPs ingestion and digestive transit were studied in the worm *Hediste diversicolor* and in sole, *Solea solea*. MPs ingestion is rapid (<1-2h) but the time to egestion depends on the species. The copepod *Eurytemora affinis* is also capable of ingesting and eliminating MPs of a few microns. These three species were exposed to ground samples of plastic litter collected in the Seine Estuary. These samples were composed of PE (62%), PP (28%) and PVC (5%) and contained high levels of PAHs, phthalates, alkyphenols and metals (Zn, Pb, Cr, Cd, etc.). Chronic exposure to these MPs led to no lethal toxicity but medium and long-term sublethal effects. Indeed, copepods exposed over three generations to 3 or 300 µg/L of MPs exhibited effects on the structure and dynamics of the population mainly on the 2nd or even the 3rd generation. Exposures of worms to a mixture of PP and PE MPs at high concentrations (10 and 50 mg/L) resulted in decreased coelomocyte viability but no significant effects observed on phagocytosis. Finally, exposure of juvenile soles to worms contaminated with 1 or 100/kg of sediment of a mixture of MPs led to a lightening of the skin colour and to a disrupted chromatic adaptation of MPs-exposed individuals. Sole larvae exposed to prev having ingested PVC particles coated with BaP showed, before metamorphosis, a slowdown in development and alterations of their swimming behaviour pattern.

Keywords : contamination , estuarine species , Microplastics , toxic effects , toxicokinetic



# Contamination and effects of microplastics in the Seine estuary's food web. First results from the Plastic-Seine research project (# 334343)



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#### Introduction:

The Seine River has an highly populated and industrial watershed (76.800 km<sup>2</sup>) and has been considered for decades as one of the most contaminated river and estuary in Europe (Cachot et al., 2006). Even though a lot of studies have reported organic or metallic contamination in this ecosystem very few reports focused on macro and microplastics (MPs).

The Plastic-Seine program (Flow and impacts of microplastics in the Seine estuary) is funded by Seine-Aval and CPIER Vallée de Seine. It involves six French laboratories associated in a joint research project to study in an integrated approach, the occurrence and levels of contamination of the Seine Estuary by microplastics in all compartments of the ecosystem including seven representative species of the food web and also possible impacts of MPs exposure on biology traits, physiology and behavior of three estuarine species, the worm Hediste diversicolor, the copepod Eurytemora affinis and the flatfish Solea solea.



Figure 1: Seine estuary (West of France), a 180 km long and

highly contaminated macr tidal estuary (Google Earth)

#### WP1: Occurrence of microplastics in abiotic compartments

See Gasperi et al., oral presentation # 333668. The fate of microplastics along salinity gradient and tidal cycles in a well-mixed estuary: a case study of the Seine estuary



#### WP2: Microplastic contamination in biota

Fig. 2: Sampling sites in the Seine estuary for seven representative estuarine species of the food web



Table 1: Suspected microplastics in gut or in soft tissue of 6 species collected in 2017 and 2018 in the Seine estuary

Species	Number of individual	% of contamination	Main type	Main type	Size range (μm)	Main polymers
Hediste diversicolor	120 pools of 3	61	Fibres and fragments	Black, blue or red	100-4000	PE + PP + PS
Palaemon longirostris	107 pools of 3	44.5	Fibres	Black or blue	100-6000	PS
Mytilus edulis	36 pools of 3	100	Fibres	Black	250-500	NI
Platichthys flesus	21	70-80	Fibres	Blue or red	< 800	PA + PET + PUR
Solea solea	101	80-98	Fibres	Black or blue	100-5000	NI
Dicentrarchus Iabrax	77	50	Fibres and fragments	Blue or red	100-2000	PET

polyethylene, PS: polystyrene, NI: non identified

Fig. 3: Main colours and polymers (FT-IR identification) of MPs isolated from gut of juveniles of Seabass Dicentrarchus labrax



> All species studied contained MPs but the occurrence varied widely between species and individuals. Fibers and fragments are the most abundant particles. Most of them are colored in blue/black or red.

> Several polymers were identified but PE and PET are the most abundant ones

WP3:Transfer and effect of MPs on three representative estuarine species : worms, copepods and soles



Fig. 4 : Experimental design. Exposure of copepods via the water column. Exposure of annelids via sediment. Sole larvae were fed with copepods and sole juveniles were fed with worms



polymers of MPs by post-larvae of Sole

> Efficient ingestion and egestion of MPs by worm and sole. Retention time of MPs in the gut of worms and soles is about 1-2h and 22-30h respectively.

No lethal effects of MPs on worms and sole. No effects on growth and metabolism in sole but significant effects on skin color and camouflage behavior.



Fig. 6 : Behavior of sole juveniles following 20 days of feeding with MP-contaminated worms.



Individuals/cohort (2L)

individuals/cohort (2L)

Fig. 7 : Chronic exposure to MPs of the copenod Eurytemora affinis over two generations using protocol of Souissi et al., (2016).

#### Conclusions:

V MPs were detected in all studied species of the food web (mussels, worms and fish) but with high variability in contamination between species and individuals.

mm

- V High efficiency of ingestion and egestion of MPs by worms and flatfish. Small MPs less than 10 μm can be ingested by copepods.
- ✓ No lethal effects on worms and soles but significant effects on skin color and behavior of soles.
- ✓ Effects of MPs on survival and development of copepod at the 2<sup>nd</sup> and 3<sup>rd</sup> generations.

Effects of MPs on development and survival of copepods at the 2<sup>nd</sup> and 3<sup>rd</sup> generation.









# A European assessment of plastic ingestion in the Norway lobster (*Nephrops norvegicus*)

Carrassón Maite, Carreras-Colom Ester, Cartes Joan E., Rodríguez-Romeu Oriol, Constenla María, Welden Natalie, Soler-Membrives Anna.

Several species among the marine organisms have been proposed as potential monitors of plastic pollution in the environment. Of these, Nephrops norvegicus, an abundant commercial species with a wide geographical distribution (from the NW Atlantic Coast to the E Mediterranean Sea) and for which varying levels of ingested plastics have already been reported at different sites (Murray and Cowie, 2011; Cau et al., 2019), seems of particular interest. In this study, we provide new data on plastic ingestion in N. norvegicus from several locations. Individuals were collected from the Clyde Sea, off Galicia, the Balearic Sea and the Gulf of Cadiz at depths ranging between 50 and 600 m. Stomach contents were screened for the presence of plastic items, which were characterized by means of light microscopy and a subsample analysed through FTIR for polymer identification. Plastics occurred in 75% of the individuals, commonly accumulating in the form of balls of tangled fibres (up to 36% of the individuals from the Clyde Sea) with the exception of the off Galicia area, where none of the individuals presented a ball. Almost all items identified were fibres of lengths ranging between 0.2 and 44.6 mm (films and fragments represented less than 1%) and a mean abundance of 7.5 items per individual overall. Despite showing a similar composition of polymers, significant differences in the mean fibre load and size range were observed between locations and even between different samplings performed at the same location. Since the diet composition of most of these populations, in general, has been considered similar (Cristo and Cartes, 1998) differences in the levels of ingested plastics could be mainly related to differences in the bioavailability of plastics in the environment. Overall, this study gives support to the potential use of N. norvegicus as a European monitor of plastic pollution.

Keywords : crustacean , monitor , Nephrops norvegicus , Plastic ingestion

# A EUROPEAN ASSESSMENT OF PLASTIC INCESTION IN THE NORWAY LOBSTER (NEPHROPS NORVEGICUS)

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#### INTRODUCTION

Several marine species have been proposed as potential monitors of plastic pollution in the environment. Of these, Nephrops norvegicus, an abundant commercial species with a wide geographical distribution and for which varying levels of ingested plastics have already been reported at different sites (Murray and Cowie, 2011; Cau et al., 2019), seems of particular interest. In this study, we provide new data on plastic ingestion in N. norvegicus from several locations across Europe with special emphasis on the characterization of fibres.

#### **MATERIALS & METHODS**





Fig. 1. Sampling locations (small turquoise dots) at each region and percentage of individuals showing fibres (lengths >5mm), tangled up fibres or fragments in their foregut content. \* Particular attention given to the Galicia sampling where no fibres >5mm

#### **CHARACTERIZATION & POLYMER IDENTIFICATION**



corresponding FTIR spectra. Spectral range: 3600-850 cm<sup>-1</sup>, Scale bars: black = 0.5 mm; white = 1 mm.

#### POLYMER COMPOSITION



Mediterranean Sea

stalan Department of Agriculture, Livestock, Fisheries and Food (European Martime and Fisheries Fund (EMFFI) project. **PLASMAR**' (RTI2018-094/806-B-100) and by ras-Colom benefits from an FPU grant (MICIL: FPU19/034/30). Our thanks especially to Dr. Angel Matea-Ramirez (ED-Centro Dreanográfico de Málaza) a the **ISUNEPCA17** cruise for providing the material from the Guif of Cádiz. is work was partially supported by the Spanish Ministry of Science, Innovation and Universities (MICIU) project "PLASMAR" (RTI2018 the Catalan Department of Agriculture, Livestock, Fisher and to the ISUNEPCA17 cruise for p

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#### **FIBRE ABUNDANCE & LOAD**



Fig. 3. Mean values of fibre abundance and total load of fibres (in mm) per individual and sampling location. Significant differences are depicted with letters. Two main groups can be defined according to the level of fibre abundance and load regardless of the region.

#### MAIN FINDINGS

- Prevalence of anthropogenic particles, particularly fibres, is extended across Europe populations of Nephrops norvegicus (Fig. 1).
  - Only one fragment observed in over 200 individuals inspected.
- Visual categorization coupled with polymer identification techniques (FTIR) (Fig. 2) can be a useful approach to characterize ingested fibres.
- Fibre abundance (in number) and fibre load (as the sum of lengths of all fibres) show equivalent results.
- In several comparisons, differences on fibre load and polymer composition between close samplings are greater than between distant samplings (e.g. Clyde Sea vs Mediterranean Sea) (Fig. 3 and 4) pointing out the potential importance of local sources of fibre pollution and/or the hydrodynamic conditions of the area.
- Since the diet composition of most of these populations, in general, has been considered similar (Cristo and Cartes, 1998), differences in the levels of ingested plastics could be mainly related to differences in the bioavailability of plastics in the environment.



UNB Unive



# **Ingestion of microplastics by fishes of an estuarine trophic chain in the Western Atlantic**

Justino Anne, Lenoble Veronique, Pelage Latifa, Ferreira Guilherme, Passarone Rafaela, Frédou Thierry, Frédou Flávia.

Marine ecosystems are reported to be contaminated by microplastics (¿ 5 mm), whereas the ecological mechanisms involved in the ingestion of debris by marine organisms are relatively unknown. This study explores an estuarine trophic chain, in a tropical ecosystem, aiming at understanding the patterns responsible for the different ingestion rates of plastic debris observed in three fish species (a predator and two of its main prey). A total of 82 fishes were collected through the local fishery, 30 individuals of Centropomus undecimalis (Piscivore), 21 of Bardiella ronchus (Zoobenthivore) and 31 of Gobionellus stomatus (Detritivore). For secure assessment of microplastic contamination in the digestive tracts of fishes, a digestion protocol was applied with the implementation of procedural blanks. Ingestion of microplastic differed significantly considering the trophic level. C. undecimalis is the most contaminated species  $(3.3 \pm 2.9 \text{ MPs fish-1})$  (77% FO), followed by G. stomatus  $(1.7 \pm 1.5 \text{ MPs fish-1})$  (74% FO) and B. ronchus  $(1.2 \pm 1.3 \text{ MPs fish-1})$ (67% FO). Additionally, the length of MPs ingested varied according to the species. G. stomatus (1.7  $\pm$  2.3 mm fish-1) ingested the longest MPs, followed by B. ronchus (0.8  $\pm$  0.7 mm fish-1) and C. undecimalis ( $0.5 \pm 0.6$  mm fish-1). Regarding the types of MPs ingested by fishes, most were fibres (47%), beads (40%) and fragments (13%), and varied between the species, C. undecimalis (68% beads, 28% fibres and 4% fragments), B. ronchus (23% beads, 62% fibres and 15% fragments), and G. stomatus (4% of beads, 71% of fibres and 25% of fragments). Our findings suggest that piscivores fish are more susceptible to be contaminated by microplastics since the ingestion rates increased with the trophic level. Our study also highlights a useful protocol of gut digestion applied for estuarine organisms, which can be replicated to other similar species and ecosystems.

Keywords : Extraction method , Plastic pollution , Trophic guild , Trophic transfer , Tropical estuary



# INGESTION OF MICROPLASTICS BY FISHES OF AN ESTUARINE TROPHIC CHAIN IN THE WESTERN ATLANTIC

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#### BACKGROUND

The whole marine ecosystems are reported to be contaminated by microplastics (MPs), whereas the ecological mechanisms involved in the ingestion of debris by marine organisms are relatively unknown. However, recent researches point out the trophic transfer (Fig.1) as a possible pathway to contaminate species at a high trophic level (Nelms et al., 2018; Ferreira et al., 2019).



Fig. 1. Trophic transfer scheme - Ingestion of contaminated prey by a high trophic level predator

This study explores an estuarine trophic chain, in a tropical ecosystem, aiming at understanding the patterns responsible for the different ingestion rates of plastic debris observed in three fish species (a predator and two of its main prey).

#### **METHODOLOGY**

Situated in a Marine Protected Area (MPA Santa Cruz), the Estuarine Complex of Santa Cruz Channel (Fig.2) is a typical model of an urban tropical estuary, which provide several ecosystems services, but is susceptible to anthropogenic impacts. A total of 82 fishes were collected through the local fishery, 30 individuals of *Centropomus undecimalis* (Piscivore), 21 of *Bairdiella ronchus* (Zoobenthivore) and 31 of *Gobionellus stomatus* (Detritivore). For a reliable assessment of microplastic contamination in the digestive tracts of fishes, a digestion protocol using NaOH (1M) was applied with the implementation of procedural blanks. We also followed the recent recommendations by Markic et al. (2019) and Hermsen et al. (2018) to avoid cross-contamination and minimize over/underestimation of microplastics samples.



Fig. 2. Map of the Estuarine Complex of the Santa Cruz Channel - Brazil
ACKNOWLEDGMENTS







#### **RESULTS**

• Ingestion of microplastic differed significantly depending on the trophic level. The predator, *C. undecimalis* was the most contaminated species (3.3 ± 2.9 MPs fish<sup>-1</sup>), followed by their two prey *G. stomatus* (1.7 ± 1.5 MPs fish<sup>-1</sup>) and *B. ronchus* (1.2 ± 1.3 MPs fish<sup>-1</sup>).



• Regarding the types of MPs ingested by fishes, most were fibres (47%), beads (40%) and fragments (13%), and varied between the species, *C. undecimalis* (68% beads, 28% fibres and 4% fragments), *B. ronchus* (23% beads, 62% fibres and 15% fragments), and *G. stomatus* (4% of beads, 71% of fibres and 25% of fragments).



The length of MPs ingested also varied according to the species. G. stomatus (1.7  $\pm$  2.3 mm fish<sup>-1</sup>) ingested the longest MPs, followed by B. ronchus (0.8  $\pm$  0.7 mm fish<sup>-1</sup>) and C. undecimalis (0.5  $\pm$  0.6 mm fish<sup>-1</sup>)

#### **CONCLUSION**

Our findings suggest that piscivores fish are more susceptible to be contaminated by microplastics since the ingestion rates increased with the trophic level. Our study also highlights a useful protocol of gut digestion applied for estuarine organisms, which can be replicated to other similar species and ecosystems.

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# Microplastics in marine macrophytes on the underwater slope of the Sambian Peninsula (the Baltic Sea)

Esiukova Elena, Lobchuk Olga, Volodina Aleksandra, Kupriyanova Anastasia, Chubarenko Irina.

Macroalgae and macrophytes are important as habitats and spawning sites for many species of fish and invertebrates living in the Baltic Sea. The goal of our investigation to check whether growing macrophytes also concentrate and retain plastics, particularly that of microplastic (MP, 0.2-5 mm here) size range. Three summer expeditions in the southeastern part of the Baltic Sea (supported by the Russian Science Foundation, grant No. 19-17-00041) were conducted (July 30, August 5 and 7, 2019) in sea coastal zone, where communities of attached macroalgae (Furcellaria lumbricalis, Coccotylus truncatus, Polysiphonia fucoides, Cladophora rupestris, etc.) are developed on underwater boulders. Sampling of macrophytes was performed: (i) directly from growing thickets on underwater slope; (ii) by the diver working from the boat, in shallow coastal waters (floating torn off filaments); (iii) from the beach. Along with sampling of growing algae (from area 25×25 cm2) on the depths from 3.0 to 8.6 m, (distance from the shore - from 60 to 850 m), a hand pump was used to sample 20-100 liters of seawater from both algae thicket and algae-free water in surroundings. MPs were found in all the collected samples. Analysis shows on average 1.7 (in the range 1.1-5.3) times higher MPs contamination in water samples taken from the algae thickets (0.7-9 items per liter) than in those taken from the plant-free areas nearby (0.3-5.9 items per liter). Number of MPs per unit area (total) is in the range of 48-3088 items per m2. Fibres are the prevalent type of MPs in water and seaweed. Plant thalli are entangled by fibres. The majority of microparticles are fibres, mainly colorless and blue, but also red, black, golden, and yellow. Filamentous seaweed (Polysiphonia fucoides, Cladophora rupestris, Cladophora glomerata, and etc.) collect more fibres than cartilaginous Furcellaria lumbricalis and Coccotylus truncatus.

Keywords : macrophytes , microplastics , sampling , the Baltic Sea , underwater slope



Abstract number 334304

#### Microplastics in marine macrophytes on the underwater slope of the Sambian peninsula (the Baltic Sea) E. Esiukova, O. Lobchuk, A. Volodina, A. Kupriyanova, I. Chubarenko



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Study area the southeastern Baltic Sea: off the Sambiar peninsula (nearby Cape Sampling . Taran). locations (8 stations) within the are rec square



Study area



Macroalgae and other macrophytes are important as habitats and spawning sites for many species of fish and invertebrates living in the Baltic Sea. The goal of our investigation to check whether growing macrophytes also concentrate and retain plastics, particularly that of microplastic (MP, 0.2-5 mm here) size range. Three summer expeditions in the southeastern part of the Baltic Sea (Fig. 1) were conducted (July 30, August 5 and 7, 2019) in sea coastal zone, where communities of attached macroalgae (Furcellaria lumbricalis, Coccotylus truncatus, Polysiphonia fucoides, Cladophora rupestris, etc. (Fig. 2)) are developed on (i) directly from growing thickets on underwater slope; (ii) by the diver working from the boat, in shallow coastal waters (floating torn off filaments); (iii) from the beach.

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Along with sampling of growing algae (from area 25×25 cm<sup>2</sup>) (*Fig. 3*) on the depths from 3.0 to 8.6 m, (distance from the shore from 60 to 850 m (Fig. 1)), a hand pump was used to sample 20-100 liters of seawater from both algae thicket and algae-free water in surroundings (*Fig. 3*).

MPs were found in all the collected samples (*Fig. 4*). Analysis shows on average 1.7 (in the range 1.1-5.3) times higher MPs contamination in water samples taken from the algae thickets (0.7-9 items per liter) than in those taken from the plant-free areas nearby (0.3-5) items per liter). Number of MPs per unit area (total) is in the range of 48-3088 items per m<sup>2</sup>. Fibres are the prevalent type of MPs in water and seaweed. Plant thalli are entangled by fibres. The majority of microparticles are fibres, mainly colorless and blue, but also red, black, golden, and yellow. Filamentous seaweed (Polysiphonia fuccides, Cladophora rupestris, Cladophora glomerata, and etc.) collect more fibres cartilaginous Furcellaria lumbricalis and Coccotylus truncatus.



Fig. 2. Algae of each species: Furcellaria lumbricalis (F. Iumbricalis), Coccotylus truncatus (C. truncatus), and Filamentous (Polysiphonia fucoides, Cladophora rupestris, Cladophora glomerata, and etc.) (Filamentous)

#### **Microplastics extraction**

Microplastics were extracted from the samples using the method employed by (Masura et al., 2015) with recommendations by (Zobkov and Esiukova, 2017a.b; Esiukova et al., 2020), and new modifications

in brief, it includes: wet peroxide oxidation (H<sub>2</sub>O<sub>2</sub> (30%) at 75 °C) → calcite fraction removal by HCl solution → filtering (100 µm)  $\rightarrow$  density separation (1.6 g mL<sup>-1</sup>) if there is sand matter filtering (100  $\mu$ m)  $\rightarrow$  examination under a stereomicroscope with the magnification from 10× to 40× directly on the surface of the filter according to (Norén, 2007)  $\rightarrow$  MPs identification with a Raman spectrometer.

The extracted microparticles were classified into three generic groups: fragments, films, and fibers according to (Chubarenko et al., 2018)



Fig. 3. Sampling of macrophytes was performed: (a) directly on underwater slope from growing thickets and algaefree water in surroundings (hand pump was used); (b) by the diver working from the boat, in shallow coastal waters (areas with filamentous algae (at depths of 3.2 and 4 m) and with perennial algae Furcellaria (depths of 5.6 and 8.2 m)



Fig. 5. Mass fraction of algae of each species (in percent of the total sample mass of algae at the station): Furcellaria lumbricalis (M\_F. lumbricalis), Coccotylus truncatus (M\_C. truncatus), and Filamentous (Polysiphonia fucoides, Cladophora rupestris, Cladophora glomerata, and etc.) (M\_Filamentous);



Fig. 8. Microplastics abundance (N/V, items per liter) and distribution inside algae thickets and outside of algae

References



Fig. 6. At each station, several samplings were performed: outside the algae and inside the algae thicket. Comparison between the number of microparticles (items per liter) in water outside of the algae and inside the algae thickets (with replicates)



Fig. 9. Microplastics abundance (N/S, items per m<sup>2</sup>) and distribution (in percent of the total number of microparticles) by three generic groups (fragments, films, and fibers).

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Fig. 4. Micro particles found in water: a) outside of algae, b) within algae thickets.



Fig. 7. Correlation between the number of microparticles (item per liter) and mass fraction of algae of each species (percent of the total mass of algae at the station)

#### Conclusions

- Marine macrophytes and water in-between them show high concentration of plastic particles. Marine macrophytes do retain microplastics
- Water within thickets is on total average 1.7 (average by station from 1.1 to 5.3) times more contaminated than water in neighboring areas, which are free of vegetation. Minimum / maximum abundance of MPs at the stations with vegetation is 0.7 / 9 items per liter, while in vegetationfree water - 0.3 / 5.9 items per liter.
- Fibres are the prevalent type of microplastics in water and within thickets, and their content is up to 3.8 fibres per liter vs 8.5 fibres per liter respectively

Plant thalli are entangled by fibers

- Number of MPs per unit area (total) is in the range of 48-3088 items per m<sup>2</sup>
- Filamentous seweed (Polysiphonia fucoides, Cladophora rupestris, Cladophora glomerata, and etc.) collect more fibers than Furcellaria lumbricalis and Coccotylus truncatus.

Investigations are supported by the Russian Science Foundation, grant No. 19-17-00041

Results



# Session 26.7\_Me. Chaired by Matthias Völkl, Bayreuth

Stable isotope determination and plastic ingestion in farmed Sparus aurata and Mytilus galloprovincialis in an integrated multi-trophic aquaculture systems Ripolles Vincent, Alomar Carmen, Compa Montserrat, Deudero Salud. Paper number 334388

# Occurrence of microplastics in the digestive tract of the European seabass (*Dicentrarchus labrax*) cultivated in the Canary Islands

Sánchez Almeida Raquel, Hernández-Sánchez Cintia, González-Sálamo Javier, Villanova-Solano Cristina, Hernández-Borges Javier. Paper number 334416

# Can a short-term exposure to microplastics affect the biochemical and metabolic responses of farmed meagre larvae (*Argyrosomus regius*; Asso, 1801)?

Campos Diana, Rodrigues Andreia C. M., Rocha Rui J. M., Martins Roberto, Candeias-Mendes Ana, Castanho Sara, Pousão-Ferreira Pedro, Soares Amadeu M. V. M., Patrício Silva Ana L.. Paper number 334533

**Solving a sticky situation: Microplastic analysis of lipid-rich tissue** *Dawson Amanda, Motti Cherie, Kroon Frederieke.* Paper number 334599

Chronic exposure to polystyrene microparticles induces changes in the proteome of *Daphnia magna* 

Trotter Benjamin, Wilde Lena, Brehm Julian, Arnold Georg J., Fröhlich Thomas, Laforsch Christian. Paper number 334247

# **Stable isotope determination and plastic ingestion in farmed** *Sparus aurata* **and** *Mytilus galloprovincialis* **in an integrated multi-trophic aquaculture systems** Ripolles Vincent, Alomar Carmen, Compa Montserrat, Deudero Salud.

Plastic debris are ubiquitous and are also found in marine aquaculture. Two key species, Sparus aurata (sea bream) and Mytilus galloprovincialis (Mediterranean mussel), were tested for four months to assess the effects of plastics derived from a multi-trophic aquaculture system integrated into the natural environment and these impacts on biological parameters (Condition index, Fullness index and size). For this, stable isotope analyses of carbon ( $\delta$ 12C and  $\delta$ 13C) and nitrogen ( $\delta$ 14N and  $\delta$ 15N) was performed. Additionally, gastrointestinal tracts of fish and soft tissues of mussel were digested with potassium hydroxide to analyse and quantify ingestion of plastics. For S. aurata, 24.44% individuals had ingested plastics with a total of 145 particles identified using visual sorting. Visually, 79.30% items identified were macrofilaments derived from aquacultures facilities. Sea bream of outdoor cage sites showed an average enrichment of 0.55‰ in δ13C and an average impoverishment of 0.56 % in  $\delta$ 15N compared to indoor treatment. For M. galloprovincialis, 89.52%individuals had ingested plastics with a total of 709 particles identified. 98.20% of plastic items identified were fibres. δ13C mean values in mussels' soft tissue were more negative in cages (-21.42  $\pm 0.38\%$ ) than in control sites, but differences were not significant.  $\delta 15N$  mean values were significantly higher in cages  $(6.01 \pm 0.32\%)$  than in control sites. Despite physical damages, no correlation between plastic ingestion and biological parameters was observed. No relationship was found between plastic ingestion and isotopic values in the two species. Isotopic differences were mainly due to different food sources with control sites and no to the plastic ingestion.

Keywords : Control index , Mediterranean aquaculture , Plastic ingestion , Stable isotopes



Ingestion of transparent microfibers and accumulation of microplastics observed
 CI and size → unaffected by ingestion of microplastics
 Under the influence of sea bream culture → filtration of organic waste

Acknowledgements : This study was funded by the Aquaculture derived plastics: Impacts and effects in marine food webs (ACUPLASTIC) project from the National Program of Investigation, Development and Innovation orientated towards challenges of society from the Spanish Government (grant number CTM2017-88332-R).

# Occurrence of microplastics in the digestive tract of the European seabass (*Dicentrarchus labrax*) cultivated in the Canary Islands

Sánchez Almeida Raquel, Hernández-Sánchez Cintia, González-Sálamo Javier, Villanova-Solano Cristina, Hernández-Borges Javier.

The European seabass is distributed in the eastern Atlantic, from Iceland and Norway to Senegal, including the Canary Islands, the Mediterranean and the Black Sea. It is a demersal species that inhabits coastal waters down to 100 m deep, in various types of estuary and lagoon bottoms, although in the northern zone they migrate to deeper waters of the high seas during winter. It is a species of great commercial interest that is currently produced around several hundred thousand tons per year. In fact, it is the most important and widely cultivated commercial fish in the Mediterranean whose main producers are Greece, Turkey, Spain, Croatia and Egypt. In Spain, the Canary Islands, is the second major producer. Microplastics have been observed in a wide variety of fish with considerable variability in levels of contamination in different species and geographic locations. Farmed fish are not exempt of this problem and their monitoring is of special interest since little is known concerning the accumulation of microplastics in fish from major fish farms and mariculture areas. In the present work we have studied the presence of microplastics particles in the digestive tracts of several specimens of the European seabass cultivated in fish farms located in Canarian waters. The specimens were bought in local markets to guarantee their origin. After the dissection and extraction of the digestive tracts, their content was digested with KOH and the content filtered. Microplastics were visualized under a stereomicroscope and classified by shape, size and colour. From the 45 examined specimens, 66.7 % presented microplastics, mainly fibres (94.6 %), with an average of 2.2 items per fish and blue as the predominant colour (45%).

Keywords : European sea bass , fibres , microplastics , sea farm , stomachs

### ANALYSIS OF OCCURRENCE OF MICROPLASTICS IN THE DIGESTIVE TRACT OF THE EUROPEAN SEABASS (DICENTRARCHUS LABRAX) CULTIVATED IN THE CANARY ISLANDS

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<sup>2</sup> Departs nto de O e, Spain. de La Lac



# **INTRODUCTION**

The European seabass is distributed in the eastern Atlantic, from Iceland and Norway to Senegal, including the Canary Islands, the Mediterranean and the Black Sea. It is a demersal species that inhabits coastal waters down to 100 m deep, in various types of estuary and lagoon bottoms, although in the northern zone they migrate to deeper waters of the high seas during winter. It is a species of great commercial interest that is currently produced around several hundred thousand tons per year. In fact, it is the most important and widely cultivated commercial fish in the Mediterranean whose main producers are Greece, Turkey, Spain, Croatia and Egypt, In Spain, the Canary Islands, is the second maior producer<sup>1</sup>

Microplastics have been observed in a wide variety of fish with considerable variability in levels of contamination in different species and geographic locations<sup>2</sup>. Farmed fish are not exempt of this problem and their monitoring is of special interest since little is known concerning the accumulation of microplastics in fish from major fish farms and mariculture areas.

In the present work we have studied the presence of microplastics particles in the digestive tracts of several specimens of the European seabass cultivated in fish farms located in Canarian waters. The specimens were bought in local markets to guarantee their origin. After the dissection and extraction of the digestive tracts, their content was digested with KOH and the content filtered. Microplastics were visualized under a stereomicroscope and classified by shape, size and colour. From the 45 examined specimens, 66.7 % presented microplastics, mainly fibres (94.6 %), with an average of 2.2 items per fish and blue as the predominant colour (45 %).

# EXPERIMENTAL



### **CONCLUSIONS**

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- > From the analysis of the 45 specimens of Dicentrarchus labrax, 66.7 % of the specimens presented microplastics in their digestive tracts, with an average of 2.2 items per fish.
- > Regarding morphological classification, only fibres (94.6 %) and fragments (5.4 %) were found in the analysed samples. The fibres had an average length of 2187 μm (range 337-7277 μm).
- > From the colours classification, it was found that most microplastics were blue (46 %), followed by black (33 %), although other colours were also found.
- > IR analyses is being carried out in order to determine the composition of the plastic particles found in Dicentrarchus labrax.

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# Can a short-term exposure to microplastics affect the biochemical and metabolic responses of farmed meagre larvae (*Argyrosomus regius*; Asso, 1801)?

Campos Diana, Rodrigues Andreia C. M., Rocha Rui J. M., Martins Roberto, Candeias-Mendes Ana, Castanho Sara, Pousão-Ferreira Pedro, Soares Amadeu M. V. M., Patrício Silva Ana L..

The ubiquitous presence and increasing concentrations of microplastics (; 5 mm in size), particularly in aquaculture facilities, raise concerns regarding its interaction with farmed organisms and potential contamination of human food supply. Adverse physiological and metabolic responses from microplastic ingestion in aquatic organisms have mostly been addressed under laboratory conditions after exposure to very high levels. Moreover, little is known considering farmed species. This study aimed at evaluating the short-term sub-lethal effects on the oxidative stress status, neurotoxicity and metabolic level of meagre fish larvae Argyrosomus regius (15 days post-hatching), caused by 3 h of exposure to polyethylene microplastics (PEMPs; tested concentrations: 0.1, 1 and 10 mg/L; n=16, in the presence or absence of food (alive Artemia nauplii, 5 org/mL). After 3 h exposure, fish larvae presented PEMPs in the gut:  $1 \pm 0.2$  to  $1.1 \pm 0.2$  and ; 1 to  $1.3 \pm 0.4$  particles/larvae in the absence and presence of food, respectively. In the absence of food, PEMP ingestion: (1) affected the antioxidant defences of meagre fish larvae through a generalised inhibition of the catalase activity, decrease in glutathione-S-transferases, and an increase of total glutathione in the medium and highest tested concentrations; without, however, cause oxidative damage (LPO); (2) inhibited acetylcholinesterase activity; (3) incremented the aerobic energy production. Such biochemical effects were similar in the presence of food, except for the aerobic energy production, which increased significantly in the lowest and medium tested PEMP concentrations. Although a short exposure to PEMPs in relevant aquaculture scenarios (presence of food) seems not to compromise fish larvae homeostasis, it cannot rule out potential adverse effects under a chronic exposure – with potential alterations on fish development, nutritional value and fitness.

Keywords : aerobic energy production , biomarkers , neurotoxicity , oxidative stress , Polyethylene









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FCT COMPETE 2020

#### Introduction

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- The presence of microplastics (< 5 mm in size) in aquaculture facilities is a reality, as most materials used to build and maintain the culture systems rely on plastics.</p>
- The ingestion of microplastics by organisms with different feeding-guilds seems to lead to impairments in their growth, development, and reproduction.
- Thus it is imperative to understand the accumulation and effects of microplastics in farmed fish since they are a food source to the humans.

Aim: Evaluation of the sub-lethal effects on the oxidative stress status, neurotoxicity and metabolic level of meagre fish larvae Argyrosomus regius after a short-term exposure to polyethylene microplastics (PE-MPs).



Fig. 1 – Effects of PE-MPs on feeding activity (A); aerobic energy production - via ETS (B); neurotransmission – via ChE activity (C); oxidative damage - via lipid peroxidation (LPO) (D); enzymatic antioxidante defences – via catalase activity (E); non-enzymatic antioxidante defences – via total glutathione levels (tGSH) (F); and detoxification mechanisms – via Glutathione-S-transferase activity (GST) (G) of *A. regius* larvae. The biochemical analyses (B-G) were performed in the presence and absence of food. All data is expressed as mean ± SEM. (F) P C 0.05 – Dunref's test against the negative control "O").

#### **CONCLUSIONS**

- The presence of PE-MPs inhibited the feeding activity of A. regius larvae; however, few particles of MPs were found inside the organisms.
- After short-term exposure to PE-MPs, larvae presented no oxidative damage, potentially prevented through an increment in aerobic energy production (ETS) and in nonenzymatic antioxidant defences (increase in tGSH levels) at exposure concentration ≥ 1 mg/L; enzymatic antioxidant capacity (CAT) was inhibited at ≥ 0.1 mg/L.
- All PE-MPs exposure concentrations caused an inhibition of ChE activity, indicating potential neurotoxicity.
- Results suggest that energy production through aerobic metabolism was required to overcome low/moderate levels of oxidative stress → but it do es not rule out potential long-term (cumulative) effects that could threatens the organisms' health and life traits, and compromise aquaculture efficiency.

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# Solving a sticky situation: Microplastic analysis of lipid-rich tissue

Dawson Amanda, Motti Cherie, Kroon Frederieke.

Given current concerns regarding the extent of microplastic contamination in the environment, routine monitoring for microplastics in biological tissues is becoming increasingly common place. However, complex sample matrices, such as lipid-rich tissues, require multiple pre-treatment steps which may lead to increased sample processing time and costs, and a reduction in microplastic recovery rates thereby hindering monitoring efforts. Lipid-rich (fat) tissues often pose difficulties for traditional potassium hydroxide (KOH) digestion methods due to saponification. This reaction produces a suspension of glycerol and fatty acids (soaps), which may entrap microplastics inhibiting their recovery and clog filters thus reducing the efficiency of the filtration or inhibiting it altogether. In this study, the incorporation of 100% ethanol (EtOH) to existing KOH digestion methods was found to completely redissolve the viscous saponified gel formed in these reactions, with a digestion efficiency greater than 97% for all treated lipid-rich tissue samples. Recovery of spiked polyethylene and polystyrene fragments, and rayon and polyester fibers, ranged from 93% to 100%. The addition of EtOH did not induce physical or chemical degradation on these polymers. The inclusion of an ad hoc decision-making tool within the digestion workflow reduced pre-processing time for samples and allowed for solid saponified samples to be completely redissolved. This validated workflow facilitates high through-put sampling of biota, by enabling lipid-rich tissues to be filtered with a high degree of efficiency thereby successfully separating microplastics from their gelatinous matrix.

Keywords : ethanol (EtOH) , fat deposit , fish , gel , methods , potassium hydroxide (KOH) , saponification

# Solving a Sticky Situation: Microplastic Analysis of Lipid-Rich Tissue

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# Adding Ethanol to Potassium hydroxide digestions reduces the viscosity of lipid- rich solutions aiding filtration

# Background

- Potassium hydroxide (KOH) digestions are used to separate MPs from tissue
- Many fish have high lipid content
- Lipid-rich tissue + aqueous 10% KOH = Saponification
- Heat and agitation increase the rate of saponification
- Once saponified, samples are often too viscous to pour and cannot be filtered

# This study developed a workflow to treat samples which have undergone saponification

### **Methods**

- Fish tissue is dissected (1,2)
- Tissue digestion using: 10% KOH 1:10 w/v, 40°C, 72h OR
  - 1:4 w/v, ~22°C (RT), 14d (3)
- Visual assessment of digesta for saponification (4)
- If saponification occurred, digesta is treated with 100% ethanol (EtOH) 1:10 v/v (EtOH:Sample) (5.a)
- If soap is completely liquefied, then digesta is filtered (6)
- If soap persists, more EtOH is added to reach 1:4 v/v (5.b)
- Repeat as necessary until soap is dispersed (5.b)
- Digesta is filtered sequentially over 547, 263 and 26 μm stainless steel filters to retrieve MPs (7)

#### **Questions? Contact Me**

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### Table 1. Effect of ethanol treatment on spiked MPs

Polymer	Recovery (%)	Spectral Match (%)	Carbonyl index	Physical Appearance
PE	93.3	98.6%	NC	NC
PES	100	98.0%	NC	NC
PS	100	99.3%	NC	NC
Rayon	93.3	97.1%	NC	Y- bleached*

NC – No Change; \*changes due to KOH exposure prior to adding EtO



Figure 1. Saponified *Lates calcifer* GIT, A) Untreated, B) Treated



Poster 334599

# Results

- All soaps formed through digestion were dispersed when treated with either 1:10 or 1:4 EtOH
- Untreated samples were unable to be filtered, due to complete saponification
- Digestion efficiency for EtOH treated samples ranged from 99-100%
- Spiked MPs were unchanged by EtOH treatment (Table 1)
- Treatment can be implemented ad hoc (only if saponification occurs)



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# Chronic exposure to polystyrene microparticles induces changes in the proteome of *Daphnia magna*

Trotter Benjamin, Wilde Lena, Brehm Julian, Arnold Georg J., Fröhlich Thomas, Laforsch Christian.

In the past years, the research focus on effects of microplastics (MP) on aquatic organisms shifted from marine systems towards freshwater systems, where microplastics occur in similar concentrations. An important freshwater model organism in the MP field is the cladoceran Daphnia. which inhibits a central role in ecosystems and has been established as a test organism in ecotoxicology. Chronic exposure of Daphnia magna (n = 30) to polystyrene (PS) microparticles led to a significant (p-value ; 0.001) reduction in body size and number of offspring. The aim of our study was to shed light on underlying molecular effects induced by microplastic ingestion in D. magna. Since proteins, e.g. enzymes are specifically relevant for the physiology of an organism, we assessed beside morphology and life history parameters, the effects of PS MP at the proteomic level. Animals (n = 30) from the same clone, cultured under the same condition served as controls. By the use of a sophisticated mass spectrometry-based approach, we were able to identify 28696 different peptides which could be assigned to 3784 different proteins. Using a bioinformatic workflow, customized for D. magna data, we found 44 proteins being significantly altered in abundance (FDR ; 0.05) in the PS treated samples. Among the proteins increased in the PS treated group were several transferases (e.g. GABA transaminase and different sulfotransferases), which are i.a. involved in catalyzing the degradation of neurotransmitters and survival of cells. In the downregulated group we found proteins connected to biotic- and inorganic stress and reproduction. Strikingly, we were able to identify four digestive enzymes which are significantly downregulated in the PS treated animals, indicating a limited nutrient uptake. This could also explain the significantly smaller body length and increased mortality of the treated daphnids, previously observed.

Keywords : Daphnia , mass spectrometry , Microplastics , polystyrene , proteomics



# Chronic exposure to polystyrene microparticles induces changes in the proteome of *Daphnia magna* [ID: 334247]

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#### INTRODUCTION

Worldwide production of synthetic polymers and the influx of inappropriately disposed plastic litter into ecosystems has grown exponentially since the 1980ies. Especially the abundance and effects of microplastics (MP, < 5 mm) on marine and freshwater ecosystems gained attention and is discussed controversially. Many recent studies show significant effects of MP on different fresh-water taxa, while others record no such effects.

The cladoceran *Daphnia* has been studied intensively as model organism, because it inhibits a central role in ecosystems and is shown to ingest MP of different morphology and polymer type. Some studies already showed substantial negative effects on daphnids. To unravel the underlying molecular mechanisms this work combines phenotypic analysis with sophisticated proteome analysis.

#### STUDY DESIGN





#### SUMMARY & DISCUSSION

- Chronic PS exposition of *D. magna* and *D. longicephala* resulted in morphological alterations and reduced numbers of offspring
- Whereas proteome alteration in *D. longicephala* were less pronounced, MP exposure induced significant alterations in the proteomes of *D. magna*
- Several sulfotransferases crucial for cell survival were found among the upregulated proteins

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- The enzyme with the strongest increase was (S)-3-amino-2methylpropionate transaminase, which is also known to react with GABA, a prominent inhibitory neurotransmitter
- Strikingly, several downregulated proteins are related to cellular stress
   and reproduction
- Four digestive enzymes were decreased, indicating a reduced nutrient uptake which may explain smaller body size





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Deutsche Forschungsgemeinschaft Geman Research Foundation



# Session 26.7\_Ma. Chaired by Mateo Cordier, Guyancourt

# Systematic identification of microplastics in abyssal and hadal sediments of the Kuril Kamchatka trench

Serena Abel, Sebastian Primpke, Ivo Int-Veen, Angelika Brandt, Gunnar Gerdts Paper number 337476

# Particle size distribution and shape identification of microplastics using ImageJ, Matlab and a self-developed Processing Tool

*Fritz Melanie, Pelikan Dominik, Albanna Mohammed, Hahn Barbara, Fischer Christian B.*. Paper number 334498

# What you net depends on if you grab: sampling method affects measured microplastic concentration

*Watkins Lisa, Sullivan Patrick J., Walter M. Todd.* Paper number 334600

**Combining microplastic surface sampling with manta trawl and microplastic ingestion in fish species to improve plastic assessment in the marine ecosystem: a case study in the Adriatic Sea.** *Giani Dario, Panti Cristina, Fossi Maria Cristina, Baini Matteo, Concato Margherita, Galli Matteo.* Paper number 334720

# Systematic identification of microplastics in abyssal and hadal sediments of the Kuril Kamchatka trench

Serena Abel, Sebastian Primpke, Ivo Int-Veen, Angelika Brandt, Gunnar Gerdts

The occurrence of microplastics throughout marine environments worldwide, from pelagic to benthic habitats, has become serious cause for concern. Hadal zones were recently described as the "trash bins of the oceans" and ultimate sink for marine plastic debris. The Kuril region covers a substantial area of the North Pacific Ocean and is characterised by high biological productivity, intense marine traffic through the Kuril straits, and anthropogenic activity. Moreover, strong tidal currents and eddy activity, as well as the influence of Pacific currents, have the potential for long distance transport and retention of microplastics in this area. To verify the hypothesis that the underlying Kuril Kamchatka Trench might accumulate microplastics from the surrounding environments and act as the final sink for high quantities of microplastics, we analysed eight sediment samples collected in the Kuril Kamchatka Trench at a depth range of 5143 to 8250 m during the Kuril Kamchatka Biodiversity Studies II (KuramBio II) expedition in summer 2016. Microplastics were characterised via Micro Fourier Transform Infrared spectroscopy. All samples were analysed in their entirety to avoid inaccuracies due to extrapolations of microplastic concentrations and polymer diversities, which would otherwise be based on commonly applied representative aliquots. The number of microplastic particles detected ranged from 14 to 209 kg-1 sediment (dry weight) with a total of 15 different plastic polymers detected. Polypropylene accounted for the largest proportion (33.2 %), followed by acrylates/polyurethane/varnish (19%) and oxidized polypropylene (17.4%). By comparing extrapolated sample aliquots with in toto results, it was shown that aliquot-based extrapolations lead to severe under- or overestimations of microplastic concentrations, and an underestimation of polymer diversity.

Keywords : Microplastic ; Deep ; sea sediment ; FTIR spectroscopy ; SiMPle
SENCKENBERG



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Abstract number: 337476

### introduction

occurrence of microplastics throughout marine environments worldwide, from pelagic to benthic habitats, has become serious cause for concern. Hadal zones were recently described as the "trash bins of the oceans" and ultimate sink for marine plastic debris. The Kuril region covers a substantial area of the North Pacific Ocean and is characterised by high biological productivity, intense marine traffic through the Kuril straits, and anthropogenic activity. Strong tidal currents and eddy activity, as well as the influence of Pacific currents, have the potential for long distance transport and retention of microplastics in this area. The Kuril Kamchatka Trench might accumulate microplastics from the surrounding environments and act as the final sink for high quantities of microplastics



### Oceanic trenches:

- The deepest environment on our planet (6000-10.989 m).
- Of the 20 major trenches, 17 are found in the Pacific basin.
- Relatively delicate ecosystems.
- Hotspot of biodiversity.

The Kuril Kamchatka trench:

- Located in the North-west Pacific Ocean.
- The trench formed as a result of the subduction zone, which formed in the late Cretaceous.
- The deepest point is at 10.542 m.
- The trench is 2250 km long
- The Kurile current (Oyashio) and Kuril counter current (Kuroshio), influence this area and may be responsible for long-distance litter transport.

The aim of this study was to quantify the MP present in the deep KKT, to characterise the MP particles by size and polymer type, and to contribute to a further understanding of MP accumulation in deep-sea trenches. To examine the potential inaccuracies of aliquot-based estimations, samples were both aliquot sub-sampling and in toto sample analysis were performed on each sample.



### Particle size distribution and shape identification of microplastics using ImageJ, Matlab and a self-developed Processing Tool

Fritz Melanie, Pelikan Dominik, Albanna Mohammed, Hahn Barbara, Fischer Christian B.

Besides the challenging extraction of microplastics from diverse matrices and their chemical analysis, it is also tricky to quantify particle sizes and shapes accurately. By checking particle size data of different studies, it becomes evidential that the data are hardly comparable. This is mainly due to the different techniques and modes of displaying particle size distributions (PSD), additionally to the non-conform sampling strategies and statistical variations. Drawbacks of the common methods are limits in sample numbers, analytical speed, complexity in handling, inefficiency or nonrepeatability by destructive methods. Furthermore, non-reliable information on shape by simplifying measurements applying the concept of equivalent spheres are inappropriate for irregular shaped particles as fibers. We developed an ImageJ plugin, a Matlab code and a self-written Processing tool to test best practice in particle analysis for a series of scanning electron microscope images of polyethylene particles (PE) and polyethylene terephthalate (PET) fibers. The prerequisite was to run large data sets at once in a short time. To consider the irregularity of particles, the reciprocal aspect ratio and feret major axis ratio was calculated. As reference, we used manually corrected images, whereby particle overlaps, artifacts or missing particle segments were elaborately processed for each image separately. For PET fibers, results show that Matlab and ImageJ have a significantly greater deviation than Processing Tool caused by conglomerations detecting fewer fibers than actually present. Interestingly the Matlab results for PE particles are much closer to the reference value than those generated with ImageJ or Processing Tool. We conclude that the appropriate software should be chosen depending on the application and the object(s) to be viewed, because considerable variations may occur based on the specifics of the programs itself. Nevertheless, PSDs and shapes for the present microplastics can be determined for all three methods quickly and accurately in good agreement.

Keywords : counting , fibers , image analysis , irregular particles , quantification , shape , size

### **MICR02020**

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### Particle size distribution and shape identification of microplastics using ImageJ, Matlab and a self-developed Processing Tool

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### What you net depends on if you grab: sampling method affects measured microplastic concentration

Watkins Lisa, Sullivan Patrick J., Walter M. Todd.

Methods used to quantify microplastics are diverse and oftentimes incompatible. In particular, factors that in theory should be unrelated to measured microplastic concentration, such as sampled volume and sampling device used, are in fact strongly correlated with it. We confirmed this by performing a meta-analysis of 100 surface water microplastic studies, including those that pair grab and net samples during collection. We then used the unexpected relationship between sampled volume, sampling device used and concentration measured to itemize and quantify potential explanations for the systematic discrepancies observed.

Keywords : freshwater , grab , marine , metaanalysis , method , microplastics , net , surface water



Cornell Water Lab

### Combining microplastic surface sampling with manta trawl and microplastic ingestion in fish species to improve plastic assessment in the marine ecosystem: a case study in the Adriatic Sea

Giani Dario, Panti Cristina, Fossi Maria Cristina, Baini Matteo, Concato Margherita, Galli Matteo.

Microplastics distributed along the water column with different patterns of accumulation. The monitoring of microplastics in the environment can lead to over or underestimation of the risk posed to marine species. Within the project AdriCleanFish, aimed to investigate the occurrence of plastics in the environment and the impacts on fish species, the data obtained from surface water sampling with manta trawl (mesh 0.3 mm) were compared with microplastics isolated from the gastrointestinal tract of 6 different fish species; 3 pelagic species (Engraulis encrasicolus, Sardina pilchardus, Trachurus trachurus) and 3 demersal species (Merluccius merluccius, Mullus barbatus, Solea solea) from two different sampling site in the Adriatic Sea. The two types of sampling were carried out in the same area thanks to the collaboration with professional fishermen. The microplastics were sorted and characterized by size class and, comparing the manta samples with microplastics detected in fish species, the environmental microplastics with dimensions between 0.3 and 0.5 mm were not found in any sample, while microplastics belonging to this size class was identified in fish species. It emerged that surface microplastics and particles isolated from pelagic species are similar in size, between 1 and 5 mm, while smaller microplastics were isolated mostly in demersal organisms. The same comparison was made considering the different shapes of microplastics. Fibers are mostly found in demersal fish species, on the other hand they are the least frequently found with manta sampling. These differences underline the importance of the simultaneous sampling of fish species belonging to different habitats combined with sea surface sampling, as they can represent comprehensively the contamination of microplastics in the same study area and the related effect on marine organisms.

Keywords : microplastics/ manta trawl/ microplastic ingestion



### Combining microplastic surface sampling with manta trawl and microplastic ingestion in fish species to improve plastic assessment in the marine ecosystem: a case study in the Adriatic Sea



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### **Introduction**

Plastic pollution in the ocean represent one of the biggest worldwide environmental topic, the distribution of plastic particles in the marine environment and in biota is the main issue of several studies (Lusher et al., 2015; Auta et al., 2017; Avio et al., 2020). Microplastics distributed along the water column with different patterns of accumulation. The monitoring of microplastics in the environment can lead to over or underestimation of the risk posed to marine species.

### Materials and methods

Within the project AdriCleanFish, aimed to investigate the occurrence of plastics in the environment and the impacts on fish species, the data obtained from surface water sampling with manta trawl (mesh 0.3 mm) were compared with microplastics isolated from the gastrointestinal tract of 6 different fish species; 3 pelagic species (*Engraulis encrasicolus, Sardina pilchardus, Trachurus trachurus*) and 3 demersal species (*Merluccius merluccius, Mullus barbatus, Solea solea*) from two different sampling site in the Adriatic Sea (Chioggia and Civitanova Marche). The two types of sampling were carried out in the same area thanks to the collaboration with professional fishermen (Fig. 1).



### **Results and discussion**

The microplastics were sorted and characterized by size class and, comparing the manta samples with microplastics detected in fish species, the environmental microplastics with dimensions between 0.3 and 0.5 mm were not found in any surface water sample, while microplastics belonging to this size class was identified in fish species (Fig. 2A;2B;2C). It emerged that surface microplastics and particles isolated from pelagic species are similar in size, between 1 and 5 mm, while smaller microplastics were isolated mostly in demersal organisms (Fig. 2D). The isolation method used for fish species has also made it possible to investigate the abundance of microplastics less than 0.3 mm in size (limit of detection for surface sampling with manta trawl), which represent 8% of the microplastics ingested (Fig. 2C). The same comparison was made considering the different shapes of microplastics (Fig. 3A;3B;3C). Fibers are mostly found in demersal fish species (Fig. 3C); on the other hand they are the least frequently found with manta sampling (Fig. 3A;3B).



### Conclusion

These differences underline the importance of the simultaneous sampling of fish species belonging to different habitats combined with sea surface sampling, as they can represent comprehensively the contamination of microplastics in the same study area and the related effect on marine organisms.

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**Poster session 26.8** 

### Session 26.8\_O. Chaired by Eva Cardona, Menorca

### Neustonic marine microplastics and zooplankton in the coastal waters of Cabrera MPA (North-Western Mediterranean Sea)

fagiano valentina, alomar carme, compa ferrer montserrat, mateu-vicens guillem, deudero salud. Paper number 334376

**Presence of microplastics in stingrays (Hypanus guttatus) from Brazilian Amazon Coast** Pegado Tamyris, Brabo Lúcio, Sarti Francesco, Gava Thaís, Nunes Jorge, Chelazzi David, Cincinelli Alessandra, Monteiro Raqueline, Martinelli Filho José Eduardo, Giarrizzo Tommaso. Paper number 334400

### Uptake, distribution and excretion of microplastic fibres in the green sea urchin: an experimental exposure

Abrahams Alexandra K., Bourgeon Sophie, Sørensen Lisbet, Herzke Dorte, Booth Andy, Halsband Claudia. Paper number 334471

### Microplastics as a Trojan horse of pathogens and antimicrobial resistance (BACT&PLAST Project)

Balleste Elisenda, Sanchez-Vidal Anna, Lucena Francisco. Paper number 333664

### Neustonic marine microplastics and zooplankton in the coastal waters of Cabrera MPA (North-Western Mediterranean Sea)

Fagiano Valentina, Alomar Carme, Compa Ferrer Montserrat, Mateu-Vicens Guillem, Deudero Salud.

Currently, microplastics (; 5 mm) are ubiquitous in the marine environments posing a growing treat to marine protected areas (MPAs) created to prevent habitat lost and to preserves biodiversity. Within the effects of microplastics (MP) on biota it is of special interest to highlight their effects on zooplanktonic organisms having a key role on local food web structures. As to which are the effects of MP abundances on zooplanktonic community assemblage and composition, particularly in coastal ecosystems, it is still an open question. The present study addresses MP distribution and composition within Cabrera MPA (north-western Mediterranean Sea) as well its effects on local zooplanktonic community assemblage. MP average abundances found within Cabrera MPA  $[0.35 (\pm 0.29)$ items/m3] showed higher values than those reported for the majority of western Mediterranean Sea basins and for the closest more anthropized area of Mallorca  $[0.16 (\pm 0.29) \text{ items/m3}]$ . A micro-scale accumulation area was detected within Cabrera MPA presenting an MP abundance [19.32  $\pm$  (14.45) items/m3] higher than those reported for the subtropical gyres by previous studies. MP composition suggested far contamination sources as predominant. Moreover, MP appeared able to influence the local zooplanktonic community assemblage without altering the zooplankton average abundance. An interesting positive correlation was found between MP and the abundance of the planktonic stage (Tretomphalus) of the foraminifer Rosalina globularis. This species seems able to utilize MP for its dispersion, while a negative correlation with Copepoda abundance was detected suggesting a role of this taxa in MP removal from surface waters. This work highlights an important and complex interaction between zooplankton and MP, being able to mutually influence their distribution and composition. Moreover, results confirm that MPAs are not protected from MP pollution.

Keywords : Balearic , Microplastic: zooplankton ratios , Microplastics , MPAs , Zooplankton

# NEUSTONIC MARINE MICROPLASTICS AND ZOOPLANKTON IN THE COASTAL WATERS OF CABRERA MPA (NW



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As to which are the effects of MPs abundances on special interest to highlight their effects on zooplanktonic Within the effects of microplastics (MPs) on biota it is of local zooplanktonic community assemblage composition within Cabrera MPA as well its influence on particularly in coastal ecosystems, it is still an open question zooplanktonic community assemblage and composition, organisms having a key role on local food web structures $^{(1)}$ The present study addresses MPs distribution and





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MPs average abundances found within items/m3]. anthropized area of Mallorca [0.16 ( $\pm$  0.29] those detected Cabrera MPA showed higher values than at the closest more

- MPs contamination sources as predominant composition suggested far
- MPs appeared able to influence the local zooplanktonic community assemblage with
- A positive correlation was found between no effects on its global abundance.
- a negative correlation with Copepoda able to utilize MPs for its dispersion, while MPs and Tretomphalus. This species seems
- waters of this taxa in MPs removal from surface abundance was detected suggesting a role
- interaction between zooplankton and MP, being

that

plastics

Б

- This work highlights an important and complex

### **Presence of microplastics in stingrays (***Hypanus guttatus***) from Brazilian Amazon Coast**

Pegado Tamyris, Brabo Lúcio, Sarti Francesco, Gava Thaís, Nunes Jorge, Chelazzi David, Cincinelli Alessandra, Monteiro Raqueline, Martinelli Filho José Eduardo, Giarrizzo Tommaso.

Is known that microplastics (MPs) are widely distributed in the aquatic environment, reaching even the remotest areas of the oceans. Given their small size and abundance, MPs are easily ingested by a wide range of organisms during their normal feeding activities. These particles can be considered as a potential exposure route to persistent organic pollutants (POP's) and metals. In view of this scenario, we aimed to documents the ingestion of microplastics by Longnose stingrays in the Western Atlantic Ocean. We examined 23 specimens of Hypanus guttatus from the Brazilian Amazon coast and found microplastic particles in the stomach contents of almost a third of the individuals. Fibers were the most frequent item (82%), blue was the most frequent color (47%) and Polyethylene terephthalate (PET) was the most frequent polymer recorded (35%), as identified by 2D imaging -Fourier Transform Infrared (FTIR). The ingestion of microplastics has not been recorded previously in H. guttatus. The findings of the present study thus provide an important baseline for future studies of microplastic ingestion by dasyatid rays and other batoid species in the Atlantic Ocean, and contribute to the broader understanding of the spatial and temporal dimensions of the growing problem of plastic pollution in aquatic ecosystems and organisms.

Keywords : 2D FTIR Imaging , Elasmobranchii , Longnose stingray , Plastic pollution



### Presence of microplastics in stingrays (*Hypannus guttatus*) from Brazilian Amazon Coast



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### Introduction

- Microplastics  $\rightarrow$  size <5 mm;

- Widely distributed along aquatic environments;

- Easily ingested by a wide rang of organisms;

- Potential exposure route to persistent organic pollutants (POP's) and metals;

- Ingestion by elasmobranchs has fewer records compared to bony fish;

- *Hypannys guttatus* is an important stingray consumed by Latin American populations.

- We aimed document the ingestion of microplastics by Longnose stingrays *Hypannys guttatus* in the Western Atlantic Ocean.

### **Material and Methods**

We examined the stomach contents of 23 *Hypanus guttatus* (Fig.1) that inhabits the complex estuarine located in the southern extreme of the Brazilian Amazonian coast, more precisely in Maranhão Gulf. The specimens were captured by local fishers' longlines and gillnets between 2018 and 2019. Microplastics were categorized by shape and color, and the polymers were identified by 2D imaging - Fourier Transform Infrared (FTIR).



Fig.1: An specien of *Hypannus guttatus* captured in the southern extreme of the Brazilian Amazon coast.

### **Results and Discussion**

17 microplastics found in 7 of 23 specimens (FO% = 30,4);

Could be explained by the foraging strategy of the species/generalist top predator.

Fibers were the most frequent item (82%) (Fig 2);

Blue was the most frequent color (47%);

Most abundant microplastic category in marine environments;

Proximity to areas of urban development;

Oceanographic phenomena (e.g. macro-tidal currents) may contribute to the ample dispersal of microplastic.



Fig.2: Category most frequent in the stomachs of Longnose stingrays (*H. guttatus*) captured in southern extreme of the Brazilian Amazon coast.

Polyethylene terephthalate (PET) was the most frequent polymer recorded (35%) (Fig 3).

Most produced worldwide;

Dense polymer, sink and hits the bottom where the stingrays feeds.



PET PP PA ABS PET+SBR PE

Fig.3: Percentage of polymers found in the stomachs of Longnose stingrays (*H. guttatus*) captured in southern extreme of the Brazilian Amazon coast.

Conclusions

- First record of microplastic ingestion by *H. guttatus*, providing baseline values for future studies approaching rays in Atlantic Ocean;

- Contribute to the broader understanding of the growing problem of plastic pollution in aquatic ecosystems and organisms.

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### Uptake, distribution and excretion of microplastic fibres in the green sea urchin: an experimental exposure

Abrahams Alexandra K., Bourgeon Sophie, Sørensen Lisbet, Herzke Dorte, Booth Andy, Halsband Claudia.

Synthetic microplastic fibres (MPFs) from textiles constitute a major proportion of microplastic pollution in marine systems, having been frequently reported in all environmental compartments, including the digestive systems of many types of biota. Despite their widespread distribution, the direct interactions of MPFs with biota remain poorly understood. This study compares ingestion and egestion of acrylic fibres in the green sea urchin Strongylocentrotus droebachiensis. For reference, natural wool fibres were also studied. In addition to 'pristine' acrylic and wool fibres, 'aged' fibres exposed to natural seawater over two weeks were also used to study the influence of biofouling on ingestion rates. Fibres of 1-5 mm length were dispersed in the incubation seawater at concentrations of approximately 43 fibres per mL and sea urchins were then added and exposed individually for 48 hours. All fibre types (wool fouled and unfouled; acrylic fouled and unfouled) were readily ingested and subsequently egested with the faeces. No negative effects of fibre ingestion on the urchins were recorded. High numbers of fibres were counted in faecal pellets, suggesting that the fibres pass through the gastrointestinal tract and leave the organisms packaged in faecal material. The implications of biofouling on fibre uptake rates in benthic grazers and the fate of microplastic fibres upon repackaging in faecal matter are discussed.

Keywords : acrylic , biofouling , egestion , faecal matter , ingestion , microplastic fibres , sea urchin

# Uptake, distribution and excretion of microplastic fibres in green sea urchin

## Strongylocentrotus droebachiensis: an experimental exposure

Alexandra Abrahams<sup>a,»</sup>, Sophie Bourgeon<sup>»</sup>, Lisbet Sørensen<sup>e</sup>, Dorte Herzke<sup>a</sup>, Andy M. Booth<sup>e</sup>, Claudia Halsband Son GES

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### BACKGROUND

abundant polymers in the ocean Microplastic (MP) fibres, mainly derived from laundry of synthetic textiles, are

Yet, more research is needed on the fate of MP and different polymer types under different environmental conditions<sup>2,3</sup>

This study investigates the fate of MP fibres in the Arctic benthic environment



igure 1: S. droebachiens collected from Kvaløya, Norw

droebachiensis of natural wool fibre and a synthetic acrylic fibre, and investigate influence of biofouling STUDY AIM: To investigate ingestion, retention time and egestion in sea urchin Strongylocentrotus



and 134 hours, and all remaining individuals dissected at 134 hours. Remaining individuals relocated into new beakers with fresh seawater for depuration. Fecal pellets were collected at 72was 48 hours, then fecal pellets collected, and three individuals from each treatment were dissected for gut contents urchins were exposed individually to each fibre types, in addition to six controls without fibres. Exposure time to fibres -igure 2: Study design with two fibre types (natural wool and acrylic) in two states (clean and biofouled). Six adult sea



(orange; no analysis) water at in situ temperature, c) sea urchin diameter measurement , d) dissection, e) dissected sea urchin with gonads Figure 3: Experimental set up. a) measuring aliquots of seawater containing fibres, b) beakers incubated in running sea

### PRELIMINARY RESULTS

- counts and lowest variation between Clean acrylic treatment has lowest fibre analysed, and fibres have been found in So far, 25% of fecal pellets from each aims to be completed by May 2021 Labwork is currently underway. The study individuals fecal pellets of all treatments (fig 4 & 5) individual (48 hour exposure) have been
- The number of biofouled acrylic fibers per Both clean and biofouled wool have large pellet is higher than that of non-biofouled
- wool has highest median count variation in fibre count per individual. Clear
- provide information on retention time Gut contents is awaiting analysis and will

Marine Environment: A Global Assessment v GESAMP\_microplastics%20full%20study.pdf IUCN.CH.2017.01.en REFERENCES <sup>1</sup> Boucher, J., & Friot, D. (2017). Primary Microplastics in the Ocean: A Global Evaluation of Sources. doi:dx.doi.org/10.2305/ sustainability assessment. Sci Total Environ, <sup>1</sup> Henry, B., Laitala, K., & Klepp, I. G. (2019). Microfibres from apparel and home textiles: Pros ent. Sci Total Environ, 652, 483-494. doi:10.1016/j.scitotenv.2018.10.166 <sup>3</sup> GESAMP. (2015).

### OUTLOOK

- Occurence of fibres packed in fecal material in all treatments demonstrates ingestion and excretion of fibres through the gastrointestinal tract
- Biofouling possibly promotes ingestion of acrylic fibres, but not wool fibres.
- analysis of gut content. and/or longer retention time will be revealed by egestion between fiber types is due to lower uptake Whether the observed differences on fecal



fibres (red arrows), c) dissected fecal pellet with fibres separated (red arrows) Figure 4: a) fecal pellets stored in ethanol until analysis b) intact fecal pe



GESAMP NO. 90). http rospects for including microplastics in en Sources, Fate and Effects of Micropla. :s in the 'Plastic in the Arctic"

Hopland Sperre for technical assistance. ACKNOWLEDGEMENTS Thanks to Kristine This research is funded by The FRAM Centre Flagship

### Microplastics as a Trojan horse of pathogens and antimicrobial resistance (BACT&PLAST Project)

Balleste Elisenda, Sanchez-Vidal Anna, Lucena Francisco.

Microplastics (MP) are already detected in all the environments becoming a global concern. They are quickly colonized by microorganisms forming a biofilm. The health risks associated to MP are still unknow but debated. One of the suggested risks it is the potential of biofilms on MP to harbor and disseminate pathogens. Some studies have already detected pathogens on MP. Besides, crowded biofilms are also a hotspot of horizontal gene transfer i.e. antibiotic resistance genes (ARGs). The transmission of waterborne pathogens and ARGs is a public health concern and one of the WHO urgencies. Wastewater treatment plants are one of the main sources of MPs, faecal bacteria and ARG in the environment, where they are in contact. Thus, faecal bacteria can get attached to the MP and reach the aquatic ecosystems in a more protected structure than as free bacteria. Bact&Plast aims to link these 3 different elements to evaluate the potential of microplastics to shelter and disseminate waterborne pathogens and ARGs. Bact&Plast combines an experimental approach using mesocosmos with environmental monitoring. First, we will evaluate the bacterial colonization of MP inquiring differences between plastic polymers and environmental conditions as the main driver of this process. To achieve this objective, we will use mesocosomos in controlled conditions and the evolution of the biofilm will be followed using massive sequencing to determine the bacterial diversity and functional diversity. A similar approach will be used to detect the potential of faecal bacteria to get attached on the biofilms. For this purpose, mesocosms will be performed with treated sewage diluted in freshwater and seawater. Faecal indicator bacteria (FIB), potential opportunistic pathogens and ARGs will be followed in the biofilm-MPs and water. Finally, the obtained data will be compared with MPs from environmental samples: sewage effluent, river water and sediments and sea water and sediments.

Keywords : Antibiotic Resistance Genes , Bacteria , Biofilm , Microplastics , Pathogens

### Microplastics as a Trojan horse of pathogens and BARCELONA antimicrobial resistance - BACT&PLAST - Num 333664



### Elisenda Ballesté<sup>1</sup>, Anna Sanchez-Vidal<sup>2</sup>, Francisco Lucena<sup>1</sup>

<sup>1</sup>Dept. de Genètica, Microbiologia i Estadística. Universitat de Barcelona. <sup>2</sup>Dept. de Dinàmica de la Terra i de l'Oceà. Universitat de Barcelona. eballeste@ub.edu

- Microplastics (MP) are detected in all the environments becoming a global Besides, crowded biofilms are also a hotspot of horizontal gene transfer concern.
  - They are quickly colonized by microorganisms forming a biofilm.
  - The health risks associated to MP are still unknown but debated.
  - One of the suggested risks is the potential of biofilms on MP to harbor and disseminate pathogens.
- i.e. antibiotic resistance genes (ARGs).
- Wastewater treatment plants are one of the main sources of MPs, faecal bacteria and ARG in the environment, where they are in contact.
- Thus, faecal bacteria can get attached to the MP and reach the aquatic ecosystems in a more protected structure than as free bacteria.

Bact&Plast aims to link MP, sewage and biofilms to evaluate the potential of microplastics to shelter and disseminate waterborne pathogens and ARGs.



Microbial community analysis Pathogen screening

ARG monitoring





MARS

### WP1. Evaluation the settlement of bacterial biofilms on microplastics

### Task 1. Mesocosmos experiment

**Differences between polymers** 



Mesocosmos will be kept for one month and samples will be collected periodically.

Task 2. Characterization of the bacterial communities of the plastisphere

- · Epifluorescence microscopy after DAPI staining.
- Scanning electronic microscopy to observe the structure of the biofilm.
- · Follow-up of the colonisation process by molecular analysis: 16S rRNA massive sequencing.

### Task 3. Characterization of the resistome of the plastisphere

- · Shot-gun metagenomics
- Identification of antibiotic resistance genes
- Specific resistant genes will be quantified using qPCR

### WP 2. Assess the potential of the plastic biofilm to act as a shelter for pathogens and ARGs

### Task 4. Sewage Mesocosmos experiment



### Task 5. Monitoring of faecal indicator bacteria, environmental waterborne pathogens and ARGs

- Culture methods: E. coli (ISO 9308-1), Enteroccocci (ISO 7899-2:2000), Pseudomonas spp (Pseudomonas agar base), Vibrio spp. (TCBS), Total bacteria
- **qPCR:** *E. coli* and Enterococci and ARGs: β-lactamases, aminoglycosides, methicillin, quinolones, sulfonamides and tetracyclines

### Task 6. Characterization of the bacterial communities of the plastisphere in a faecally polluted environment

• Massive sequencing of the 16S rRNA gene

### WP 3. Analysis of bacterial communities of MP collected from the environment



Task 8. Characterization of the microbial community of the plastisphere from environmental samples

- **gPCR:** *E. coli* and Enterococci and ARGs: β-lactamases, aminoglycosides, methicillin, quinolones, sulfonamides and tetracyclines
- Massive sequencing of the 16S rRNA gene
- Shot-gun metagenomics

### Task 9. Identification of the plastic polymers

Microplastic particles will be chemically identified using the Fourier transformed infrared spectrometer (FTIR).

### Session 26.8\_Me. Chaired by Miguel Tamayo, Madrid

### Micro- and nanoplastic exposure effects in microalgae: a meta-analysis of standard growth inhibition tests

*Reichelt Sophia, Gorokhova Elena.* Paper number 332718

### Trout gut cells as a model to understand the effects of polystyrene nanoplastics on the gut immune function

*García Ordóñez Marlid, Solà i De Dios Roger, Brandts Irene, Teles Mariana, Roher Nerea.* Paper number 334390

### Chemical digestion methods: what are the real impacts on microplastics?

Gulizia Alexandra, Brodie Eve, Santana Marina, Bloom Sarah, Corbett Tayla, Daunmuller Renee, Motti Cherie, Vamvounis George. Paper number 334454

### Impact of plastic nanoparticles on marine and freshwater algae: *Thalassiosira weissflogii* and *Desmodesmus subspicatus*

*Feurtet-Mazel Agnès, Gonzalez Patrice, Braat Jeanne, Gaubert Joy, Gigault Julien, Latchere Oïhana, Châtel Amélie, Baudrimont Magalie.* Paper number 334432

### Micro- and nanoplastic exposure effects in microalgae: a meta-analysis of standard growth inhibition tests

Reichelt Sophia, Gorokhova Elena.

The effects of micro-and nanoplastics (MNP) on aquatic life have been subject to a vivid discussion about their environmental impacts. Algae are the base of the aquatic food-web and therefore highly relevant for the ecosystem stability. Yet, the reported outcomes of MNP are adverse, ranging from inhibition to stimulation of growth. Therefore, synthesis of available data is needed for reliable risk assessment. We performed a meta-analysis study to assess the effect of MNP exposure on algal growth. Twenty studies published between 2010 and 2020 and representing 16 algal species and five polymer materials administered as particles in size range 0.04 to 3000  $\mu$ m were included in this meta-analysis. A random-effect model was used to estimate the effect size in three datasets: (1) Low concentration range

Keywords : analysis , growth , meta , microalgae , microplastics , nanoplastics , particulate matter , risk assessment



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2018

Rogstad et al., 2019 Su et al., 2019 Zhao et al., 2019 Gorokhova et al., 2020 Cwart et al. 2020

Published: 08/2020 in Frontiers in Environmental Science Reichelt and Gorokhova et al., 2020 Nano- and microplastic exposure effects in algae: a meta-analysis of standard growth inhibition tests

### Trout gut cells as a model to understand the effects of polystyrene nanoplastics on the gut immune function

García Ordóñez Marlid, Solà i De Dios Roger, Brandts Irene, Teles Mariana, Roher Nerea.

The increasing and ubiquitous presence of nanoplastics (NPs) in the aquatic environment demands urgent assessment in order to understand their impact on ecosystems and human health. An important open question is how NPs exposure would interfere with the normal function of the immune system of aquatic organisms. In this context, we aim to understand whether the gut immune homeostasis could be disturbed by NPs exposure. Our previous studies using zebrafish as a model demonstrated that polystyrene nanoplastics (PS-NPs) were efficiently taken up by zebrafish liver (ZFL) cells tending to accumulate in lysosomes, possibly in an attempt of the cell machinery to metabolize PS-NPs. We also observed a modulation of the expression of relevant immune genes in ZFL cells, consecutively exposed to PS-NPs and Poly-(I:C), a viral-like stimulus. In this study, we evaluate the effect of PS-NPs (45 nm) on the immune response of RTgutGC cells (trout gut cells), since fish gastrointestinal epithelium is one of the principal portals of entry for both NPs and pathogens in teleosts. Our results show that: first, PS-NPs do not exert any cytotoxicity on RTgutGC cells; second, PS-NPs are efficiently taken up and accumulate in lysosomes, modulating the number and size of these organelles; third, PS-NPs alone barely affect expression of immune representative genes but trigger a synergic immune response to Poly-(I:C) (viral-like challenge) and LPS (bacterial-like challenge). Hence, these results point out the existence of altered cellular and molecular events that could have a relevant impact on gut mucosal immune response.

Keywords : immune response , lysosomes , nanoplastics , RTgutGC

### Trout gut cells...



### as a model to understand the effects of polystyrene nanoplastics on gut immune function

García, M.; Solà, R.; Brandts, I.; Teles, M.; Roher, N.

November 2020

The increasing and ubiquitous presence of nanoplastics (NPs) in the aquatic environment demands urgent assessment in order to understand their impact on ecosystems and human health. An important open question is how the exposure to NPs might interfere with the normal function of the immune system of aquatic organisms. Our previous study using zebrafish as a model demonstrated that polystyrene NPs (PS-NPs) were efficiently taken up by zebrafish liver cells (ZFL) accumulating in lysosomes, possibly in an attempt of the cell machinery to metabolize PS-NPs.

In this study, we aim to understand whether the gut immune homeostasis could be disturbed by exposure to NPs. Hence we evaluate the effect of PS-NPs on the immune response of RTgutGC cells, since fish gastrointestinal epithelium is one of the principal portals of entry for both NPs and pathogens in teleosts.

### Methodology



### . Results

### Uptake

Flow cytometry analysis showed a dose-dependent uptake of PS-NPs by RTqutGC cells after 24 h of exposure. (Fig.1). Confocal microscopy confirmed cellular accu colocalization in lysosomes (Fig.2). of PS-NPs and sh Contro 25 ug/ml PS-NPs



Fig.1: Flow cytometry analysis (mean fluorescence intensity in RTgutGC).

### Gene expression

0

Conclusions

Exposure of RTgutGC cells to PS-NPs (up to 50 mg/l) did not induce expression of proinflammatory genes. Nevertheless, when we co-exposed cells to PS-NPs and a viral (poly(I:C)) or bacterial challenge (LPS), this triggered a synergistic immune response, with the up-regulation of the expression of antiviral genes or pro-inflammatory cytokines (Fig.3)

63



There is a dose-dependent uptake and accumulation of PS-NPs in RTgutGC cells.

63x zoor Fig.2: Confocal images of RTgutGC cells. Cells were stained with Lysotracker, Hoechst and CellMask. Fluorescent PS-NPs incorporate Dragon Green as a dye.

63)

Cytotoxicity

The exposure to PS-NPs did not cause toxicity to RTgutGC cells, after 24h incubation for the testes concentrations (Fig.4).

Fig.4: Viability of RTgutGC cells assessed with the MTT assay



Immune stimulation 4

Nuc

### Bibliography

- 1.
- PS-NPs are not cytotoxic to RTgutGC cells at concentrations of up to 50 mg/l.
  As in ZFL, in RTgutGC cells PS-NPs colocalized in lysosomes.
  Co-exposure of RTgutGC cells to PS-NPs and PAMPs (Poly(I:C)) or LPS) triggers a synergistic response, modulating the expression of pro-inflammatory cytokines and antiviral genes.
- Brandts, L. et al. (2020) 'Polystyrene nanoplastics accumulate in ZFL cell lysosomes and in zebrafish larvae after acute exposure, inducing a synergistic immune response in vitro without affecting larval survival in vivo', Environmental Science: Nano. Royal Society of Chemistry, 7(8), pp. 2410–2422. doi: 10.1039/d0en00553c.
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- з.

### Chemical digestion methods: what are the real impacts on microplastics?

Gulizia Alexandra, Brodie Eve, Santana Marina, Bloom Sarah, Corbett Tayla, Daunmuller Renee, Motti Cherie, Vamvounis George.

Field collection and laboratory-controlled exposure studies are critical to establishing the toxicity and long-term consequences of aquatic plastic debris and their associated chemical contaminants (e.g. plasticisers) on organisms. A critical aspect of these studies relates to the separation and retrieval of plastics from biological sample matrices using a variety of extraction techniques. Popular separation methods for micro and nano-size plastics involve prolonged, high temperature treatment with a range of alkali, oxidative and/or acidic chemical reagents (e.g. 60oC with nitric acid). While these techniques offer a more efficient and robust means of sample clarification, their strongly acidic and oxidative properties have been associated with physical damages, enhanced polymer reactivity and fragment deterioration. Considering many identification and characterisation techniques postrecovery involve qualitative visual assessment and comparative spectroscopy, it is imperative that the chemical and physical characteristics of the plastic polymers are not impacted during chemical digestion. Here, we apply an analytical approach to characterise and quantify the reactivity and degradative impacts of common chemical digestion methods on polystyrene-based microplastics, chosen because of their high prevalence in aquatic environments globally. Based on these results, chemical digestion methods will be ranked to determine their appropriateness for microplastic separation, and recommendations for updates to future protocols will be offered.

Keywords : chemical digestion , extraction methods , microplastics , polymer degradation



## hemical digestion methods: What are the impacts on microplastics?

<u>Alexandra M. Gulizia</u> ª.<sup>b</sup>, Eve Brodie ª, Marina MF. Santana <sup>a.b.c</sup>, Sarah B. Bloom ª, Renee Daunmuller ª, Tayla Corbett <sup>a</sup>, Cherie A. Motti <sup>a,b,c</sup>, George Vamvounis\* <sup>a,b,c</sup>

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Australian Institute of Marine Science (AIMS), Australia



### 1. Introduction

conditions which can damage the plastic polymer. biomass, these methods often utilise harsh chemical and exposure studies. However, to efficiently remove and quantified from animal biota during field collection Chemical digestion methods allow plastics to be extracted



plastics in the environment, suitable chemical digestion To ensure accurate quantification and classification of methods need to be implemented 3.com; favpng.com; sci360.org

### 2. Methods

were exposed to the following digestion conditions in the Prepared polystyrene microplastics (300 µm - 1 mm) tahle helow

	$H_2O_2$
12, 24,	NaOH
1	КОН
Time (ho	Reagent
	<b>Time (h</b> c 12, 24,

or george.vamvounis@jcu.edu.au For more information contact: <u>alexandra.gulizia@my.jcu.edu.au</u>

### □ Treatments with HNO<sub>3</sub> resulted in a significant decrease in polymer chain length (e.g. molecular weight), changes to signature peak 3. Results and Discussion

environments in the FT-IR spectra and severe mechanical damages indicating polymer degradation 1200 6



**Molecular Weight** 

(g/mol)



Figure 1. Chemical and physical analyses revealed (a) polymer degradation, (b) infrared spectral dissimilarity and (c) fragment discolouration and deep surface cracking associated with high temperature  $HNO_3$  treatments

**Digestion Treatment** 

hr

HNO3

75

There was significant swelling and pitting after all 90°C KOH, NaOH and H<sub>2</sub>O<sub>2</sub> digestions. This indicated that microplastics may be

more sensitive to digestion conditions given their manufacturing origins and/or environmental histories



Figure 2. Microplastics swollen and pitted after 48-hour,  $90^{\circ}C$  digestions with (a) KOH, (b) NaOH and (c)  $H_2O_2$  at 2.5X magnification

### 4. Conclusions

□ Surface morphology may impact fragment reactivity during digestion

High temperatures digestions are associated with enhanced reagent reactivity, polymer and fragment degradation

HNO<sub>3</sub> digestions above 60°C may not be suitable for microplastic extractions

This research was made possible by the Australian Government RTP Scholarship and Stipend, AIMS@JCU and the JCU AAC

### Impact of plastic nanoparticles on marine and freshwater algae: *Thalassiosira* weissflogii and *Desmodesmus subspicatus*

Feurtet-Mazel Agnès, Gonzalez Patrice, Braat Jeanne, Gaubert Joy, Gigault Julien, Latchere Oïhana, Châtel Amélie, Baudrimont Magalie.

Every year, 5 to 10 million tons of plastic are dumped into the oceans and accumulate in huge ocean gyres. Freshwaters are the main transportation route for plastics to the oceans. This plastic debris undergo very strong physico-chemical stresses under the effect of various biotic and abiotic factors, and thus fragment. The fragmented macroscopic plastics lead to the formation of «microplastics» (between 20  $\mu$ m and 5 mm), and also «nanoplastics» (between 1 and 1000 nm). These nanoplastics are all the more dangerous as they can penetrate the food chains from the very first links: the primary producers. As part of the TROPHIPLAST project, this study aims to investigate the impact of nanoplastics from the Garonne river on two primary producers: Desmodesmus subspicatus, a freshwater green microalgae, and Thalassiosira weissflogii, a marine diatom. Both species were exposed for up to 96 h to 0.1, 1, 10, 100  $\mu$ /L of nanoplastics obtained from plastics samples of Garonne river. The results show that at these concentrations, there is no significant effect of nanoplastics on microalgae growth. However, there are some variations in the expression of photosynthesis genes for Thalassiosira weissflogii.

Keywords : freshwater and marine microalgae , growth effects , impacts , nanoplastics





Every year, 5 to 10 million tons of plastic are dumped into the oceans and

This plastic debris undergo very strong physico-chemical stresses under the

The fragmented macroscopic plastics lead to the formation of «microplastics»

These nanoplastics (NPs) can penetrate the food chains from the very first links: the primary producers and may be the cause of deleterious effects from

As part of the TROPHIPLAST project\*, this study aims to investigate the effects

and impact of nanoplastics from the Garonne River (SW, France) on two primary producers: Desmodesmus subspicatus, a freshwater green microalgae,

Freshwaters are the main transportation route for plastics to the oceans.

effect of various biotic and abiotic factors, and thus fragment.

(20 µm-5 mm), and also «nanoplastics» (1-1000 nm).









### Impact of plastic nanoparticles on marine and freshwater algae: Thalassiosira weissflogii and Desmodesmus subspicatus

Agnès Feurtet-Mazel<sup>1</sup>, Patrice Gonzalez<sup>1</sup>, Jeanne Braat<sup>1</sup>, Joy Gaubert<sup>1</sup>, Julien Gigault<sup>2</sup>, Oïhana Latchere<sup>3</sup>, Amélie Châtel<sup>3</sup>, Magalie Baudrimont<sup>1</sup>

### **Objectives**

the first trophic level.

accumulate in huge ocean gyres.

### Methodology

### **Exposure conditions:**

- ✓ Environmental NPs (99% <40 nm, PP, PS)</p>
- ✓ [NPs] tested: 0 0,1 1 10 100  $\mu$ g/L (same range than environmental concentrations in oceans: 8 pg/L to 0,1 mg/L in hot spots)
- Exposure duration: 96h (orbital shaker)

### **Biological criteria:**

- ✓ Gene expression by real time qPCR

Algal growth

- **Optical density**
- Cellular enumeration on counting cell





### **Results and**

and Thalassiosira weissflogii, a marine diatom.



### Coulter counter and OD overestimate algal densities, but the growth tendancies stays similar. Direct numeration of algae on counting cells

seems to be the best to correctly estimate algal densities.

### Gene expression levels in T. weissflogii





Functions	Response to oxidative stress		Photosynthesis		Mitochondrial metabolism			Detoxification
Genes	sodMn	sodFe	psaA	d1	cox1	nd5	125	mdr1
24h								
18h – 0,1 μg/L			2	3				
)6h – 100 μg/L			2	5,5				2,5

Over the NPs range tested:

- Induction of genes involved in photosynthesis (d1 and psaA) only after 48h at 0,1  $\mu g/L$  and after 96h at 100  $\mu g/L$  Moderate toxic effetcts only detectable at 100  $\mu g/L$  at 96h with overexpression of
- efflux mechanisms (mdr1)
- No differential expression detected at 24h
- Over-expression of genes involved in photosynthesis revealed an increased energy demand probably link to NPs.

### Conclusion

For both species, no significant effect of environmental NPs (PP, PS) collected in the Garonne River was observed on algal growth exposed to environmental concentrations (0,1 to 100 µg/L). The induction of genes involved in photosynthesis (d1 and psaA) has been reported, suggesting an increase of the energy demand in the algae and consequently an increase of the ATP production mediated by photosynthesis. This increase in energy demand could propably be linked to the effects of NPs or more probably to the contaminants carried by NPs. This hypothesis is emphasized by the induction of detoxification mechanisms for the highest dose at 96h. However, effects of NPs seems to be limited since no modification of algae growth has been observed.

Complementary experiments are planned in the Trophiplast project, to investigate the potentiality of NPs to transfer to higher trophic levels (e.g. bivalves) and the possible indirect impacts in relation with the ingestion of algae previoulsly exposed to NPs.

\* TROPHIPLAST project: « Micro- and nanoplastic particules toxicity on aquatic organisms along a freshwater-marine continuum : evaluation of a trophic transfer », supported by ANSES 2018-2022



Multisizer 4<sup>e</sup> Coulter counter

Thlassiosir weissflogi

### Session 26.8\_Ma. Chaired by Mateo Cordier, Guyancourt

A deep learning-based algorithm to automatically identify fluorescently stained MP *Kvæstad Bjarne, Farkas Julia, Krause Dan, Aas Marianne, Andy Booth.* Paper number 334491

**Qualitative analysis of the ecocorona on plastic surfaces** *Rynek Robby, Lechtenfeld Oliver, Reemtsma Thorsten, Wagner Stephan.* Paper number 334320

**Density Separation of Microplastics from solid sample matrices** *Schütze Berit, Thomas Daniela, Brunotte Habil Joachim.* Paper number 335550

**Strategies for the separation of microplastics from water via density modification** *Martínez De Pedro Zahara, Munoz Macarena, Ortiz David, Casas Jose A.*. Paper number 334335

### A deep learning-based algorithm to automatically identify fluorescently stained MP

Kvæstad Bjarne, Farkas Julia, Krause Dan, Aas Marianne, Andy Booth.

There are multiple approaches available for identifying and quantifying the number of microplastic (MP) particles present in environmental samples. The application of the different techniques depends on multiple factors including instrument purchase and running costs, instrumental availability, ease of use, time for analysis, parameter(s) of interest and quality of the final data. Fluorescent staining and imaging of MP particles is a method that is cheap to implement, easy to conduct and which offers high potential for rapid screening of large numbers of samples. While there are clear and acknowledged limitations with respect to some of the other methods and analysis systems (e.g. lack of specificity in identification, false positives, sensitivity), one major issue preventing the wider acceptance of this approach as a screening technique is a lack of robust and reproducible automation in the processing of images containing fluorescent MP on filters. Manually analysing filters with stained MP is not only a tedious and time-consuming task, but it can be very inaccurate and suffers with reproducibility issues both within an individual lab and across labs. To improve accuracy and reproducibility, whilst reducing time and cost of analysis, we have developed an algorithm based on deep learning to automatically identify the stained MP. The algorithm consists of a custom Fully Convolutional Network (FCN) architecture for particle segmentation and classical image processing for acquiring metrics including individual particle area, length and width of each particle. This algorithm can process data from one filter (consisting of ten 50-megapixel images) in less than one minute, completely unattended.

Keywords : Machine Learning , Microplastics , Quantifying

### A deep learning-based algorithm to automatically identify fluorescently stained MP

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### Introduction

There are multiple approaches available for identifying and quantifying the number of microplastic (MP) particles present in environmental samples. Fluorescent staining and imaging of MP particles is one method that is easy to implement and offers high potential for rapid screening of large numbers of samples. However, manually analysing filters with stained MP is not only a tedious and time-consuming task, but it can be very inaccurate and suffers with reproducibility issues, we aim to solve these issues using state-of-art deep learning techniques.

### **Materials & Methods**

The MP imaging rig is comprised of a macro lens attached to a 54-megapixel camera, which is mounted on a computer controlled desktop-sized CNC (Computer Numerical Control) machine. This setup automatically images the entire 41-mm diameter filter with 10 photographs at a resolution of approximately 2 µm per pixel. The lens includes an orange filter (529 nm wavelength) while the imaging stage is illuminated with two blue/green light sources with a peak wavelength of 475 nm. For image processing we have developed an algorithm based on deep learning to automatically identify the stained MP. The algorithm consists of a custom Fully Convolutional Network (FCN, Fig. 1) architecture for particle segmentation and can automatically adapt to image input sizes of 128x128 pixels and above.



Figure 1. illustrates the neural net architecture used in this method, the network consists of only convolution and max pool layers.

The output is a binary soft mask of size N-128xM-128 of a NxM input image, where the pixel values in the output mask can be any value between zero to one, depending on the confidence of the MP detections. Using a FCN architecture simplifies the training set since the adaptive input size makes it possible to train on a set of small images of 128x128 pixels, with the annotation value being either 1 (MP) or 0 (background) per image.

### Results

When processing full size images (> 128 pixels), the network will automatically scale to create a output image containing the segmentation values for the detected MPs. By thresholding this output image, we can filter the detected MP particles on confidence from the neural net (Fig. 2).

Input image

Output image



Figure 2. Image from the imaging equipment (left) and the neural net output (right) showing the detected MPs (white).

Using techniques such as Topological Structural Analysis (Suzuki, 1985) and ellipse fitting, we can acquire metrics like area, length and width for all detected particles. This algorithm can process ten partial images representing the total area of a single filter in less than one minute. When testing this system in a ongoing EU Joint Research Centre interlaboratory comparison test, we scored in the middle of the expected range.

### Conclusion

The system is comprised of cheap of the shelf hardware compared to more advanced systems, combined with fast image acquisition and processing the system offers consistent and accurate results for rapid screening of MP extracted from environmental samples, which is also time and cost effective compared to manual analysis.



References



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### Qualitative analysis of the ecocorona on plastic surfaces

Rynek Robby, Lechtenfeld Oliver, Reemtsma Thorsten, Wagner Stephan.

As microplastic particles enter the aquatic environment natural organic matter (NOM) molecules attach to the plastic surface first. These attached organic molecules are termed ecocorona. In a second stage microorganisms may attach at the plastic surface which is covered to some extend with the ecocorona and form a biofilm. Previously, it has been demonstrated that biofilm evolution is strongly depending on the surface chemistry of plastic particles (Lorite et al., 2011). These findings imply that the chemical composition of the ecocorona may vary and it may be dependent on the polymer type, the weathering stage of the polymer surface and the NOM composition in the water. To evaluate early stage sorption processes of NOM on different plastic surfaces we incubated artificially UV-aged and pristine polymer particles (PS, PET, LDPE) and a reference material (glass) in sea water from the Pacific Ocean. The sea water was obtained from various positions in the Pacific to account for possibly different NOM composition. Afterwards the organic matter was desorbed from the plastic surfaces, analyzed by Fourier-transform ion cyclotron resonance mass spectrometry (FT-ICR-MS) and compared to each other. To account for measurement uncertainty, which may play a crucial role for data interpretation various control and blank samples were analyzed in parallel. Here we present our preliminary results for the comparison of different polymer samples. In brief, we observed differences in the chemical composition of the ecocorona between aged and non-aged particles and between different polymers with about 50% of shared molecular formulas. For all ecocorona samples the proportion of aliphatic substances was over 90%. Further data analysis will be done to identify differences between the adsorbed NOM on the analyzed particles and link them to the NOM composition of the corresponding sea water samples. [1] Lorite et al., J. Colloid Interface Sci., 359, 289-295, 2011.

Keywords : ecocorona , FT , ICR , microplastic , MS , NOM

### **UFZ** HELMHOLTZ Zentrum für Umweltforschung

### Qualitative analysis of the ecocorona on plastic surfaces

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### **Motivation and Aims**



for later biofilm formation. Previously it has been demonstrated that biofilm evolution is strongly dependent on the surface chemistry of plastic particles.<sup>[1]</sup> These findings imply that the ecocorona may has an influence on biofilm growth and may be dependent on the polymer surface.

Our aims are:

1 - Detection and comparison of NOM extracted from polymer surfaces with Fouriertransform ion cyclotron resonance mass spectrometry (FT-ICR-MS).

2 - Evaluation of differences in NOM quality adsorbed to different polymer particles.

3 - Evaluation of the influence of weathering of the polymer surface on the quality of adsorbed NOM.

Fig. 1 - Role of early stage sorption processes on biofilm growth.

### Key findings

- FT-ICR-MS is a suitable method to detect and qualify the ecocorona on plastic material.
- Marine NOM molecules attach to the surface of different plastic particles and form an ecocorona.
  - Preliminary results show differences between various polymer materials and weathering stages.

### Methods

- Incubation of pristine and artificially UV-weathered plastic particles in sea water from the Pacific Ocean for 1 hour and formation of the ecocorona.
- Extraction of adsorbed NOM from the polymer surface and clean-up with solid phase extraction (SPE).
- FT-ICR-MS measurement of extracts.
- Data evaluation and sample comparison including HCA, O/C and H/C ratios and presence/absence of molecule formulas.



Fig. 2 - Workflow for the qualitative analysis of the ecocorona on plastic surfaces.

### **References and Acknowledgements**

[1] Lorite et al., J. Colloid Interface Sci., 359, 289-295, 2011.

ProVIS - Centre for Chemical Microscopy



### **Preliminary results**

We established a workflow to evaluate the quality of adsorbed NOM on polymer particles and determined the molecular masses, molecule formulas and number of atoms (C, H, N, O, S) of chemical compounds present in ecocorona extracts with FT-ICR-MS. (Fig. 3)



Fig. 3 - van Krevelen plot of an ecocorona sample. Each spot represents a molecule formula present in the sample and its corresponding H/C and O/C ratios

Here we present some results from our first ecocorona measurements:

1 - A fraction of marine NOM attaches to the polymer surface to form an ecocorona. (Fig. 4 A)

2 - The quality of the ecocorona depends on the type of polymer of the plastic particle. (Fig. 4 B)

3 - Weathering of the polymer surface has and influence on the quality of adsorbed NOM. (Fig. 4 C)



Fig. 4 - Venn plots for the comparison of molecular formulas present in different samples. The overlap represents molecular formulas that were detected in both samples





### **Density Separation of Microplastics from solid sample matrices**

Schütze Berit, Thomas Daniela, Brunotte Habil Joachim.

While extent of contamination and effects of MPs in aquatic systems are under current study, terrestrial ecosystems have only recently come into focus (Rillig et al. 2012). Main sources are expected to be application of sewage sludge and compost as well as fragmentation of macroplastics in soil. As it is estimated that plastic release in terrestrial systems is 4-23 times higher than into the oceans (Horton et al. 2017), there is an urgent need to quantify contents and specify effects in soil. Definitions of microplastic as well as sample treatment methods and measurement techniques for MP analysis differ strongly within the scientific community. Consequently, previous studies calculated strongly varying concentrations of 0.34-43,000 particles/kg soil and 500 µm from soil was developed based on previously applied methods. Furthermore, tests of influence during separation of MPs were conducted by measuring surface changes with ATR-FTIR as well as changes in size of particles. Artificial substrates and compost with varying content of organic matter were tested. Four different solutions were applied in density separation to separate MPs from substrates: distilled water ( $\rho = 1.0$ g/cm<sup>3</sup>), NaCl ( $\rho = 1.19$  g/cm<sup>3</sup>), sodium hexametaphosphate (SHMP) ( $\rho = 1.30$  g/cm<sup>3</sup>) and NaBr ( $\rho =$ 1.53 g/cm<sup>3</sup>). Furthermore, organic matter was removed by digestion with H2O2. Recovery rates of 87.3-100.3 % for conventional polymers (PE, PP, PVC, PET) and 38.2-78.2 % (PLA, PBS, MaterBi film) for biodegradable polymers were measured with NaBr that showed highest recovery of applied density solutions.

Keywords : biodegradable plastic , compost , density separation , plastic debris , sediment , soil , spectroscopy



### Density Separation of Microplastics from Solid Sample Matrices

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### Aim of the Study

**Material & Methods** 

A reliable method for the analysis of MPs in environmental samples, especially in Solid Sample Matrices (e.g. soil), was not established until today, leading to a lack of comparability of studies. The aim of this study was to examine and improve previously applied methods based on Density Separation to isolate conventional and biodegradable MPs (> 500  $\mu$ m) from different Solid Sample Matrices with solutions of varying density.



FIG 1: Method procedure for the recovery tests of MPs in Solid Sample Matrices by Density Separation.

### Results

- Highest recovery rates were achieved with NaBr ( $\rho$  = 1.53 g/cm <sup>3</sup>)
- Conventional polymers (colored):

94.7 % ± 10 %

Biodegradable polymers (translucent):

57.6 % ± 32 %

- Influence of the method on some polymers (PE, MB) by treatment with H<sub>2</sub>O<sub>2</sub> at 60 °C and by mechanical influence
- Density of density solution recovery rates
- Content of organic matter recovery rates



**FIG 2:** Results for the recovery tests of MPs in different Solid Sample Matrices by Density Separation with NaBr.

### Conclusion

Complex solid matrices with increasing organic content (Compost > OECD 222 > Sand) make separation of Microplastics (MPs) more challenging. Consequently, removal of organic matter is a crucial step for sample preparation. Furthermore, recovery rates were higher for conventional than for biodegradable polymers. Due to the results of influence tests, it is concluded that lower recovery rates of biodegradable polymers mainly depend on appearance of polymers with reduced visibility during analysis of samples and not on chemical properties of the polymer. An automated chemical analysis could improve the method by decreasing visual dependency. Besides, beakers were not detected to be suitable for density separation as stirring led to fragmentation of Microplastics and corrosion of beakers. For further experiments, application of density solutions with  $\rho \ge 1.5 \text{ g/cm}^3$  are recommended.



● PE ● PP ● PET ● PVC ③ Biodegradable polymer FIG 3: Results for the recovery tests of MPs in artificial soil (OECD 222) by Density Separation with different solutions of varying density.



**Strategies for the separation of microplastics from water via density modification** Martínez De Pedro Zahara, Munoz Macarena, Ortiz David, Casas Jose A.

The widespread occurrence of microplastics (MPs) in the aquatic environment represents one of the most important environmental concerns nowadays. Although MPs can enter the environment through multiple pathways, wastewater treatment plants (WWTPs) have been recognized as important sources for MPs introduction into the aquatic systems. It is estimated that rivers, the main recipients of WWTPs discharges, transport up to 90% of the global MPs load into the sea (Schmidt et al., Environ. Sci. Technol. 2017, 51, 12246). Only in Europe, 520000 tons/year of MPs are released in WWTPs effluents (Alimi et al., Environ, Sci. Technol, 2018, 52, 1704). In this context, the development of innovative water treatment processes that allow the effective removal of MPs at WWTPs is crucial. This is an important challenge as the small size of MPs and their low chemical and biological reactivity significantly limit their elimination. The methods developed so far have been mainly focused on their sampling, showing important limitations for water treatment. In our research group, the development of strategies for MPs separation from water via density modification are being investigated. In this work, the removal of polystyrene (PS) and polyester fibers (PE) has been evaluated by their interaction with high-dense hematite (Fe2O3) microparticles. We have found that these mineral particles covered completely both PS and PE MPs leading to a significant increase on their density, which facilitates separation by sedimentation. On the other hand, carbon coating of MPs is being also investigated as an alternative approach to separate MPs. In this case, the removal of polyethylene terephthalate (PET) has been investigated by its coverage with a low-dense activated carbon, which allowed to increase the hydrophobic properties of the particles. These can adsorb air bubbles and float by reducing their density, favoring MPs separation via dissolved air flotation (DAF).

Keywords : activated carbon , hematite , microplastic , water treatment







### Strategies for the separation of microplastics from water via density modification

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**Poster session 26.9**
### Session 26.9\_O. Chaired by Eva Cardona, Menorca

### Microplastics at the water column: Their distribution and dynamics at the Eastern NorthAtlantic ocean down to 1150 meters

Daura Vega-Moreno, Bárbara Abaroa-Pérez, Paula Domínguez Rein-Loring, Carmen Presas-Navarro, Eugenio Fraile-Nuez, Francisco Machín. Paper number 337446

### European flat-oysters (*Ostrea edulis*) under anthropogenic pressure: An assessment of the combined chronic exposure effects of a warming marine environment and increasing microplastic pollution

Mackay-Roberts Nicholas, Paul Nina, Gerdts Gunnar, Pogoda Bernadette, Lannig Gisela, Lucassen Magnus, Bock Christian. Paper number 334519

### Ingestion of micro-litter in the deep-sea fish *Phycis blennoides* (Brünnich, 1768) along the Spanish Catalan coast

*Villodres Daniel, Dallarés Sara, Constenla María, Carrassón Maite.* Paper number 334560

### Preliminary evaluation of microplastic ingestion and phthalates load in Mediterranean Lanternfish (*Myctophum punctatum*)

Baini Matteo, Galli Matteo, Giani Dario, Panti Cristina, Rosso Massimiliano, Fossi Maria Cristina. Paper number 334573

### Same but different: A framework to design and compare riverbank plastic monitoring strategies

*Vriend Paul, Roebroek Caspar T.j., Van Emmerik Tim.* Paper number 334559

### Airborne microplastic deposition across the Weser River Basin

*Kernchen Sarmite, Held Andreas, Löder Martin G. J., Laforsch Christian.* Paper number 334353

### Microplastics at the water column: Their distribution and dynamics at the Eastern NorthAtlantic ocean down to 1150 meters

Daura Vega-Moreno, Bárbara Abaroa-Pérez, Paula Domínguez Rein-Loring, Carmen Presas-Navarro, Eugenio Fraile-Nuez, Francisco Machín.

Nowadays it is widely known that pollution by microplastics at the open ocean covers immense areas. Buoyant plastics tend to accumulate in areas of convergence at the sea surface such as subtropical gyres, while non-buoyant plastics accumulate at the seafloor. However, model studies have revealed that the total amount of plastic in the different oceans is not well correlated with the measured concentrations on the sea surface and the sea floor, showing a significant amount of missing plastic in the oceans. This deviation could be related to an underestimation of the role played by small fragments of plastic and fibers in the oceans. Furthermore, microplastic fragments with a density lower than the density of seawater have been gathered hundreds of meters below the sea surface in the Pacific Ocean due to their size and shape.

The main objective of this study is to carry out an equivalent analysis in the Atlantic Ocean. A total number of 44 samples were collected during four different oceanographic cruises between February and December 2019 and from the sea surface down to 1150 meters depth at the open ocean waters of the Canary Islands region (Spain). For each sample, 72 litres of seawater were filtered on board with a mesh size of 100  $\mu$ m, where the presence of microplastics has been clearly observed. Our results reveal the presence of microplastics at least up to 1100 meters depth, at the open ocean waters of the Northeastern Atlantic Subtropical Gyre. These microplastics have being horizontally transported by the ocean dynamics as passive drifters.

Keywords : Marine microplastics ; water column ; depth samples ; oceanic dynamic ; physical parameters ; passive drifters





### Microplastics at the water column: Their distribution and dynamics at the Eastern Northatlantic ocean down to 1150 meters -337446-

Daura Vega-Moreno<sup>\*1</sup>, Bárbara Abaroa-Pérez<sup>1</sup>, Paula Domínguez Rein-Loring<sup>1</sup>, Carmen Presas-Navarro<sup>2</sup>,

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### Abstract

Nowadays it is widely known that pollution by microplastics at the open ocean covers immense areas. Buoyant plastics tend to accumulate in areas of convergence at the sea surface such as subtropical gyres, while non-buoyant plastics accumulate at the seafloor. However, model studies have revealed that the total amount of plastic in the different oceans is not well correlated with the measured concentrations on the sea surface and the sea floor, showing a significant amount of missing plastic in the oceans. This deviation could be related to an underestimation of the role played by small fragments of plastic and fibers in the oceans. Furthermore, microplastic fragments with a density lower than the density of seawater have been gathered hundreds of meters below the sea surface in the Pacific Ocean due to their size and shape.



The main objective of this study is to carry out an equivalent analysis in the Atlantic Ocean. A total number of 51 samples were collected during four different oceanographic cruises between February and December 2019 and from the sea surface down to 1150 meters depth at the open ocean waters of the Canary Islands region (Spain). For each sample, 72 litres of seawater were filtered on board with a mesh size of 100 µm, where the presence of microplastics has been clearly observed. Our results reveal the presence of microplastics at least up to 1100 meters depth, at the open ocean waters of the Northeastern Atlantic Subtropical Gyre. These microplastics have being horizontally transported by the ocean dynamics as passive drifters.

### Results and conclusions

### Microplastic vertical distribution

Microplastics appear in all the samples analyzed between surface and 1150 meters depth, with an especially predominant presence of fibres, but small fragments can also be found down to the maximum sample depth.



and M04 at 1152 m (Nov'19) (right). (72 liters filtered per sample)

1242,651

Figure 2. MP samples collected between 0 & 280 meters depth (left), 0 & 523

m (center) and 0 & 550 m (right). (Black box- filtered volume per sample)

In these figures there are clear evidences of the presence of MP at the water column up to 1150 meters depth.



Figure 3. Microplastic vertical distribution of fibers (left) and fragments (right) sampled

### Ocean dynamic and convergence areas

As passive drifters, the MP spatial distribution is largely related to the underlying velocity field. The accumulation of MP might respond to the existence of long-lived mesoscale convergent structures, which is the case of wakes and eddies.

Velocities decrease with depth, being much higher at 1 m depth that at 250 m (Figure 4). The flow below 250 m north of the islands is largely affected during winter and fall by the presence of a meddy just north of Gran Canaria, which highly conditions the circulation and the convergent/divergent areas. On the other hand, south of the islands we may observe alternative patterns of convergent/divergent areas, likely related to the presence of mesoscale eddies.



Figure 4. Divergence of the velocity field during winter 2019 (first four panels, left) and fall 2019 (right) at 4 different depths: surface, 250 m, 500 m and 1100 m. Velocity fields are averaged during the 15 days previous to the samplings (numerical model IBI).

### Relation between MP vertical distribution and ocean dynamic

MP vertical distribution (Figure 3) might be indicating that the long-term velocity field north of the Canary Islands is not inducing a large variability in the MP spatial distribution. However, for stations M1 to M4, located south of the islands, the concentrations are notably higher than from ESTOC and variable with depth, likely as a consequence of the eddies that might be contributing to MP accumulation and vertical transfer along the water column.

### Acknowledgment

We would like to thank the Oceanic Platform of the Canary Islands (Plataforma Oceánica de Canarias, PLOCAN) for their support and sharing their research facilities with us (including our participation at two oceanographic cruises) and to the Spanish Institute of Oceanography (IEO) for their support in the context of VULcanología CAnaria SubmariNA project (VULCANA-II, IEO-2019-2021) funded by IEO with the participation in two oceanographic cruises during 2019.

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### European flat-oysters (*Ostrea edulis*) under anthropogenic pressure: An assessment of the combined chronic exposure effects of a warming marine environment and increasing microplastic pollution

Mackay-Roberts Nicholas, Paul Nina, Gerdts Gunnar, Pogoda Bernadette, Lannig Gisela, Lucassen Magnus, Bock Christian.

Marine organisms are naturally exposed to multiple environmental drivers and stressors due to the highly dynamic nature of the marine habitats in which they live. Anthropogenic activities have, in some cases, exacerbated these natural drivers, as with climate-change induced water temperature increase, or added novel stressors, such as the ingestion of microplastic pollution. The ability of marine organisms to tolerate combined drivers and stressors, from a physiological to ecological level, will greatly affect the distribution, functioning, and survival of populations, and is in general a poorly understood subject despite its vital importance. In this study, a chronic exposure experiment will be performed using a replicated multi-stressor mesocosm setup, examining the individual and combined impacts of increased water temperature (+3.1 oC), and environmentally-relevant suspended microplastic concentrations (10µm size, 50 MP l-1). The European flat-oyster (Ostrea edulis Linnaeus 1758) was selected for this study due to it being of both conservation and commercial importance, and, as a sessile filter-feeding bivalve, is greatly susceptible to exposure to both of these stressors. Six hundred adult O. edulis will be exposed to four different treatments consisting of increased temperature, increased MP, both stressors, or neither, for 200 days. Haemolymph, tissue, and whole organism samples will be taken throughout the exposure, for analysis of a wide range of biochemical and physiological functions, and quantification of MP retention. Growth, clearance and egestion rates, respiration and mortality will be recorded for each treatment. Variations in MP dynamics will be monitored, as will the biodeposition rates of the oysters themselves. The large time series dataset generated from this exposure study, covering a broad range of biotic and abiotic metrics, will provide a better understanding of the effects of MP ingestion by bivalve molluscs in both the current climate and under future conditions.

Keywords : Chronic Exposure , Microplastics , Multi , Oysters , stressor , Temperature

### European flat oysters (Ostrea edulis) under anthropogenic pressure:

### Assessing the combined effects of increasing microplastic pollution and a warming marine environment

N. MACKAY-ROBERTS (1), N. PAUL (2), H. NEUBAUER (1,3), H. MORENO (1), B. POGODA (1), G. LANNIG (2), M. LUCASSEN (2), C. BOCK (2), & G. GERDTS (1)

### INTRODUCTION

Microplastics (MP) have been identified as an emergent environmental threat, especially for filter-feeding organisms such as bivalves. This project focuses on the chronic multistressor impacts of environmentally relevant MP concentrations under a predicted warming scenario for elucidating potential additive, synergistic, or antagonistic effects on the physiology and performance of the ecologically important European flat oyster (Ostrea edulis). Additionally, we will examine changes to MP dynamics between treatments due to potential variability in the filtration, biodeposition and aggregation of MP caused by the oysters themselves.

### **MATERIALS & METHODS**

Sixteen semi-recirculating mesocosms (500 L vol.) were constructed for this experiment at the Alfred Wegener Institute marine station on Helgoland, Germany. Each mesocosm is supplied with running unfiltered seawater (400 L/h), and has one of four treatments applied. Three replicate mesocosms containing 50 O. edulis and one control without oysters are assigned to each treatment. Oyster samples from each mesocosm are taken in triplicate every 40 days over 200 days of exposure.

Individual O. edulis are assessed and sampled for a broad range of measurements, including clearance rate, growth rate, condition index, and hemolymph microbiome. Ingested MP counts are performed following alkaline digestion (10 % KOH, 50 °C, 48 h) of the soft tissue. Simultaneous MP sampling is conducted for water and sediment from each mesocosm. Additional O. edulis from each treatment are assessed for respiration rates and sampled for physiological tissue analysis (oxidative stress, energetics, metabolomics). Weekly water samples are taken for food quality (phytoplankton) assessments using flow cytometry and fluorescence spectroscopy, and further water samples are taken for monitoring MP concentrations. The experimental exposure period began in September 2020 and will continue through to May 2021



### FOUR TREATMENTS • Dosed microplastics with or without raised temperature • Non-dosed with or without raised temperature

**RAISED TEMPERATURE** 

### **DOSED MICROPLASTICS**

Eight mesocosms (heated and unheated) are dosed with fluorescent MP beads (polystyrene; 10 µm) to create an exposure concentration of 50 MP/L Deads (polysylprelative to the inflow rate. Concentrated MP solutions are stored in reservoirs on magnetic stirrers and automatically dosed every 15 minutes into the external mixing tanks of the mesocosms. Wastewater from the mesocosms is filtered to 1 µm to prevent environmental release.



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### above the annuent water temperature to annuence Cooper Moder Intercomparison Project predicted warming for the North Sea by 2099. Real time monitoring data of the ambient and warmed mesocosm temperatures is relayed to a computerised switching cabinet which turns off corresponding heaters once target temperatures are reached. AMBIENT . . HEATED

Eight mesocosms (with and without dosed MP) are warmed to +3.1 °C

above the ambient water temperature to simulate Coupled Model

sor location (Red)

### Ostrea edulis

Linnaeus 1758 Common name: European Flat Oyster or Native Oyster Depth Range: Littoral fringe - 50 m Native Distribution: North Atlantic (Norway -Morocco) Mediterranean Sea. Black Sea

Once common in European waters, this species became functionally extinct following overfishing and disease events in the 20th century. Restoration efforts are underway to protect and expand existing populations, and to reintroduce this species to previously inhabited areas.

For more information on O. edulis restoration projects please visit https://noraeurope.eu/ or scan the OR code.





### Figure 4 Photograph of an adult Ostrea edulis showing flat upper valve surface

### RESEARCH **OBJECTIVES**

- Determine the chronic effects of MP exposure for O. edulis under natural seasonal conditions
- Assess responses to the combined effects of increased temperature and MP exposure from the whole organism to cellular level.
- Model the aquatic distribution of MP in relation to absence/presence of O. edulis.
- Identify species-specific vulnerabilities to these anthropogenic pressures for eluding potential ecological consequences.

### ACKNOWLEDGMENTS

The authors wish to thank the technical staff of the Biologische Anstalt Helgoland for their assistance in the construction of this experiment.

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HELMHOLTZ

### Ingestion of micro-litter in the deep-sea fish *Phycis blennoides* (Brünnich, 1768) along the Spanish Catalan coast

Villodres Daniel, Dallarés Sara, Constenla María, Carrassón Maite.

The presence of micro-litter (ML) (including plastic beads, films, fragments and anthropogenic fibres, ; 5 mm) in aquatic environments has attracted increasing attention of the scientific community in the last decades. Specially, the Mediterranean Sea is one of the most affected seas on the planet by high concentrations of plastics. However, their presence and impact on biota are still scarce. In the present study, 42 specimens of the greater forkbeard (Phycis blennoides), were sampled from two different areas off the Catalan coast subject to different anthropogenic impacts (20 from off Barcelona and 22 from off the Ebro Delta). The gastrointestinal tracts (intestines and pyloric caeca, since stomachs were everted in almost all cases) of the fish were analysed to assess ML ingestion levels and composition. Fish biometric measurements as well as parasitological infection levels were also recorded in order to assess a possible effect of ML on fish health (condition indexes and parasitism). In addition, ML characterization was performed based on visual an FTIR analyses. According to results, presence of ML was observed in 72.72% individuals sampled off Ebro Delta and in 95% individuals sampled off Barcelona. Each specimen contained an average of 1.7 and 5.4 ML in Ebro Delta and in Barcelona, respectively. Ingested ML in Ebro Delta consisted in 97.4% fibres and 2.6% small fragments, and fibres showed a mean length of 1.64 mm and a mean diameter of 0.017 mm. Similarly, ML present in specimens from Barcelona consisted in 97.2% fibres and 2.8% small fragments, and fibres displayed a mean length of 1.68 mm and a mean diameter of 0.021 mm. Current results point to Barcelona as an area of more ingestion for MPs than Ebro Delta, probably due to higher environmental availability. Details about fibre composition and fish health indicators will be reported during the congress.

Keywords : bioindicators , deep sea , marine pollution , Mediterranean , Microlitter , microplastics , Phycis blennoides

### **INGESTION OF MICROPLASTICS IN THE DEEP-SEA FISH** PHYCIS BLENNOIDES (BRÜNNICH, 1788) ALONG THE Universitat Autònoma **SPANISH CATALAN COAST**

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### INTRODUCTION

de Barcelona

Microplastics (MPs)

- Ubiquitous plastic particles, man-made and smaller than 5 mm (Thompson et al., 2004)
- The effects of ingestion in natural environments are still poorly known (Wright et al., 2013)

The aim of this study is to evaluate the levels of microplastic ingestion in Phycis blennoides, the most abundant benthopelagic fish on the upper slope (200-690 m depth) of the NW Mediterranean, its potential effects on fish health and the geographical variation among 3 localities with diferent impacts (Barcelona, Ebro River Delta and Blanes





sampling areas						
Area	Barcelona	Blanes	Delta			
Mean fish standard length (cm) (SL)	17.47 ± 1.08ª	19.19 ± 1.20 <sup>b</sup>	16.11 ± 1.82 <sup>c</sup>			
Mean fish eviscerated weight (g) (EW)	56.76 ± 8.84	81.77 ± 13.86	44.97 ± 17.54			
Fulton's condition factor (K)	$1.06 \pm 0.09^{a}$	1.15 ± 0.07 <sup>b</sup>	0.64 ± 0.32 <sup>a</sup>			
Mean HSI (%)	$3.46 \pm 0.98^{a}$	$4.04 \pm 1.49^{a}$	2.37 ± 0.67 <sup>b</sup>			
Mean GSI (%)	0.06 ± 0.03 <sup>a</sup>	0.11 ± 0.03 <sup>b</sup>	0.08 ± 0.03ª			

ipt letters indicate significant differences among sampling areas (One-Way ANOVA and post-hoc ar

Table 2. Prevalence (%) and AFs parameters in P. blennoides from the three sampling areas

Area	Barcelona	Blanes	Delta
AF Prevalence (%)	90ª	50 <sup>b</sup>	73ª
Mean Abundance	5.15ª	0.75 <sup>b</sup>	1.72 <sup>b</sup>
Mean AF length (µm)	1.68 ± 1.05ª	4.81 ± 3.74 <sup>b</sup>	1.89 ± 1.33ª
Mean AF diameter (µm)	0.02 ± 0.02ª	0.024 ± 0.01 <sup>a</sup>	0.01 ± 0.005 <sup>a</sup>

Figure 1. Mean abundance (a) and prevalence (%) (b) of AFs present in intestine and pyloric caeca contents of P. blennoides across localities



Different superscript letters and numbers indicate sign

- No relationships were observed between area and number of CMM and the rest of parameters (health indices, AFs parameters) nor differences in CMM among areas.
- No relationships were observed between health indicators and AFs parameters.



This work was partially supported by the Spanish Ministry of Science. Innovation and Universities (MICIU) project "PLASMAR" (RTI2018-094806-B-100) and by the Catalan Department of Agriculture, Livestock, Fisheries and Food (European Maritime and Fisheries Fund (EMFF)) project "SOMPESCA" (ARP059/19/00003).

health of the fish

### DISCUSSION



P. blennoides from Barcelona presented the highest abundance and prevalence (%) of AFs



### **Preliminary evaluation of microplastic ingestion and phthalates load in Mediterranean Lanternfish (***Myctophum punctatum***)**

Baini Matteo, Galli Matteo, Giani Dario, Panti Cristina, Rosso Massimiliano, Fossi Maria Cristina.

Myctophids plays a key role in trophic webs from the continental slope and pelagic waters linking the zooplankton to top predators, such as cetaceans. They feed mainly on copepods, euphausiids, amphipods, and decapods during upward nightly migration into the epipelagic zone where floating plastic debris is described to accumulate. As a consequence of their vertical migration, these fish may reflect the ingestion of microplastics (MPs) along the water column and be exposed to load of hazardous chemicals such as Phthalates esters (PAEs) leached from these particles. Within the Plastic Busters MPAs project, 20 specimens of Myctophum punctatum have been collected using a plankton net during a sampling campaign carried out in the North-Western Mediterranean Sea during summer 2018. Fish were properly processed to evaluate the potential relationship between the ingestion of MPs and the levels of chemicals released from the ingested particles. The gastrointestinal tract of each organism was chemically digested using KOH and filtered through a glass-fiber membrane. Levels of four different PAEs (DIBP, DBP, DEHP and BBzP) were evaluated in the fish muscle, as plastic tracers. MPs isolated were characterized by size, shape, colour, and chemical composition by FTIR spectroscopy. The presence of MPs confirms the hazard associated with the debris ingestion for this species and its transfer throughout the marine trophic web. The chemical analysis revealed for the first time the levels of PAEs in the muscle tissue of Mediterranean lanternfish, confirming their bioavailability for these organisms. Being these toxic chemicals used as plasticizers, their presence could be due to the leaching of these compounds from floating plastics debris ingested during the feeding activity. The results provide data on an ecologically valuable and poorly investigated species, emphasizing the challenges between the plastic debris accumulation and the associated risks to wildlife.

Keywords : Endocrine disruptors , Microplastic ingestion , Phatalate esters , Plastic tracers

### Preliminary evaluation of microplastic ingestion and phthalates load in Mediterranean Lanternfish (Myctophum punctatum)



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### INDRODUCTION

Myctophids plays a key role in trophic webs from the continental slope and pelagic waters linking the zooplankton to top predators, such as cetaceans. They feed mainly on copepods, euphausiids, amphipods, and decapods during upward nightly migration into the epipelagic zone where floating plastic debris is described to accumulate. As a consequence of their vertical migration, these fish may reflect the ingestion of microplastics (MPs) along the water column and be exposed to load of hazardous chemicals such as Phthalates Esters (PAEs) leached from these particles.

### **MATERIAL & METHODS**

Within the Plastic Busters MPAs project, 20 specimens of Myctophum punctatum have been collected using a plankton net during a sampling campaign carried out in the North-Western Mediterranean Sea during summer 2018. Fish were properly processed to evaluate the potential relationship between the ingestion of MPs and the levels of chemicals released from the ingested particles. The gastrointestinal tract of each organism was chemically digested using KOH and filtered through a glass-fiber membrane. Levels of four different phthalates (DIBP, DBP, DEHP and BBzP) were evaluated in the fish muscle, as plastic tracers. MPs isolated were characterized by size, shape, colour, and chemical composition by FTIR spectroscopy.



RESULTS

The presence of MPs confirms the hazard associated with the debris ingestion for this species and its transfer throughout the marine trophic web. The chemical analysis revealed for the first time the levels of PAEs in the muscle tissue of Mediterranean lanternfish, confirming their bioavailability for these organisms. Being these toxic chemicals used as plasticizers, their presence could be due to the leaching of these compounds from floating plastics debris ingested during the feeding activity. The results provide data on an ecologically valuable and poorly investigated species, emphasizing the challenges between the plastic debris accumulation and the associated risks to wildlife



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### Same but different: A framework to design and compare riverbank plastic monitoring strategies

Vriend Paul, Roebroek Caspar T.j., Van Emmerik Tim.

Plastic pollution in rivers negatively impacts human livelihood and aquatic ecosystems. Monitoring data are crucial for a better understanding of sources, sinks and transport mechanisms of riverine macroplastics. In turn, such understanding is key to develop effective plastic pollution prevention, mitigation, and removal strategies. Riverine plastic is mostly studied through the monitoring of floating plastic and through the quantification of plastic deposited on riverbanks. Existing riverbank plastic measurement methods vary greatly, which complicates direct comparison of data collected with different monitoring strategies. We present a framework to better compare and to aid the design of riverbank plastic monitoring methods, which is based on four common elements distilled from riverbank (plastic) litter monitoring methods currently in use. This framework can be used by scientists and practitioners to find the right trade-offs between the data required to answer specific research questions, and the available resources. Subsequently, we use the framework to suggest effective monitoring strategies for three frequently asked research questions. With this presentation, we aim to provide a first step toward harmonization of riverbank plastic litter monitoring efforts.

Keywords : citizen scientists , hydrology , litter , macroplastic , marine litter , microplastic , observations



# A Framework to Design and Compare Riverbank Plastic Monitoring Strategies Paul Vriend<sup>1</sup>, Caspar Roebroek<sup>1</sup> & Tim van Emmerik<sup>1</sup> Same but Different:

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# 1. Introduction

- Data on presence of macroplastic on riverbanks is essential for design of mitigation and removal strategies
- complicating comparison of data between programs Current methods to quantify riverbank plastic vary greatly,
- Goal of this paper is to create a framework that can be used step towards standardization to compare or aid design of monitoring methods, as a first

### 2. Methods

- Determine the range of possibilities for each of these Analyze current monitoring efforts for common elements
- elements
- Determine effort required for each end of this range

# Convert to framework

# 3.1 The Framework

- Four main elements: Space, Time, Observers, and
- Each main elements can be further divided in sub-elements Categorization
- Kange of possibilities shows what options can be chosen for currently used in literature each element, with left and right showing the extremes

Categorization	Observers				Time			Space	Element
Category Size Range		Duration	Structure	Frequency	Period	Structure	Sampling area	Domain	Sub-element
Material Based Macro	Citizen Scientists	Singular	Structured	Yearly	4 Weeks	Structured	Subsampling	Sub-basin	
† į	*	1	*	Ť	•	•••	•		
	÷	•	•	•••		•		•	Range
Identity Based Macro and Micro	Trained Professionals	Multi-year	Unstructured	Daily	Single day	Unstructured	Sampling larger area	Multi-basin	

Fig. 1 – A schematic representation of the proposed framework for riverbank plastic pollution quantification protocols. The range of possibilities is given for each element within the framework. The colored dots represent where the Plastic Prates' (blue), Schone Rivieren<sup>2</sup> (green), Battulga's (vellow), and CrowdWater' (red) are on this scale of possibilities.

### 3.2 Space

- Domain: scale at which sampling is performed
- Structure: method in which sampling locations are **Sampling area**: the space in which samples are taken

chosen



Fig. 2 - An overview of sampling areas (shaded areas, black dashed lines for microlitter analysis) for multiple riverbank plastic quantification protocols, and the NOAA beach litter protocol<sup>6</sup> to exemplify random sampling.

### 3.3 Time

- measurement round are taken **Period**: the timeframe in which measurements for a
- Frequency: number of sampling rounds in a year
- Structure: whether to use a structured protocol (using rounds and frequency) or randomly sample throughout the year
- **Duration**: range of time in which plastic is sampled



Fig. 3 - The four elements of time depicted on two timelines, where timeline (A) represents structured temporal sampling, and timeline (B) depicts unstructured temporal sampling. The duration is the total time that samples are taken, the frequency is the number of samples that are taken annually, and the period the time that samples are considered as one measuring round.

Sampling or one riverbank

<sup>3</sup>Battulga et al. (2019) <sup>4</sup>van Emmerik et al. (2020b) <sup>5</sup>Lippiatt et al. (2013)

# 3.3 Observers

- Group of people that sample riverbank macroplastic
- Growing interest for citizen scientists due to possibility to sample a large domain, though can affect data quality

# 3.5 Categorization

- Category: level of detail in characterization of items
- Categorization can be aggregated to higher levels to compare
- between monitoring strategies (see figure below)
- Size range: Macro-, microplastic, or both



Fig. 4 - An example of riverbank plastic dassification, where the upper layer represents the most detailed categorization (identity based) hased on OSPAK etagogization, not an exhaustive bits), and each layer below represents a higher level of aggregation. The type of categorization and how this categorization is achieved is listed on the right side.

# Trade-off analysis

- Limited amount of resources, so trade-offs are made
- Score each range from least effort to most effort



Fig. 5 - Graphical representation of the choices made for each element for the Plastic Frates protool<sup>11</sup> (A), Schone Rivieren protocol<sup>15</sup> (B), the Battling protocol<sup>15</sup> (D), and the CrowdWate protocol<sup>16</sup> (D). Where each axis represents the following elements: (1) Sampling scale, (2) Store–Sampling area, (2) Time–Frequency (4) Time–Paricia, (5) Time–Duration, 6. Observers, 7. Categorization, and (8) Size range. For each axis, the inner part represents lop priority, and the outer part represents ligh priority. The sub-element of structure for time and space vere excluded since these factors do not influence total cost.

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<sup>1</sup>Kiessling et al. (2019) <sup>2</sup>van Emmerik et al. (2020a)

References

### Airborne microplastic deposition across the Weser River Basin

Kernchen Sarmite, Held Andreas, Löder Martin G. J., Laforsch Christian.

Contrary to the well-studied microplastic (MP) contamination in aquatic and terrestrial systems, the atmosphere has gained much less attention. Previous studies focused on MP in atmospheric samples and have found measurable amounts of synthetic polymers in bulk deposition, snow, settled dust and in air. So far, exposure to airborne MP by inhalation has been linked with respiratory system diseases and lung cancer, although comprehensive studies on human health and risks are urgently needed. To better understand the health effects of airborne MPs, much more research in-depth is required regarding the levels of contamination in air, the role of atmospheric transport and deposition, and the physics and chemistry of those pollutants. This work aims at assessing MP deposition rates across the German River Weser basin, which connects urban, agricultural and rural areas with the North Sea. Total atmospheric deposition was sampled monthly from March to September/October (2018) at four sites (Wasserkuppe, Kassel, Bremen and Bremerhaven) and twice at two sites (Rinteln, Solling) in March and in September, while site Kassel was sampled monthly over one year from April 2019 to April 2020 for wet-only and dry-only atmospheric deposition. Samples were treated following a novel purification protocol based on oxidative-enzymatic digestion in order to remove organic and inorganic residues. The isolated MP fraction was analyzed with FTIR microscopy which provides polymer specific information about the particle count, size distribution and particle shape. All sampled sites contained measurable amounts of MPs with deposition rates ranging from 10 to 367 particles/m2/day (99±84, mean±SD). Airborne MP deposition rates were higher in urban (Bremerhaven, Bremen, Kassel) than in rural areas (Wasserkuppe, Solling, Rinteln) and have seemingly no seasonal characteristics. In the studied particle diameter range (¿ 11 µm), PP, PE and PET are the predominant plastic types in air in Middle and North-West Germany.

Keywords : airborne microplastics , deposition rate , FTIR , plastic pollution , Weser river basin



# Airborne microplastic deposition across the Weser river basin

# S. Kernchen<sup>\*1</sup>, M. G. J. Löder<sup>2</sup>, C. Laforsch<sup>2</sup> and A. Held<sup>3</sup>

## Introduction

connects urban, agricultural and rural areas in studied across the Weser river basin, which this work MP deposition fluxes (DFs) were atmosphere has gained much less attention. In microplastic (MP) contamination aquatic, terrestrial environmental concern globally. Contrarily to Germany with the North Sea. Increasing plastic pollution is of growing and biological systems, in the



# Experimental Work

digestion to remove organic and inorganic residues. The atmospheric deposition. Samples were treated following a site KasselWasser was sampled monthly over one year from Total novel purification protocol based on oxidative-enzymatic strategically different sites monthly from March to isolated MP fraction was analyzed with FTIR microscopy in a April 2019 to April 2020 for wet-only and dry-only two sites (Rinteln and Solling) in spring and autumn, while KasselWasser, hanseWasser and Bremerhaven) and twice at September/October (2018) at four sites (Wasserkuppe, size range from  $11 \ \mu m$  to > 500  $\mu m$ . atmospheric deposition was collected at XIS

### Results

amounts of MPs with deposition fluxes corresponds to an average deposition of 0.05 ranging from 10 to 367 particles (N) per  $m^2$ All sampled forms (96.9%) per kg ha<sup>-1</sup> annually. MP fragments in different day (99±84, sites contained measurable comprised most of the mean±SD), which

MPs were made of PP, PE, PET, PVC, and PS particles made of 17 different synthetic detected plastic particles, followed by fibres polymers were identified. The predominant range from 11  $\mu$ m to 100  $\mu$ m. In total, particles were in the smallest studied size (2.9%) and spheres (0.2%). 92% out of all

approximately 5% of the total deposition. accounting for 82% of all particles. Deposition deposition. Dry deposition comprised only MPs were significantly higher than dry-only r=0.8, p<0.05). Wet-only deposition fluxes of significantly with precipitation (Pearson's fluxes at the site hanseWasser correlated

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total deposition flux [N m<sup>-2</sup> day<sup>-1</sup>] of MPs at the site hanseWasser and averaged wind direction. The highest deposition flux was observed downwind of the city Bremen size distribution of deposited non-fibrous MPs; (d) plastic-type distribution in a set of 32 total deposition measurements; (e) wind rose plot of the relationship betweer Fig. a-e. (a) Sampling sites; (b) total atmospheric deposition fluxes of MPs (mean±SD). Numbers above bars correspond to the population density (2019) at the site; (c)



### Summary

- × MP total deposition fluxes were higher in urban areas than in rural areas
- × Deposition fluxes across the Weser river basin showed no clear seasonal characteristics
- ⊁ No tight correlation between population density and deposition fluxes was observed.
- × We cannot confirm that wastewater treatment plants generate airborne MPs
- × Rainfall plays a major role in airborne MP scavenging from the atmosphere
- ⊁ Wind characteristics can be helpful to interpret MPs amounts in the air.



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### Session 26.9\_Me. Chaired by Miguel Tamayo, Madrid

### The impact of nanoplastic on Antarctic krill embryonic development in current and future acidified conditions of the Southern Ocean

Rowlands Emily, Galloway Tamara, Cole Matthew, Lewis Ceri, Peck Victoria, Thorpe Sally, Manno Clara.

Paper number 334748

### Effects of in vitro and in vivo exposure of polystyrene nanoplastics in the marine mussel *Mytilus galloprovincialis*

Gonçalves Joanna Melissa, Vilke Juliano Marcelo, Da Costa Mestre Nélia, Garcia Da Fonseca Taína, Serrão Sousa Vânia, Bebianno Maria João. Paper number 335135

**Immune response of terrestrial crustacean** *Porcellio scaber* **to microplastics exposure** *Jemec Kokalj Anita, Drobne Damjana, Dolar Andraz.* Paper number 335544

**Coping with synthetic and natural microparticles: brown shrimp and antioxidant defence** *Korez Špela, Gutow Lars, Saborowski Reinhard.* Paper number 334431

### A method to quantitatively analyse Microplastic transport and retention in an experimental flume environment

Boos Jan-Pascal, Gilfedder Benjamin, Frei Sven. Paper number 334407

### The impact of nanoplastic on Antarctic krill embryonic development in current and future acidified conditions of the Southern Ocean

Rowlands Emily, Galloway Tamara, Cole Matthew, Lewis Ceri, Peck Victoria, Thorpe Sally, Manno Clara.

Antarctic krill (Euphausia. Superba) are amongst the most important and abundant filter-feeding metazoans in the Southern Ocean. The negative effects of rapid warming and ocean acidification (OA) have already been acknowledged for the species. Less explored is the impact of increasing plastic pollution, which the Southern Ocean has failed to avoid. Antarctic krill might be of increased risk of exposure to plastic particulates due to the ability of sea-ice to act as a sink for plastic particulates coupled with krill's reliance on sea-ice, though may have diminished coping mechanisms for reasons such as weak genetic variability. Nanoplastic, the smallest form of plastic debris, is predicted to be the most hazardous yet a paucity of studies explore its potential toxicological impacts on this keystone species. The potential multi-stressor effects of plastic particulates coupled with the climate stressors exasperated in the polar regions are also unexplored. Here, we investigate the single and combined effects of NP (spherical, aminated (NP-NH2), yellow-green fluorescent polystyrene nanoparticles) and OA (pCO2-manipulated seawater, pH 7.7) on the embryonic development of eggs produced by Antarctic Krill. Organisms were collected in the Atlantic sector of the Southern Ocean. Eggs from a single female were incubated at 0.5 °C with three treatments: with 0.16 µm nanoplastic spheres, in acidified conditions, plus with the combined treatment of nanoplastic (0.16µm) and acidification. We present results to date on this study enhancing our understanding of the potential impact of plastic pollution on Krill populations, at critical yet potentially most vulnerable embryonic life stages, in their current and predicted future environment. This is acknowledged to be a critical future research direction since addressing the toxicology of plastic particulates in singularity will fail to mimic future multi-stress conditions.

Keywords : Antarctic krill, Ecotoxicology, Nanoplastic, Ocean Acidification

### The potential impact of nanoplastic on Antarctic krill embryonic development in current and future acidified conditions of the Southern Ocean

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### Introduction



### Fig 1. Antarctic krill lifecycle (Carr, 2014)

### **Hypotheses**

- Nanoplastic will deleteriously impact the embryonic development of *E. superba.*
- The synergistic impact of nanoplastic and ocean acidification will increase detrimental consequences on embryonic development.

### Incubation

Organisms were collected in the Atlantic sector of the Southern Ocean.

Embryos (N~680) from a single female were incubated at 0.5 °C in a multiwell plate with treatments of 0.16 µm spherical, aminated (NP-NH<sub>2</sub>), yellow-green fluorescent polystyrene nanoparticles at a final concentration of 2.5 µg/ml; in acidified conditions (pH 7.7); or with the combined treatment of nanoplastic and acidified water. Three multiwells contained 0.22 µm filtered seawater as a control. All treatments were carried out in triplicate.

### Analyses

Embryonic development

- Classified at the incubation end point as :
- no development, 2. two-cell, 3. multi-cell, 4. limb bud, 5. entrapped nauplii, 6. hatched nauplii.



Microplastic
 Nanoplastic
 Chemical

Fig 2. Potential pathways and fates of micro- and nano-plastics in the polar regions. Potential interactions with other anthropogenic chemical and climate stressors, and interactions with pelagic filter-feeding communities including Antarctic krill (Rowlands et al., 2020).

### Nanoplastic quantification

Surfaces of a subset of embryos were imaged via scanning electron microscope (SEM) to determine absence/presence of nanoplastic adhered to the outer embryo surfaces (Fig 3). The residual of digested krill eggs will also be imaged via SEM to determine whether nanoplastic is able to penetrate the chorion (membrane).



### Summary

Results from this multi-stressor nanoplastic/ocean acidification incubation will enhance our understanding of the potential impact of plastic pollution on Antarctic krill, at critical yet potentially most vulnerable embryonic life stages, in their current and predicted future environment. This is acknowledged to be a critical future research direction since addressing the toxicology of plastic particulates in singularity will fail to mimic future multi-stress conditions.

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### Effects of in vitro and in vivo exposure of polystyrene nanoplastics in the marine mussel *Mytilus galloprovincialis*

Gonçalves Joanna Melissa, Vilke Juliano Marcelo, Da Costa Mestre Nélia, Garcia Da Fonseca Taína, Serrão Sousa Vânia, Bebianno Maria João.

Plastic debris has been a worldwide concern over the past years, wherein macro- and micro-plastics have been scientists' main focus. However, there still remains a scarcity of information on the effects that nanoplastics pursue on marine biota. This study aimed to comprehend the effects on 50 nm polystyrene nanoplastics (nPS, 10 µg/L) in an *in vitro* (24 h) and *in vivo* (21 days) exposure in the marine mussel Mytilus galloprovincialis. Characterization of nPS was conducted, whereby the Zpotential and the hydrodynamic diameter were calculated in both freshwater and seawater. Mussel haemocytes were exposed in vitro to nPS and cell viability was assessed using the neutral red assay. In the *in vivo* exposure to nPS (10 µg/L), genotoxicity and oxidative stress were analysed in mussel gills. Results showed that nPS in freshwater had an average hydrodynamic diameter of 25 nm, with a  $\zeta$ -potential of -68.4 mV, indicating that there is no aggregation. In seawater, on the other hand, the hydrodynamic diameter increases, indicating that aggregation does occur. ζ-potential of nPS in seawater is of -0.068 mV, meaning that in these conditions, as ζ-potential is closer to zero, aggregation is favoured. Results showed that turbidity is inversely proportional to sedimentation. Regarding the *in vitro* exposure, nPS decrease cell viability significantly (*p*<0.05) and genotoxicity occur in the haemolymph of nPS exposed mussels (*p*<0.05). The antioxidant enzyme catalase (CAT), as well as the biotransformation enzyme glutathione-s-transferase (GST) after 3 days of exposure to nPS, presented a decrease in activity (*p*<0.05) at days 7 and 14 when compared to controls and the other days of exposure (p < 0.05). Oxidative stress, genotoxicity, as well as a decrease in cell viability was observed in mussels after exposure to nPS, highlighting the potential negative repercussions that nPS may have on marine biota.

Keywords : Mussels , Nanoplastics

# Effects of *in utro* and *in uno* exposure of polystyrene nanoplastics in the marine mussel *Mytilus galloprovincialis*

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### Immune response of terrestrial crustacean *Porcellio scaber* to microplastics exposure

Jemec Kokalj Anita, Drobne Damjana, Dolar Andraz.

In recent years a number of studies have reported the effects of microplastics on terrestrial invertebrates at the level of the whole organism, but considerably less is known about the sublethal effects after long-term exposure. One of the likely microplastics-induced changes in organisms are those related to innate immunity. For example, indirectly by acting on the diversity and function of intestinal microorganisms, mechanical damage to the digestive tract, effects on food quality and food intake, and release of hazardous chemical additives from microplastics. In this work, we investigated the modulation of immune processes in macro-decomposer woodlice Porcellio scaber exposed to polyester fibers and crumb rubber from end-of-life car tyres. Organisms were exposed through soil for 3 weeks and immune endpoints were followed throughout the exposure (day 1, 2, 4, 7, 14, 21). Tested immune markers were: haemocyte viability and count (total and differential), and antioxidant enzymes. Our results show that neither polyester fibers nor crumb rubber affected the survival of woodlice, but immune processes were altered during the exposure period. The most significant change was increase in total haemocyte count, and specifically granulocytes, which implies alteration of metabolic processes in haemocytes. Viability of haemocytes however was not affected which implies that effect of potential chemicals leached from microplastics (in particular crumb rubber) was not significant. Antioxidant enzyme activities varied during the exposure period but were not evidently different from controls. We did not observe a difference in effects induced by crumb rubber and polyester fibers although these particles differed significantly in their physico-chemical properties. This study shows that microplastics exposure modulates immune processes in woodlice. This implies that prolonged exposure of terrestrial invertebrates to microplastics may results in their altered immunocompetence and research in this direction is of interest.

Keywords : car tyre particles , crustacean , feeding , immune response , terrestrial invertebrates , textile fibers



### IMMUNE RESPONSE OF TERRESTRIAL CRUSTACEAN PORCELLIO SCABER TO MICROPLASTICS EXPOSURE

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**INTRODUCTION AND AIM**: Microplastics can potentially enter the terrestrial environment by fragmentation of plastic wastes, via microplastics contaminated sewage sludge deposition on agricultural land, or as a result of extreme agricultural praxes. Despite the potential presence of microplastics in terrestrial environments, data regarding the effects of microplastics on terrestrial organisms are scarce, in particular it is unclear how long-term exposure affects immune processes. In this study, we investigated the immune response of terrestrial isopod *Porcellio scaber* to crumb rubber and polyester fibers.

### ANTICIPATED IMMUNOMODULATORY EFFECTS OF MICROPLASTICS



**METHODS:** Animals were exposed to polyester textile microfibers or crumb rubber from end-of-life tyres (0.05 to 1.5% w/w) in Lufa 2.2 soil for 3 weeks (Selonen et al., 2020). Immune response was followed during and after the exposure. The following immune parameters in the haemolymph were assessed: total haemocyte count (THC); differential haemocyte count (DHC); haemocyte viability.

<u>Crumb rubber</u>: produced from mixed end-of-life passenger car tires by cryo-milling. The powder contained several different synthetic rubbers, 10-35% natural rubbers and 25-35% carbon black. Traces of metals and PAH were detected. The dominant fraction of particles lies between 80 and 110  $\mu$ m (mean 102.9  $\mu$ m).

<u>Polyester fibers</u> were prepared by cutting a fleece blanket followed by cryo-milling using a homogenizer (MillMix 20, Domel, Slovenia). The fibers had a shape of narrow strips with an approximate length 220  $\pm$  200  $\mu$ m, ranging from 12 to 2 870  $\mu$ m and height of 6  $\mu$ m (Selonen et al., 2020).

<u>Natural control:</u> wooden particles from beech tree, mean size 212  $\mu m$ 

**RESULTS**: Immune parameters are modulated upon exposure to microplastics. Total haemocyte count (THC) was slightly, but insignificantly, increased and also their viability was not affected. The number of granulocytes was increased and semigranulocyte count was decreased implying a shift in metabolic action of haemocytes. The response was time-dependent with the peak of changes in period 2-7 days. A gradual return to control levels was found after 3 weeks of exposure, however some parameters were not entirely the same as the controls. No clear microplastics dose-related trend was observed. Natural control (wooden fibers) did not induce any effects.



**<u>CONCLUSIONS</u>**: Our results reveal a clear time-dependent immunoactivation in *Porcellio scaber* exposed to microplastics. The response was most pronounced after 4-7 days and a gradual decline to nearly control levels were evident after 21 days. What this alteration in immune processes means for the immunocompetence of the organism as well as its general fitness remains to be investigated.





\* in comparison to control (control=0; source culture) a,b = comparison between treatments/day

REFERENCES: Selonen et al., Science of the Total Environment, 700, 134451. ACKNOWLEDGEMENTS: Slovenian Research Agency, through Research program "Integrative zoology and speleobiology P1-0184" and ARRS J1-2482 project

### Coping with synthetic and natural microparticles: brown shrimp and antioxidant defence

Korez Špela, Gutow Lars, Saborowski Reinhard.

Broad applicability and industrial mass production established plastics in our everyday life. Due to careless use and poor recycling strategies, synthetic plastic material emerged to one of the greatest threats to the marine environment. In the environment, plastics are prone to disintegrate into microplastics. The wide size range make the microplastics available to different marine fauna, which may mistake them with food items. Apart from the synthetic microparticles, marine organisms are persistently surrounded in their habitat by various suspended organic microparticles (e.g. remains of bivalves, cellulose fibers, chitinous remains) and inorganic microparticles (e.g. silica frustules, sediment grains). Therefore, the goal of the present study was to investigate whether these natural particles may potentially harm organisms in the same way as synthetic particles. We determined the antioxidant defense response of brown shrimp, Crangon crangon, subjected to microparticles of different origin. The shrimp were exposed to 20 mg L-1 of natural (clay, diatoms) and synthetic (TiO2, PVC, PLA) microparticles for 6, 12, 24 and 48 h. After exposure, the shrimp were dissected and the midgut gland, also referred as to digestive gland, was withdrawn. The tissue was homogenized and processed for analysis of the antioxidant enzymes superoxide dismutase, glutathione peroxidase, glutathione reductase, and the total antioxidant potential. The results, so far, show no increase in the cellular stress response in the brown shrimp, irrespective of the origin of the chemical nature of the microparticle. These findings suggest that C. crangon may better cope with microplastic pollution then others marine organisms. Comparative experiments with crustaceans from other habitats or different feeding traits are required to better rate the potentially high stress resistance towards microplastic pollution in the brown shrimp.

Keywords : clay , Crangon crangon , diatoms , oxidative stress , PLA , PVC , TiO2





### **Coping with synthetic and natural** microparticles:

### brown shrimp and the antioxidant defence

### Introduction

In the environment, plastics are prone to disintegrate into microplastics. The wide size range make microplastics available to different marine fauna, which may mistake them for food items. Apart from synthetic microparticles, marine organisms constantly encounter in their natural habitats various suspended organic (e.g. remains of bivalves, cellulose fibers, chitinous remains) and inorganic (e.g. silica frustules, sediment grains) microparticles. Therefore, the goal of the present study was to investigate whether synthetic and natural particles have similar cellular effects on the brown shrimp Crangon crangon.







HELMHOLTZ

### A method to quantitatively analyse Microplastic transport and retention in an experimental flume environment

Boos Jan-Pascal, Gilfedder Benjamin, Frei Sven.

Rivers and streams are the dominant sources of microplastic (MP) in marine environments. During transport, complex physicochemical interactions between particles, water and river sediments influence particle mobility and retention. The specific transport mechanisms of MP in fluvial systems are not yet fully understood, and the main reason lies in the limitation in reliable data derived from experimental analysis. In our subproject of the 'CRC 1357 Microplastics', we investigate the hydrodynamic mechanisms that control the transport and retention behavior of MP in open channel flows and streambed sediments. In an experimental flume environment, we create realistic hydrodynamic and hyporheic flow conditions by using various porous media (e.g. glass beads or sand) and bedform structures (e.g. riffle-pool sequences, ripples and dunes), modelled from real stream systems. The method developed here can quantitatively analyze the transport and retention of pore-scale particles (1-40  $\mu$ m) based on fluorometric techniques. Particle velocity distributions and particle transport are measured using Particle-Image-Velocimetry and Laser-Doppler-Velocimetry. With our setup, we can quantitatively investigate time-resolved MP transport and retention through the aqueous and solid phase in a flume scale experiment.

Keywords : fluvial systems , hydrodynamic analysis , microplastic transport



### Following the tiniest: How to track Microplastic particles in (sub-) surface water flow [334407]

Jan-Pascal Boos<sup>12</sup>, Benjamin Gilfedder<sup>12</sup>, Sven Frei<sup>1</sup> <sup>1</sup> Department of Hydrology, University of Bayreuth, Germany <sup>2</sup> Limnological Research Station, University of Bayreuth, Germany **CRC 1357 Microplastics** 



**MICROPLASTIC (MP) IN FLUVIAL SYSTEMS** 

Rivers and streams are the dominant pathway for MP input into marine ecosystems.

Pore-scale MP are found in river sediments, but transport mechanisms are unclear: **River** ≠ **pipe**:

- Sedimentation and Resuspension in stream
- Transfer across hyporheic interface with infiltrating stream water
- Physicochemical interactions with water and sediments

Common analysis methods are often destructive and static:

 $\rightarrow$  To investigate MP transport mechanisms, we need spatiotemporal and quantitative information on MP abundances in i) open channel flow and ii) porous media



 hydrodynamic transport (2) sedimentation and buria 3 resuspension

homoaggregation (5) heteroaggregation (6) hyporheic exchange

(7) transport in HZ (8) exchange with aquifer (9) transport in aquifer

### **METHODS**

### Microplastic detection

Spatially averaged measurement (concentration) for small particles  $(1 - 10 \,\mu\text{m})$  with a fluorescence coating:

- Planar measurement (Fluorescence-Camera-System)
- Selective measurements (Fluorometer)

### **Hydraulics**

Optical techniques to avoid disturbing the flow



### **RESULTS: PROOF-OF-CONCEPT**



Migration and formation of microplastic plumes (Colorcoded intensity indicates abundance)

Infiltration of Hyporheic Zone





Left: MP concentration in sediment increases and expands over time Right: Breakthrough curves for MP (in- and outflow) in surface stream show declining concentration (right) in time

### DISCUSSION

Mobility of MP in porous media

Hyporheic flow paths of MP can be visualized Infiltration of MP into Hyporheic Zone is shown

Quantification of exchange processes Linear response for small particles System sensibility is high (recovery rates ~ 90 %)



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### Session 26.9\_Ma. Chaired by Mateo Cordier, Guyancourt

### Microplastic formation from a newly developed biocomposite

Niu Zhiyue, Catarino Ana I, Davies Peter, Le Gall Maelenn, Curto Marco, Jiang Chulin, Dotcheva Mariana, Vancoillie Gertjan, Dhakal Hom, Vandegehuchte Michiel, Everaert Gert. Paper number 334351

**GC/MS-Orbitrap: an application for the characterization of organic plastic additives** *Akoueson Fleurine, Cbib Chaza, Dehaut Alexandre, Duflos Guillaume.* Paper number 334396

**A temporal sediment record of microplastics in Haukadalsvatn, a lake in Iceland** *Ásmundsdóttir Ásta, Gomiero Alessio, Øysæd Kjell Birger, Geyrsdóttir Áslaug.* Paper number 334475

**Insights into the oxidation of microplastics by the Fenton process** *Ortiz David, Munoz Macarena, Carbajo Jaime, De Pedro Zahara M., Casas Jose A.*. Paper number 334420

### Abundance and oceanic source of microplastics: pellets as a model on the middle coast of Rio Grande do Sul, Brazil

*Correa Marina Zimmer, Panizzon Jenifer, Heinzelmann Larissa Schemes.* Paper number 334445 \_MICRO 2020. Fate and Impact of Microplastics: Knowledge and Responsibilities. 23-27 November 2020\_\_\_\_

### Microplastic formation from a newly developed biocomposite

Niu Zhiyue, Catarino Ana I, Davies Peter, Le Gall Maelenn, Curto Marco, Jiang Chulin, Dotcheva Mariana, Vancoillie Gertjan, Dhakal Hom, Vandegehuchte Michiel, Everaert Gert.

Concerns have arisen about the environmental persistence and biological impacts of microplastic (MP) in the global ocean. An important source of MP in the marine environment is the degradation of fossil-based polymers such as polypropylene (PP) and polyethene (PE) induced by the hydromechanical forces and UV radiation. Polymers and composite materials made from a naturalsourced feedstock, like polylactic acid (PLA) and thermoplastic starch (TPS), known as biopolymers and biocomposites, are seen potential alternative with lower environmental impacts. However, to date, few studies have focused on the degradation behavior of biopolymers and biocoposites in the marine environment. As part of the Interreg 2 Seas Mers Zeeën project SeaBioComp (http://seabiocomp.eu/), we compared and quantified the MP formation of a newly developed biocomposite and a fossil-based polymer during their degradation under UV radiation. To do so, we exposed self-reinforced PLA and PP specimens in seawater to UV radiation simulating natural exposure for up to 18 months. To identify and characterize MP particles, we applied a combination of fluorescence microscopy, scanning electron microscopy coupled to an element detection system (SEM-EDX), and infrared technology (µFT-IR). Preliminary results indicate the formation of MP due to UV exposure (ongoing analysis). We anticipate that our results will contribute to assessing the risk of biocomposites which can present a more sustainable alternative to fossil-based polymers.

Keywords : biopolymer , fragmentation , microplastic formation , photodegradation , SeaBioComp , size frequency distribution





### Microplastic Formation from A Newly Developed Biocomposite

Niu Zhiyue\*, Catarino Ana Isabel, Davies Peter, Le Gall Maelenn, Curto Marco, Jiang Chulin, Dotcheva Mariana, Vancoillie Gertjan, Dhakal Hom, Vandegehuchte Michiel and Everaert Gert

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### Background

- Concerns have arisen about the environmental persistence and biological impacts of microplastic (MP) in the global ocean.
- An important source of MP in the marine environment is the degradation of fossil-based polymers such as polypropylene (PP) induced by the hydromechanical forces and Ultra-violet (UV) radiation.
- Polymers and composite materials made from a natural-sourced feedstock, like polylactic acid (PLA) known as biopolymers and biocomposites, are seen potential alternative with lower environmental impacts.
- To date, few studies have focused on the degradation behavior of biopolymers and biocoposites in the marine environment.

### Objective

 To compare and quantify the MP formation of a newly developed biocomposite and a fossil-based polymer during their degradation under UV radiation.

### Methodology

- we exposed self-reinforced PLA and PP specimens in seawater to UV radiation simulating natural exposure for up to 18 months.
- To identify and characterize MP particles, we applied a combination of fluorescence microscopy and infrared technology (µFT-IR)



Figure 1. Graphical scheme of the experimental design.

### Results

Particle identification

- PLA and PP showed similar color under UV filter and blue filter (Fig.2).
- Polymer type was confirmed using µFTIR with library spectra.
- · Recovery test: > 95% of particles detected.



Figure 2. Fluorescent microscope Images of identified PP/PLA debris. A) and C)Image captured under UV filter. B) and D) Image captured under blue filter.

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### Microplastic formation

- Significant number of PP microplastic (>50µm) formed after 1368h UV exposure; while no PLA microplastic formed.
- PP has a higher microplastic generation rate than PLA (Fig.3).
- The frequency of formed microplastic decreased with their size increasing (Fig.4).



**Figure 3.** Number of PLA/PP microplastic (>50 µm) detected against duration of UV exposure (h). The count of microplastic particles (N) from each polymer type is a function of UV exposure (UV) according to a Poisson regression analysis (confidential interval = 95%). The equations of fitted curve are as:  $N_{SR-PP} = e^{2.76+0.00179 \cdot UV}$ ;  $N_{SR-PLA} = e^{2.10+0.000871 \cdot UV}$ .



**Figure 4.** Size frequency distribution of PP microplastic (>50  $\mu$ m) after 1368h UV exposure. The Maximum density is scaled as 1. The Feret's diameter ( $\mu$ m) and area (mm<sup>2</sup>) were quantified using ImageJ.

### Conclusions

- PP are more easily degrede to microplastic than PLA under UV radiation.
- Our results will contribute to assessing the risk of biocomposites which can present a more sustainable alternative to fossil-based polymers

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### GC/MS-Orbitrap: an application for the characterization of organic plastic additives

Akoueson Fleurine, Cbib Chaza, Dehaut Alexandre, Duflos Guillaume.

Environmental pollution from plastics and microplastics (MPs) is a threat to the environment and wildlife on multiple aspects regarding exposition of different hazards. One of them is the impact of the organic plastic additives (OPAs). Various chemical additives can be added to polymers during the manufacturing process to modify and improve their physical properties for example. The analysis of OPAs has shown a growing importance since their use has become controversial as some of the additives were found to be toxic to marine organism and human being. In order to have a better understanding of the chemical impact of these additives, it is important to identify and, ideally quantify the ones included in the polymeric matrices. The analysis of OPAs and polymers is an analytical challenge due to the diversity of their chemical composition and the fact that they are most of the time several into a single plastic. Implementing easy and quick screening analysis methods, able to analyze plastic additives, have become a focus of interest. The goal of this study is to develop such a method, with pyrolysis coupled to a gas chromatography and mass spectrometery (Py-GC/MS) for the identification and, if possible, the quantification of OPAs and, consecutively, of the polymeric matrix they are included in. A first step consisted in constituting a database in GC/MS-Orbitrap with a wide variety of plastic additives. So far, a common method in GC/MS-Orbitrap has been developed for the analysis of various plastic additives with standards solution. The selection of the main additives studied was based on their use and toxicity.

Keywords : antioxidants , Common method , flame retardants , GC/MS , Orbitrap , organic plastic additives (OPAs) , plasticizers

### GC/MS-Orbitrap: an application for the characterization of organic plastic additives

### Fleurine Akoueson<sup>1,2</sup>, Chaza Chbib<sup>3</sup>, Sébastien Monchy<sup>3</sup>, Ika Paul-Pont<sup>4</sup>, Périne Doyen<sup>2</sup>, Alexandre Dehaut<sup>1</sup> & Guillaume Duflos<sup>1</sup>

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- The analysis of organic plastic additives (OPAs) has shown a growing importance since the use of some of them has become controversial. In order to have a better understanding of their chemical toxicity, it is important to identify and, ideally quantify the OPAs included in plastic polymers.
- This is an analytical challenge due to the diversity of molecules, the complexity of polymeric matrices, and the fastidious sample preparation using traditional analysis technique, such as solvent extraction, which can bring biases. In order to integrate these biases and simplify the analysis approach, pyrolysis coupled to a gas chromatography mass spectrometer (Py-GC/MS) can be used.

### Aim & Objective

• So far, a common method in GC/MS-Orbitrap has been developed for the analysis of various plastic additives.

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FATE AND IMPACTS OF MICROPLASTICS: KNOWLEDGE AND RESPONSIBILITIES

- An additive **database** has been built.
- The final goal is to apply this method with Py-GC/MS for the identification of the OPAs in polymeric samples.

### Steps & Methods

### Additives selection

- Based on their use and toxicity (bibliography, ECHA list (2019), EFSA list (2020))
- 64 molecules candidates

Function (number of molecules)	Details	
	Phthalates (20)	
Plasticizers (24)	Adipate (2)	
	Others (2)	
Plasticizora/Antiovidenta (6)	Nonylphenols (6)	
Flasticizers/Antioxidants (6)	Bisphenol A	
Antiovidente (C)	Irganox (3)	
Antioxidants (6)	Others (3)	
UVs stabilizer (5)	Phenols (5)	
	Phosphorous (7)	
	Phosphates (3)	
Flames retardants (21)	Phenols & PBDE (10)	
	Citrate (1)	
Monomers-intermediates (2)	BPS & BPF	

### GC/MS Orbitrap analysis with a single method

- Preparation of stock solutions (from 0.05 to 100μg/mL)
- Liquid injection for GC/MS analysis

Analysis parameters				
Instrument	GC/MS-Orbitrap – TRACE 1300 - DB-5 like column			
Injection T°	300°C			
Oven program	80°C (0.5 min) → 10°C/min → 330°C (1 min)			
Transfer line T°	300°C			
Ion source T°	300°C			
Scan ranges	33-750 m/z			
Injection volume	1 μL			
Injected [C]	0.32 μg/mL to 5 μg/mL			
Cut-Off	5 min			

### Creation of our own library

- Deconvolution (TraceFinder software)Selection of guantitative and
- qualitative ions



### Results



### Identification of OPAs



### Antioxidants – Irganox®



### Conclusion

- A single method in GC/MS-Orbitrap showed its performance to analyze multiple additives used for plastic production. A high resolution mass spectra database of about sixty additives was created and provide help to characterize the chemical compounds contained included in plastic.
- GC/MS-Orbitrap coupled with pyrolysis was successfully used to identify several phthalates with the same method directly from plastic samples. These preliminary result are promising for the ability of Py-GC/MS to analyze plastic additives with simple sample preparation.

### Further work

- A method using GC/MS-Orbitrap coupled with pyrolysis is under development and will be widely applied to identify the main plastic additives.
  - Quantitative development should also be implemented.



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**A temporal sediment record of microplastics in Haukadalsvatn, a lake in Iceland** Ásmundsdóttir Ásta, Gomiero Alessio, Øysæd Kjell Birger, Geyrsdóttir Áslaug.

Research has found that plastic pollution is being deposited into the fossil record with contamination increasing dramatically since the 1940s. It has been suggested that the plastic layers could be used to identify the start of the Anthropocene, the proposed geological epoch in which human activities have come to dominate the planet. In this study a well-dated sediment core collected from lake Haukadalsvatn in west Iceland, covering the whole of the Holocene, was used to provide data on the historical accumulation trend of microplastic waste. Samples were analysed from the top section of the sediment core dated from 1958 to 2002 CE as 5 years' time series. Microplastics were extracted from the sediment layers by a sequence of enzymes and oxidizing purification steps followed by ZnCl2 dense-liquid separation. The size class distribution and the chemical characterization were performed by µ-FTIR microscopy followed by GCMS-pyrolysis. Polyethylene, polypropylene, and polystyrene microplastic particles were among the most dominant identified polymers. A total of 12 polymers where identified. Proliferation of microplastics is evident in the core from the late 1960s to the present. Relatively low numbers of particles were found in older sediments or comparable to laboratory blanks. This study shows that robustly dated sediments can add an important temporal perspective to our global understanding of microplastics sources, transport pathways, distribution and temporal trends.

Keywords : Iceland , microplastics , pristine lake , sediments , temporal sediment record



### A temporal sediment record of microplastics in Haukadalsvatn, Iceland

Ásta Margrét Ásmundsdóttir<sup>1</sup> Alessio Gomiero<sup>2,\*</sup>, Kjell Birger Øysæd<sup>2</sup>, Áslaug Geirsdóttir<sup>3</sup>

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### Introduction

The atmospheric transport of microplastics (MPS) is still poorly understood but data exist indicating the capability of MPS to travel by air (Allen et al.). Data regarding the deposition of MPS in Sub-Arctic/Arctic areas is seriously lacking. The team capitalize on the fact that Icelandic lake basins act as sinks, preserving valuable information on climate and environmental changes through time due to the continuous, highresolution sediment accumulation (Geirsdóttir et al., 2019). The exceptionally secure age models depend on the high sedimentation rates, radiometric age determination (210Pb/137Cs) and the well-known historical tephrochronology of Iceland. Analysis of the sediment's organic geochemistry provides proxy information on pollutants occurrence evolution within the catchment and in the lakes. In the presented study the team analyses MPS in dated sediment layers of lake Haukadalsvatn, with the aim to identify the first MPS in Icelandic environment as well as providing important information on the time related deposition rates, size and polymer type distribution



### Materials & methods

Sampling and age determination: The lake (3,2 km<sup>2</sup> max depth 40m) is located the west of Iceland (Fig. 1), 140 km northwest from Reykjavík. A sediment core (diameter: 7 cm, length: ~1m in) was obtained in the year 2003 following Glew (1991), to capture the sediment-water interface and undisturbed upper sediments plus the last approximately 100 years (Fig. 2; Geirsdóttir et al., 2009). The sediment core was sampled every 2.5 cm for 210Pb and nearly every centimeter through the key time interval for 137Cs. 210Pb was measured by alpha spectrometry (Eakins and Morrison 1976). Unsupported 210Pb calculated by subtracting supported activity from the total activity measured at each level and dates determined according to the constant-rate-of-supply model (Appleby 2001). 137Cs activity was measured using an Ortec-EGG high-purity, germanium crystal well, photon detector coupled to a digital gamma-ray spectrometer. Maximum deposition of 137Cs associated with above ground nuclear bomb testing occurred during the period 1963-1964. Tephra layers were identified visually within the sediment cores; tephra from key horizons were taken for analysis by electron microprobe CAMECA SX50. The geochemical composition of the tephra layers was then used to identify the source volcano and eruption according to Jóhannsdóttir (2007) and Harning et al. (2019).

MPS analysis: 51 sub sections (2,5 cm) of the initial core were obtained and pooled to obtain 11 composite sediments samples, covering a time series of ≈ 10 years. This allowed processing ≈. 100 gr per sample. MPS were extracted by a density separation with ZnCl<sub>2</sub>, filtration through 1 µm stainless steel filter, multi-step procedure based on combined enzymatic and oxidizing treatments (Gomiero et al., 2019). Enzymes from Sigma were filtered on 0.7 µm (Whatman™ GF/F ™) and stored in sterile containers. The obtained purified extracts where deposited on a ZnSe window and scanned by a Nicole iN10 Infrared Microscope (ThermoFisher). Dust traps were used to evaluate possible contamination from airborne particles and procedural blanks were analyzed as part of the QA/QC procedures



### Results

The first 2,5 cm layer was dated as 2002 and the last (at 49,51 cm depth) as 1865. A total number of 22 polymer types were characterized. The earliest occurrence of plastic material identified in the composite sample covering the time span 1942 - 1952 were celluloid (CL) particles and rayon's fibers (RA). On a later stage in the sediments dated as from 1953-1964, the appearance of Bakelite (BaK) particles, polyvinyl acetate (PVA) and polyvinyl chloride (PVC) is recorded (Fig. 3). Since 1974, nylon (Ny), polyethylene (PE), cellulose acetate (CeA), polystyrene (PS) and polypropylene (PP) made their stratigraphic appearance. While the latest entry polymer types such as polyurethane (PU), polyacrylates (PCy) and polyethylene terephthalate (PET) appeared in the 1986-1992 composite sediment sample. Overall, on average 62% of the characterized particles were within 40 and 60 µm size range while the smallest were 20 µm (the limit of detection) and the largest 197 µm (Fig.2). The total number of particles progressively increased through time starting by 0,3 particles/g (DW) in the 40's up to 9 particles/g (DW) in the 90's.



### **Discussion and Conclusions**

The observed polymers' appearance sequence corresponds with the start of global mass production, use and waste-generation of plastic (Zalasiewicz et al. 2016; Geyer et al. 2017). Once accumulated within sedimentary strata, plastic particles are likely to have a variable, but generally good preservation potential. The results of the present work indicate that plastic particles are already present in enough numbers to be considered as one important permanent record of human presence on Earth. The current advances in sediments temporal trend analysis and polymer type chemical characterization enables the assessment of plastics in sedimentary strata in lake sediments in sparsely populated Arctic/Sub-Arctic regions. This offers a unique opportunity to investigate the temporal trend in plastics distribution as well as changes in the deposition rates. The support of imaging FTIR includes the far more informative infrared region of the spectrum from the very onset of the analysis and enables the detection of particles down to 20 µm, which are likely overlooked by visual inspection and therefore not included in the majority of the previously published studies in the field.

### References

### Insights into the oxidation of microplastics by the Fenton process

Ortiz David, Munoz Macarena, Carbajo Jaime, De Pedro Zahara M., Casas Jose A..

The widespread presence of microplastics (MPs) in aquatic ecosystems constitutes one of the most challenging environmental issues nowadays. The small size of these solids coupled with their low chemical and biological reactivity makes their removal from aqueous matrices particularly complex. Although advanced oxidation processes (AOPs) have been widely studied for the treatment of persistent contaminants in waters, there are only a few works focused on MPs degradation. In this study, a homogeneous and heat-activated Fenton treatment has been proposed to evaluate the changes that MPs can suffer along this oxidation process. Polystyrene (PS) particles (100-250 um). obtained from commercial plastic trays by cryogenic milling, and commercial glitter were used as MPs. The degradation experiments (5 days) were carried out at 80°C and pH 3 in a glass reactor under constant stirring (200 rpm). The reactor volume and MP mass were established at 75 mL and 100 mg, respectively. The initial concentrations of Fe3+ and H2O2 were set at 10 mg/L and 1000 mg/L, respectively. To enhance the oxidation yield, three additional H2O2 doses (75 g/L) were added per day once complete consumption of H2O2 was reached. In the same line, an additional dose of Fe3+ (750 mg/L) was also added once a day (with the first H2O2 dosage). The images taken by optical microscopy revealed slight surface changes in the studied MPs, especially in PS, where semitransparent regions were appreciated due to the decrease in the thickness of the particle in these areas. Furthermore, an analysis of the FTIR spectra revealed small changes after Fenton treatment, denoting the presence of new functional groups. Nevertheless, the mass loss observed was relatively low (5-10%), which indicates that MPs mainly undergo surface changes. These modifications can modify the MPs hydrophobicity and thus, the Fenton treatment could improve their further separation from water.

Keywords : Fenton oxidation , glitter , microplastic , polystyrene , water treatment







### 23-27 NOVEMBER 2020 LANZAROTE AND REYOND

### Insights into the oxidation of microplastics by the Fenton process

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### Abundance and oceanic source of microplastics: pellets as a model on the middle coast of Rio Grande do Sul, Brazil

Correa Marina Zimmer, Panizzon Jenifer, Heinzelmann Larissa Schemes.

Coastal regions can be affected by plastic litter even far from urban centers as a result of sources as wind, waves and tides. Since the pellets is one of microplastics widely observed on coastal sand surfaces, and considering it as a consequence of industrial spillage, our research aimed to evaluate the oceanic contribution to plastic particle pollution at coastal areas distant from cities and industrial plants. Pellets collecting were conducted in the 30 km middle coastline of Rio Grande do Sul, Brazil. Collecting methodology based on UNEP protocols consisted in active visual survey held by two collectors within three 5mX15m surface square plots marked between the dunes base and water line. Samplings were performed along three southern hemisphere seasons (spring of 2019, summer and autumn of 2020) and three different locations inside the total area, 15 km apart from each other. A one-way ANOVA was conducted to compare pellet abundance among the collecting points and among seasonal collect. A total of 342 pellets (0.17 pellets/m<sup>2</sup>) were collected in three sampling surveys, a unusual small amount to Brazilians coastal areas. The amount of pellets varied between the three points.

Keywords : Plastic pollution , Sandy beaches , Seasons.

. 3, p. 325-332, 2015.	mmission, 2009, 117 p. , Brazil. RGCI, Lisboa , v. 15, n.	vernmental Oceanographic Cor esin pellets in Salvador, Bahia,	<b>References:</b> INEP/IOC. UNEP/IOC Guidelines on Survey and Monitoring of Marine Litter. Nairobi: United Nations Environment Programme/Intergover TERNANDINO, Gerson et al. How many pellets are too many? The pellet pollution index as a tool to assess beach pollution by plastic res
	ets 57 pellets s/m <sup>2</sup> 0.08 units/m <sup>2</sup>	79 pelle 0.12 unit	55 pellets 40 pellets 0.08 units/m <sup>2</sup> 0.06 units/m <sup>2</sup> More campling is necessary to establish which abintic factors relate to seas
5	206 pellets 30 units/m² Autumr	Summer 0.	P2 247 pellets P1 0.36 units/m <sup>2</sup> P3
een sampled on seems to	differences betwe at pellet depositio year.	are no statistical $\Rightarrow$ suggests that nuous along the y	The amount of pellets varied between the three points (p<0.01) There a Higher amounts of pellets were observed at point 2, followed by seasons point 1 (p=0.02) and point 3 (p<0.01). be continu
	coastal areas.	Isual for Brazilian	Total of 342 pellets (0.17 pellets/m <sup>2</sup> ) were collected $\rightarrow$ a small amount unus
Oc	Sand	eas and among	distributed in three different points (P1, P2 and P3) over 30 km of coast; • One-way ANOVA test $\rightarrow$ to compare pellets abundance both among areaseasonal sampling.
ean	dunes	<ul> <li>Prazil → very</li> <li>venters;</li> <li>umn of 2020;</li> <li>of 5 m X 15 m</li> </ul>	<ul> <li>Surveys were conducted at the middle litoral of Rio Grande do Sul state,</li> <li>ow human density, absence of industrial activities and located far from urbar</li> <li>Three sampling effort were performed along spring 2019, summer and autu</li> <li>Active visual survey was carried out by two collectors in three square plots</li> </ul>
			MATERIAL AND METHODS
n to plastic at remote icators.	ceanic contributio on composition sing pellets as indi	To evaluate oc particle pollutic coastal areas us	Coastal regions can be affected by plastic litter, even far from urban centers, as result of sources like wind, waves and tides; One of microplastics widely found at marine coastal sand surfaces is pre- production pellets, the raw material which plastic is moulded by industry.
	OBJECTIVE		INTRODUCTION
	ELLETS AS A AZIL I, Brazil	PLASTICS: PE DO SUL, BRA leinzelmann <sup>1</sup> b, Rio Grande do Sul	334445 - ABUNDANCE AND OCEANIC SOURCE OF MICROP MODEL ON THE MIDDLE COAST OF RIO GRANDE I Marina Zimmer Correa* <sup>1</sup> , Jenifer Panizon <sup>1</sup> , Larissa Schemes He <sup>1</sup> Feevale University - ERS-239, 2755, Vila Nova, 93525-075, Novo Hamburgo, *marinazimmercorrea@gmail.com

Day 5, Friday 27 <sup>th</sup> . November 2020						
9h-10h	27.1_O	27.1_Ma	27.1_Me			
10h-10h15	27.	1_Gaia: 3 session	s brief			
10h30-11h30	27.2_O	27.2_Ma	27.2_Me			
11h30-11h45	27.	2_Gaia: 3 session	s brief			
12h-13h	27.3_O	27.3_Ma	27.3_Me			
13h-13h15	27.3_Gaia: 3 sessions brief					
14h-15h	27.4_O 27.4_Ma 27.4_Me					
15h-15h15	27.4_Gaia: 3 sessions brief					
15h17-15h30	this isthe ENDsee you in 2022OUTDOOR					

### Day 5/5, Friday 27th November 2020
### Session 27.1\_O. Chaired by Patricia Ostiategui, Gran Canaria

### Microplastics in coral reefs : a two years survey in the Maldives

Saliu Francesco, Montano Simone, Seveso Davide, Lasagni Marina, Galli Paolo. Paper number 329480

## Plastic ingestion by four seabird species in the Canadian Arctic: Comparisons across species and time

*Baak Julia, Provencher Jennifer, Mark Mallory.* Paper number 332988

## Microplastic exposure and uptake by juvenile white seabream *Diplodus sargus* (Linnaeus, 1758) in a coastal lagoon nursery ground

Müller Carolin, Erzini Karim, Cruz Joana, Dudeck Tim, Santos Corona Luana, Abrunhosa Felipe Eloy, Afonso Carlos Manuel Lourenço, Mateus Miguel Ângelo Franco, Monteiro Pedro, Ekau Werner.

Paper number 334263

### Plastic ingestion in Mediterranean seabirds: characterization and temporal trends

Serrallonga Sara, Frias-Perez Joan, Rodríguez Andrea, Militão Teresa, García Salvador, Sanchez-Vidal Anna.

Paper number 334473 Please note there's an additional author: Jacob González-Solís

### Elucidating marine biofouling recruitment on biodegradable polymers augmented with oyster shell filler

Audrezet Francois, Pochon Xavier, Von Ammon Ulla, Floerl Oliver, Le Guen Marie-Joo, Trochel Branwen, Zaiko Anastasija. Paper number 334232

### Microplastics in coral reefs : a two years survey in the Maldives

Saliu Francesco, Montano Simone, Seveso Davide, Lasagni Marina, Galli Paolo.

The impacts of microplastics (MP) on reef ecosystems are still larger unknown. Evidences from lab feeding trials that MP may be ingested by reef-building corals and cause adverse effects, i.e. necrosis and bleaching were recently collected, but ön fieldstudies are still limited. For this reason, we established in our research center located in the Faafu Atoll (Maldives) several research campaigns devoted to highlight possible correlations between the raising presence of microplastics in the reef envronments and the health of the related marine organism . The vulnerability of the species selected for the biomonitoring prompt us to develop alternative analytical approaches to lower the impacts of the research, i.e we extensively implemented the use of solid phase microextraction as non lethal and solvent free sampling method. Surveys carried out till now on the subsurface water, shore sediments and marine invertebrates showed that MP are concentrated mostly inside the atoll rims and influenced by the Monsoons. Moreover, a positive correlation between the presence of MP in the subsurface water and of pthalates in the surveyed organisms was observed.

Keywords : Coral Reef , Maldives , Marine invertebrates , Phthalates

# Plastic ingestion by four seabird species in the Canadian Arctic: Comparisons across species and time

Baak Julia, Provencher Jennifer, Mark Mallory.

Plastic pollution ingestion by seabirds is an increasing environmental problem even in remote areas such as the Arctic, yet knowledge on plastic pollution ingestion by several Arctic seabirds is limited, making it difficult to assess trends. We examined plastic pollution ingestion by northern fulmars (Fulmarus glacialis), black-legged kittiwakes (Rissa tridactyla), thick-billed murres (Uria lomvia) and black guillemots (Cepphus grylle) in the Canadian Arctic to assess species-specific and temporal differences in plastic ingestion over ten years. Seventy-two percent of fulmars and 15% of kittiwakes ingested plastic, while guillemots and murres did not. The number and mass of plastic ingested by fulmars decreased between the two periods (2008 and 2018), but the frequency of occurrence of plastic ingestion did not change, although sample sizes were less than ideal. Future research with larger samples is recommended to reinforce these trends in plastic ingestion by Arctic seabirds.

Keywords : Arctic, marine debris, plastic accumulation, plastic pollution, seabird

# Microplastic exposure and uptake by juvenile white seabream *Diplodus sargus* (Linnaeus, 1758) in a coastal lagoon nursery ground

Müller Carolin, Erzini Karim, Cruz Joana, Dudeck Tim, Santos Corona Luana, Abrunhosa Felipe Eloy, Afonso Carlos Manuel Lourenço, Mateus Miguel Ângelo Franco, Monteiro Pedro, Ekau Werner.

Estuaries and lagoons are vital nursery grounds for a variety of commercially important fish species. vet these essential habitats lie at the gateway of plastic pollution to the global oceans. Due to their size range (; 5mm), microplastic (MP) fibres and fragments entail a high bioavailability for a wide range of organisms at the base of the marine food web, among them early life stages (ELS) of fish, representing the bottleneck of population development. As ELS of seabream show a high site fidelity within their nurseries, it is hypothesized that they encounter a gradient of habitat quality and MP pollution across small spatial scales, potentially allowing for MP ingestion at an early developmental stage and consequent physiological implications. Though the number of published studies on MP ingestion across various fish taxa continuously increases, more emphasis needs to be devoted to indepth research on the contextualization of MP ingestion as well as on the potential negative effects of these pollutants on ELS of individual species in coastal environments. Thus, the aim of this field study was to investigate differences in fish growth and condition of juvenile white seabream Diplodus sargus in the Ria Formosa lagoon (Southern Portugal) along spatial gradients of prey availability, habitat quality and MP pollution. The analyses of samples are currently underway; preliminary results however suggest rather low MP uptake rates despite an omnivorous, opportunistic feeding mode of juvenile white seabream. Further analyses are expected to shed light on the potential existence of a spatial gradient in MP abundance as well as on the bioavailability of MP in relation to natural prey items. The selectivity of juvenile seabream towards MP as an artificial prey will be determined and potential physiological impacts of MP exposure and uptake will be assessed.

Keywords : early life stages of fish , ingestion , microplastics , seabream

# Plastic ingestion in Mediterranean seabirds: characterization and temporal trends

Serrallonga Sara, Frias-Perez Joan, Rodríguez Andrea, Militão Teresa, García Salvador, Sanchez-Vidal Anna.

Plastic, the main component of marine litter, is now ubiguitous in the marine environment. Ingestion of floating plastics is an emerging threat to seabirds, particularly for those already threatened. It is estimated that around 40% of the seabird species ingest plastic, which may cause wounds, intestinal blockage, decrease in feeding capacity, starvation and even death. Here, we aim to characterize the temporal trends and types of plastics ingested by seabirds in the Mediterranean Sea. For that, we assessed the plastic ingestion of 386 individuals from 10 species of Mediterranean seabirds: 113 Scopoli's (Calonectris diomedea), 68 Mediterranean (Puffinus velkouan) and, 74 Balearic (P. mauretanicus) shearwaters, 42 Audouin's (Ichthyaetus audouinii), 12 Mediterranean (I. melanocephalus), 12 black-headed (Chroicocephalus ridibundus) and 43 yellow-legged (Larus michahellis) gulls, 4 black-legged kittiwakes (Rissa tridactyla), 16 Northern gannets (Morus bassanus) and 2 great skuas (Catharacta skua). Furthermore, we characterized the plastic polymer found using a Fourier-transform infrared spectrophotometer (FTIR). Seabirds included in the study were accidentally caught by longliners operating through the Catalan Coast, in the Western Mediterranean Sea from 2003 to 2014. Our results support recent findings of a higher occurrence of plastic ingestion in the three shearwaters than in the remaining study species. Scopoli's shearwaters showed higher occurrence (73.5%) and larger number of plastics (on average 7.0  $\pm$  11.7) than Balearic (40.5%,  $2.7 \pm 1.4$ ) and Mediterranean shearwaters (38.2%,  $3.1 \pm 4.8$ ). The number of plastics ingested by shearwaters decreased from 2003 to 2014, but the total plastic mass accumulated did not change. The main plastic components found were polyethylene and polypropylene and did not differ among species. The proportion of each of these polymers resembled those found in surface waters of the NW Mediterranean. Thus, our results highlight the sensitivity of shearwaters to plastic ingestion and their suitability as sentinels for monitoring floating plastic pollution.

Keywords : Mediterranean seabirds , Plastic characterization , Plastic ingestion , Temporal trends

### Elucidating marine biofouling recruitment on biodegradable polymers augmented with oyster shell filler

Audrezet Francois, Pochon Xavier, Von Ammon Ulla, Floerl Oliver, Le Guen Marie-Joo, Trochel Branwen, Zaiko Anastasija.

Marine plastic debris (MPD) impacts on marine ecosystems is among the most important environmental concerns of the past decades. Virgin-plastic manufacture is often cheaper than recycled plastics, which increases the rate of plastic release in the environment. Along with other environmental effects, MPD can serve as vector for marine hitchhikers, facilitating spread of unwanted organisms. In this context, there is a growing demand in eco-friendly replacements to conventional plastic polymers, ideally with fit-for-purpose properties and a well understandable life cycle. In response to this demand, biodegradable polymers made of, or augmented with biological material are being considered and tested as a future alternative to plastics. In this experimental study, formulated biopolymer compounds with different concentrations of oyster shell filler were tested to determine: (i) their degradation rate in the natural marine environment; (ii) preferential recruitment of a non-indigenous tunicate Styela clava; (iii) temporal evolution and peculiarities of bacterial and eukaryotic communities associated with the different shell concentration in polymers. Composites were deployed at Nelson Marina (New Zealand) in January 2019 and dynamics of biofouling formation and biodegradation was observed over an 18-week period. Here we present the results of this investigation and outline further steps of this research.

Keywords : Biodegradable polymers , Biosecurity , invasive species , Marine environment , Metabarcoding , Plastisphere

### Session 27.1\_Me. Chaired by Sonja Oberbeckmann, Rostock

## Role of organic substances adhered on surface of polystyrene microplastics in the urban river of Mongolia

Battulga Batdulam, Kawahigashi Masayuki, Oyuntsetseg Bolormaa. Paper number 332824

### **Paint particles are a distinct and variable substrate for marine bacteria** *Tagg Alexander, Oberbeckmann Sonja, Labrenz Matthias, Fischer Dieter, Kreikemeyer Bernd.* Paper number 334175

## Marine biodegradability of bio-based and petroleum-based polymers as substitutes of conventional microbeads

Ghiglione Jean-François, Cheng Jingguang, Boris Eyheraguibel, Ter Halle Alexandra, Bruzaud Stéphane, Meistertzheim Anne-Leïla. Paper number 334301

**Meso and microplastics as an ecosystem: The plastisphere from a brazilian estuary** *Pinheiro Lara, Agostini Vanessa, Pinho Grasiela.* Paper number 334422

## Role of organic substances adhered on surface of polystyrene microplastics in the urban river of Mongolia

Battulga Batdulam, Kawahigashi Masayuki, Oyuntsetseg Bolormaa.

Plastic debris is a pervasive pollutant in marine environment released from terrestrial origin through river transport. Evaluating behavior of plastic debris especially microplastics (MPs) in river environment is vital to assess risks of plastic contamination with respect to their physical and chemical characteristics in river ecosystem. Since microplastics (MPs) have become long-lasting anthropogenic debris, the current study aimed to identify occurrence of natural organic substances on polystyrene microplastics (PS-MPs) and to evaluate behavior of the PS-MPs based on the surface property of PS-MPs in river environment. Samples of PS-MPs were collected from the river shores along the Tuul River in the urban area of Ulaanbaatar, Mongolia. Samples were digested with mixture of hydrogen peroxide (30%) and iron (II) sulfate for further spectroscopic analyses. The spectroscopic analysis was carried out for digested and non-digested samples by micro-Fourier transform infrared spectroscopy to obtain organic functional groups and surface photodegradation status of plastic particles using carbonyl index (CI). A wide spectrum of organic functional groups was identified from the samples, indicating that growth and metabolism of microorganisms promotes formation of biofilms on the surface of PS-MP particles based on the environmental factors. The CIs in the samples revealed surface chemical deterioration of PS-MPs by solar radiation before microbial growth on PS-MPs. The values of CIs could be reflected the weathered surface of PS-MPs in the environment with further potential to aggregate with other plastic particles and contaminants in the environment mediated by microbial biofilms. Owing to variation in residential time of plastic particles in the environment, PS-MPs are prone to interact with contaminants as vectors for pollutants in the aquatic environments during the development of biofilms followed by formation of MP aggregates. The fate and behavior of plastic particles and associated pollutants can be greatly influenced by development of biofilms in aquatic environments.

Keywords : biofilm , carbonyl index , organic substances , polystyrene microplastics (PS–MPs) , urban river

### Paint particles are a distinct and variable substrate for marine bacteria

Tagg Alexander, Oberbeckmann Sonja, Labrenz Matthias, Fischer Dieter, Kreikemeyer Bernd.

While paint particles are an important part of the microplastic sphere, they have, as yet, received much less research coverage, particularly regarding microplastic-microbiological interactions. This study investigated the biofilm communities of a variety of paint particles from brackish sediment using 16S rRNA gene sequencing. Paint particle biofilm communities appear to be distinct from natural (water and sediment), non-synthetic particle (cellulose) and common microplastic biofilm communities. Notably, there appears to be 1 group of sulphate-reducing bacteria from the Desulfobacteraceae family, Desulfatitalea tepidiphilia, that dominate certain paint biofilms. Of the 8 investigated paint-associated communities, four paints displayed this high Desulfobacteraceae presence. However, it is currently unclear from the chemical analysis performed of the paint surface chemistry (ATR FT-IR spectroscopy, Raman spectroscopy, SEM-EDX) what the drivers behind this might be. As such, this study provides important insights as the first to analyse microplastic-paint biofilm communities and paves the way for future research.

Keywords : 16S, Biofilm, Desulfatitalea tepidiphilia, Desulfobacteraceae, Microplastic, Sediment

## Marine biodegradability of bio-based and petroleum-based polymers as substitutes of conventional microbeads

Ghiglione Jean-François, Cheng Jingguang, Boris Eyheraguibel, Ter Halle Alexandra, Bruzaud Stéphane, Meistertzheim Anne-Leïla.

The European parliament voted to ban single-use plastic items in 2021 and transition to bio-economy includes the substitution of these banned products by biodegradable polymers. Conventional plastic microbeads classically used as abrasive agents and excipients in cosmetics contribute worldwide to more than 4% of primary microplastics discharged each year into the marine environment, i/e. between 10,900 and 38,300 tonnes / year. Here, we tested the potential of the marine "plastisphere" to biodegrade microbeads composed of petroleum-based polymers (PCL, PMMA), bio-based (PHBV, PLA) and natural products (rice, apricot) as compared to conventional PE-based polymer. After colonization of the microbeads by a microbial biofilm in circulating aquariums open to the sea for 2 months (Banyuls Bay, Mediterranean), their biodegradation was studied for an additional 2 months after transfer in a minimum medium with microbeads as the only carbon source. We used a multidisciplinary approach including changes in the physical properties (particle size, molecular weight) and chemical properties (production of oligomers) of the microbeads. Changes in microbial abundance, diversity and activity were explored by confocal microscopy, flow cytometry, Illumina sequencing, heterotrophic prokaryotic activity, energy production (ATP) and respiration (O2). This multidisciplinary approach showed efficient biodegradation of the biobased PHBV and the petroleum-based PCL, whereas biobased PLA did not show sign of biodegradation in our marine conditions. We were able to identify specific OTUs directly linked with biodegradation activities proved by the coupled chemical and biological tools. Metagenomic analysis are currently under analysis that will help to decipher the polymer degradation mechanisms under marine conditions. Finally, this work led to the development of a ministerial decree and our protocol was the subject of a SOLEAU envelope.

Keywords : biodegradation , biofilm , exfoliating microbeads , microbial ecotoxicology , Plastisphere

# Meso and microplastics as an ecosystem: The plastisphere from a brazilian estuary

Pinheiro Lara, Agostini Vanessa, Pinho Grasiela.

Plastics serve as substrate for microorganism (e.g. bacteria, protozoans, fungi and microalgae) colonization in aquatic environments, forming the Plastisphere. The Plastisphere in meso and microplastics from water and sediment collected in a salt marsh in the Patos Lagoon estuary (Brazil) was studied. For plastics in water, 150mL of water was filtered (0.45µm), and particles were selected from the filter under a microscope. For the sediment surface, plastics were manually sampled using tweezers in four zones: high marsh, middle marsh, low marsh and mudflat. All particles were analysed under a scanning electron microscope for attached microorganisms and degradation marks. Plastics (1.25-10.5mm in size) were found as fragments, pellets, films and fibres. In water, only fibres were found, and biofouling occurred in patches (not covering the entire surface). Microorganisms were found on all particles, including filamentous cyanobacteria, single-celled round- and rod-shaped bacteria, pennate and centric diatoms, and filamentous fungi, and yeasts. Broken diatom frustules were found mainly at the higher zones salt marsh. At the high marsh, biofilms also followed a patchy pattern on plastic surfaces, while in the mudflat and low marsh they were covering most of it. This might be associated with water availability, as lower zones are mostly flooded while higher zones are drier, which can cause desiccation of the fouling community. Also, fibres seemed to be less covered by microorganisms than other formats, which can be related to their smaller surface area available for colonization. Several degradation marks could be observed in the plastics surface in the high marsh, which can be a result of the less contact with water i.e. higher sunlight exposure. The patchy colonization pattern in that area could also favour photodegradation as biofilms can act as a protection against sunlight. Implications of biofouling in plastics remain to be studied in this area.

Keywords : biofilm , biofouling , colonization , salt marsh , substrate , zonation.

### Session 27.1\_Ma. Chaired by Lisa Devriese, Oostende

**Effects of long-term exposure to microfibers on ecosystem services provided by coastal mussels** *Christoforou Eleni, Dominoni Davide, Lindstrom Jan, Stilo Giulia, Spatharis Sofie.* Paper number 332450

The potential of fluorescence dyes – Comparative study of Nile red and three derivatives for the detection of microplastics *Schuhen Katrin, Sturm Michael.* Paper number 331980

# Sources of microplastic pollution and amounts of microplastics in zoobenthos and fish in the Baltic Sea

Buhhalko Natalja, Turov Polina, Lind Kati, Lips Inga. Paper number 333678

### Benthic fauna contribute to microplastic sequestration in coastal sediments

Coppock Rachel, Cole Matthew, Lindeque Pennie, Queirós Ana, Galloway Tamara, Näkki Pinja, Birgani Hannah, Richards Saskiya. Paper number 334168

# Effects of long-term exposure to microfibers on ecosystem services provided by coastal mussels

Christoforou Eleni, Dominoni Davide, Lindstrom Jan, Stilo Giulia, Spatharis Sofie.

Coastal environments are increasingly exposed to microplastic pollution. It is known that large microplastics impair the filtration capacity of bivalves, which provide the invaluable ecosystem service of alleviating the effect of coastal eutrophication through their biofiltration mechanism. However, the effect of smaller microplastics, and specifically microfibers, is still unknown despite the fact that they are the most common microplastics in the coastal marine habitats and similar in size to phytoplankton, the main food source of bivalves. Hence, in this study we investigate the effects of long-term exposure to microfibers (<100µm) on the biofiltration capacity of the blue mussels, *Mytilus edulis*. We found that long-term exposure to microfibers significantly reduced the clearance capacity, of *Tetraselmis* sp. phytoplankton, by mussels. Furthermore, a wide range of microfiber quantities were found in the digestive gland of mussels but, the most efficient phytoplankton consumers were more susceptible to microfibers could negatively impact the biofiltration capacity of the more potent individuals, thus decreasing the ecosystem service provided by the population.

Keywords : Coastal ecosystems , Ecosystem services , Microfibers , Particle selection , Phytoplankton clearance

# The potential of fluorescence dyes – Comparative study of Nile red and three derivatives for the detection of microplastics

Schuhen Katrin, Sturm Michael.

For an appropriate risk assessment of microplastics in the environment and food, it is essential to know the levels of microplastic contamination. Extensive research has been carried out in the field of microplastic detection in recent years. While common methods such as Raman spectroscopy and pyrolysis GCMS are time consuming and require trained personnel and expensive equipment, there is a need for an inexpensive and easy to use method. Staining microplastics with the Nile red (NR) fluorescent dye has great potential to meet these criteria because the only requirement is a device for fluorescence imaging [1, 2]. In our work, we tested NR and newly developed derivatives to achieve greater selectivity for plastic particles and more intense fluorescence. In addition, the influence of the use of different solvents and water at different pH values in the dyeing process was examined by analyzing the fluorescence spectra of solid samples of colored microplastics and natural particles. Finally, a standardized method was developed method developed from the knowledge acquired and tested on sea salt and wastewater. The results will be presented in our talk.

Keywords : Fluorescence dye , Fluorescence Microscopy , Microplastic detection , Nile red derivatives

# Sources of microplastic pollution and amounts of microplastics in zoobenthos and fish in the Baltic Sea

Buhhalko Natalja, Turov Polina, Lind Kati, Lips Inga.

Microplastic (MP) contamination is becoming a significant challenge with the burgeoning use of plastic. The ubiquitous presence of the MPs in the sea (Baltic Sea) has raised care about their possible environmental impacts. Our research aims were to quantify the MPs accumulated in the open and coastal area environments, including water, sediments and biota. The potential impact of different land-based sources on marine biota was analysed. The amount, distribution, shape, color and polymer types of MPs in different matrices and the potential contribution of wastewater treatment plants (WWTP) and rivers to the MPs pollution in coastal areas were studied. The WWTPs can be considered as a main source of the MPs in the Estonian waters. There is an increase in the pollution level in the city with a higher population (Tallinn). Such sources as ports, rainwater, rivers flowing through the city and beaches also play an important role in the pollution of the sea with MPs. MPs were found in 24% of analysed benthic fish and just in 8% of pelagic fish. 43% of juvenile fish contained MPs vs 20.5% of adult fish. The most abundant polymer in fish guts was polyethylene.

Keywords : Baltic Sea , fish , microplastic , pollution , zoobenthos

### **Benthic fauna contribute to microplastic sequestration in coastal sediments**

Coppock Rachel, Cole Matthew, Lindeque Pennie, Queirós Ana, Galloway Tamara, Näkki Pinja, Birgani Hannah, Richards Saskiya.

Microplastics are ubiquitous in the marine environment, however, the mechanisms governing their uptake by, and burial within, seabed habitats are poorly understood. In this study, microplastic burial and its impact on fauna-mediated sedimentary processes was quantified at three coastal sites, and the potential contribution of burrowing faunal communities to this process assessed via functional trait diversity analysis of field data. In addition, laboratory exposures were used to assess whether sediment-processing undertaken by the brittlestar Amphiura filiformis, a key species in the sampled area, could explain the burial of microplastic fibres. Field observations confirmed broad-scale burial of microplastics across the coastal seabed, consistent across sites and seasons, with microplastic sequestration linked to benthic-pelagic exchange pathways, driven by burrowing fauna. Brittlestars were observed to bury and line their burrow walls with microfibres during experiments, and their burial activity was also modified following exposure to nylon fibres, relative to controls. Collectively, these results indicate that biodiverse and functionally important seabed habitats act as microplastic sinks, with burrowing fauna contributing to this process via well-known benthic-pelagic pathways, the rates of which are modified by plastic exposure.

Keywords : benthic , burial , fate , functional traits , pelagic

### Session 27.2\_O. Chaired by Rachid Dris, Créteil

Similar size of microplastics and algae as food supply affects life history traits of two freshwater rotifers species *Drago Claudia*. Paper number 334193

### Particle size and hydrologic influence on microplastic accumulation in streambed sediments downstream of a municipal point source

Margenat Henar, Drummond Jennifer D., Nel Holly A., Krause Stefan, Stonedahl Susa H., Sabater Francesc. Paper number 334250

**Prospective study on macroplastics and microplastics within dredging sediments** *Constant Mel, Alary Claire, Billon Gabriel, Dumoulin David, De Waele Isabelle, Moreau Myriam.* Paper number 334308

# Similar size of microplastics and algae as food supply affects life history traits of two freshwater rotifers species

Drago Claudia.

The large quantities of plastic particles reported from lakes and rivers are the cause of an increasing number of microplastics studies in freshwater environment. Microplastics ingestion by freshwater biota has been demonstrated but the potential harm caused by microplastics is not clear vet. Rotifers are important primary consumers in freshwater environment, linking the primary producers and higher trophic consumers. Filter – feeding organisms such as rotifers are unselective and thus highly vulnerable to microplastics exposure. In this study we evaluate the effect of different sizes and types of microplastics (1-, 3- and 6- µm PS microspheres and 5-25 µm PA fragments) on two life history traits : population growth rate and reproduction, of two cosmopolitan rotifers: Brachionus calyciflorus and Brachionus fernandoi. The different treatments were combined with high and low concentrations of algae as food, in order to simulate optimal and stressed natural conditions. A decrease in intrinsic growth rate and reproduction was found in B. calyciflorus exposed to 3- µm PS microspheres in combination with high and low concentration of food algae. Contrastingly, B. fernandoi showed decreasing growth rate just when exposed to low concentration of food in combination with 3- µm PS microspheres. Smaller and larger PS microspheres (1-, 6-, µm) had weaker but not significant effects and the PA fragments had no effects on intrinsic growth rate and reproduction of both rotifers species. Additionally, we tested different concentrations of 3- µm PS microspheres in combination with varied food algae species. The ratio between food algae and microplastics, as well as the size and nutritional quality of food algae, may have a key role to understand the indirect effects of microplastics on rotifers.

Keywords : brachionus , intrinsic growth rate , microplastics , polyamide , polystyrene , reproduction , rotifer

## Particle size and hydrologic influence on microplastic accumulation in streambed sediments downstream of a municipal point source

Margenat Henar, Drummond Jennifer D., Nel Holly A., Krause Stefan, Stonedahl Susa H., Sabater Francesc.

Wastewater treatment plants (WWTP) act as a point source of microplastics (MP, defined as 1 - 1000um) to freshwater ecosystems, releasing over 5 million MPs/dav/WWTP to streams. Previous studies have shown a correlation between MP abundance and high-density population areas, but less is known on how river hydrodynamics play a role in MP accumulation in streams. We investigated the spatial distribution of microplastics within sediments every 15 m downstream of a WWTP effluent in Cànoves stream (Montseny, Catalonia) during Spring 2019. We compared measurements to an upstream-control site and another site near to the WWTP bypass that added untreated wastewater when discharge exceeded capacity. The 450 m reach consisted of three geomorphically altered reaches interspersed between three unaltered buffer reaches, each 75 m. During baseflow conditions, we measured per site MP abundance, length, area, particulate organic matter quantity, grain size distribution, and local hydrologic conditions (velocity, depth, and width). MPs were quantified following the Nile Red fluorescence method and characterized as large (¿ 64 µm) or small (10 to 64 um). MPs in sediment samples were mainly fragments with a higher abundance of small MPs vs. large MPs, 150 particles/g vs. 22 particles/g, respectively. We found that both small MP abundance and size was negatively correlated with distance from the point source, suggesting a combination of fragmentation of larger particles and/or preferential filtration of larger particles along the stream. A negative correlation of large MPs with distance from the WWTP was not found unless the bypass was included, suggesting these infrequent inputs are an important source of MPs to the stream, especially the larger MPs. Greater MP abundance was observed in the areas with increased organic matter and smaller sediment grain sizes. These results demonstrate the complex size-dependent microplastic transport processes that determine accumulation patterns in streambed sediments.

Keywords : hydrologic influence , particle size , pattern distribution , WWTP

**Prospective study on macroplastics and microplastics within dredging sediments** Constant Mel, Alary Claire, Billon Gabriel, Dumoulin David, De Waele Isabelle, Moreau Myriam.

Compared to ecotoxicological studies and the fate of microplastics in the ocean, only few studies had investigated the presence of microplastics within terrestrial ecosystems (Rochman, 2018). However, 80 % of marine microplastics could come from land, and soils/sediments may represent an important sink for microplastics (Jambeck et al., 2015; Rillig, 2012). To fill this gap, microplastics were collected within a dredging disposal area belonging to VNF (Voies Navigables de France, Navigable Waterways of France), along the Aa River (St-Omer, France). Four pits were dug, and samples were taken at four depths with a metal trowel. A part of them were directly sieved (10 and 5 mm) on site. to collect macroplastics. The other parts were brought back to the laboratory, where microplastics were extracted using a gravity separation method (NaI, 1.6 g mL-1). Microplastics were observed under a dissecting stereo-microscope ( $6\times$ ,  $12\times$ ,  $25\times$  and  $50\times$  magnification), separated into 5 shape categories (fibers, fragments, micro-beads, films and foams) and analyzed to determine their plastic nature with a FTIR spectrometer. Preliminary results indicated that these sediments are widely contaminated, with concentrations ranging from 1 to 77 macroplastic per kilogram and between 1 and 2822 microplastics per kilogram. No clear patterns were observed between sediment features and plastic concentrations. Films were the most abundant shape observed within the pool of macroplastics, whereas microplastics were mainly fibers and fragments. FTIR results indicated that macroplastics were mainly made of polyvinyl chloride (PVC), polyethylene (PE) and polypropylene (PP). Among microplastics fibers, polyamide, polyester and PVC were the most abundant polymer, while other shapes were PVC and PE.

Keywords : dredging sediment , Macroplastics , Microplastics

### Session 27.2\_Me. Chaired by Jean-François Ghiglione, Banyuls

**Plastic habitats: insights into algal biofilm formation on PET, PP and PVC in a freshwater lake** *Stanton Tom, Law Antonia.* Paper number 333218

**Can nanoplastic particles pass through drinking water treatment plants?** *Pulido-Reyes Gerardo, Mitrano Denise M., Kaegi Ralf, Von Gunten Urs.* Paper number 334276

### Biogenic calcite precipitation facilitates the sedimentation of microplastics in a eutrophic reservoir

Leiser Rico, Jongsma Rense, Insa Bakenhuis, Möckel Robert, Philipp Bodo, Neu Thomas R., Wendt-Potthoff Katrin. Paper number 334345

**Potential role of metal precipitation on plastic fate: transport and degradation** *Rogers Kelsey, Carreres-Calabuig Joan, Keulen Nynke, Shashoua Yvonne, Posth Nicole.* Paper number 334500

# Plastic habitats: insights into algal biofilm formation on PET, PP and PVC in a freshwater lake

Stanton Tom, Law Antonia.

Plastic materials are integral to contemporary societies. However, at the end of their life plastic products that are not disposed of responsibly can pollute the environment. The negative impacts of plastic pollution are well documented and vary with plastic polymer and type. However, in addition to the physical and chemical environmental threats posed by plastic waste, plastic in the environment can also provide a habitat for some organisms. This work documents the colonisation of plastic materials by diatom and algal communities in benthic and photic lake environments over six weeks of submersion. It finds that biofilm communities on plastic litter differ depending on position within the lake (photic and benthic), and that the development of the algal assemblage varies with plastic polymer (Low Density Polyethylene, Polyethylene Terephthalate, and Polypropylene). Moreover, SEM imaging of these plastic surfaces following submersion identified surface degradation of plastic litter, highlighting the potential for plastic degradation after four weeks. The extent of this degradation also varied between the plastic polymers under investigation. This research highlights the role of plastic waste as a habitat for algal communities, with implications for freshwater carbon fixation and microplastic generation. The recorded differences between polymer type suggest that the ability of different plastic polymers to function as habitats for algal communities varies in lake environments, as does the algal assemblages they can support. Moreover, the presence of these biofilms could act as a catalyst for the breakdown of plastic polymers into microplastics.

Keywords : Algae , Biofilm , Degradation , Diatoms , Lake Environments

**Can nanoplastic particles pass through drinking water treatment plants?** Pulido-Reyes Gerardo, Mitrano Denise M., Kaegi Ralf, Von Gunten Urs.

Microplastics have been detected in both potable water sources and bottled water, leading to questions about the efficacy of current water treatment practices. It is hypothesized that all water resources will experience an increasing influx of particulate plastic down to the nanometer dimensions as degradation products of larger plastic items. Ozonation, sand and activated carbon filtration are key treatment processes in drinking water treatment plants, yet studies addressing the impacts on and the retention efficiency of nanoplastics by these treatment steps are still lacking. Therefore, we evaluated the impact of ozonation on the physicochemical properties of metal doped nanoplastic particles (polystyrene/polyacrylonitrile, 215±1 nm diameter) and the effectiveness of sand/activated carbon filtration for nanoplastic removal. For this purpose, nanoplastics were treated with ozone in Lake Zurich water (source for drinking water production in Zurich). The nanoplastics remained colloidally stable with unchanged diameter. However, the particle surface charge decreased with exposure to increasing ozone doses. Pristine and aged sand and activated carbon, used in the treatment plant and thus coated with a biofilm, were evaluated as filtration media. Results from colloid column transport studies showed a higher degree of particle retention in used sand in comparison with pristine sand, with 92% and 42% of nanoplastic retention, respectively. However, an opposite trend was found for columns packed with activated carbon where the highest level of particle retention was obtained with pristine materials in contrast with columns packed with used activated carbon. Using the data from used substrates, a filter length of 0.9 and 2.7m would be required for reaching a 99% of nanoplastic removal for sand and activated carbon filters, respectively. In order to ensure that these estimated values can be transferred to real drinking water treatments plants, pilot plant experiments are currently being conducted in collaboration with the Water-Works of Zurich.

Keywords : activated carbon , drinking water treatments , filtration systems , Nanoplastics , ozonation , pilot plant , sand

# **Biogenic calcite precipitation facilitates the sedimentation of microplastics in a eutrophic reservoir**

Leiser Rico, Jongsma Rense, Insa Bakenhuis, Möckel Robert, Philipp Bodo, Neu Thomas R., Wendt-Potthoff Katrin.

Low-density microplastics are frequently found in sediments of many lakes and reservoirs. The processes leading to settling of initially buoyant polymers are poorly understood for inland waters. This study investigated the impact of biofilm formation and aggregation on the density of buoyant polyethylene microplastics. Biofilm formation on polyethylene films (4 x 4 x 0.15 mm) was studied in a eutrophic reservoir (Bautzen, Saxony, Germany). Additionally, aggregation dynamics of small PE microplastics (85 µm) with cyanobacteria were investigated in laboratory experiments. During summer phototrophic sessile cyanobacteria (Chamaesiphon spp. and Leptolyngbya spp.) precipitated calcite while forming biofilms on microplastics incubated in Bautzen reservoir. Subsequently the density of the biofilms increased sinking of roughly 10 % of the polyethylene particles within 29 days of incubation. In the laboratory experiments planktonic cyanobacteria (Microcystis spp.) formed large and dense cell aggregates under the influence of elevated Ca2+ concentrations. These aggregates enclosed microplastic particles and led to sinking of a small portion (0.4%) of polyethylene microplastics. This study showed that both sessile and planktonic phototrophic microorganisms mediate processes under the influence of calcium which facilitate densification and sinking of microplastics in freshwater reservoirs. Loss of buoyancy leads to particle sedimentation and could be a prerequisite for the permanent burial of microplastics within reservoir sediments.

Keywords : biofouling , calcite , calcium , cyanobacteria , reservoirs , sinking

**Potential role of metal precipitation on plastic fate: transport and degradation** Rogers Kelsey, Carreres-Calabuig Joan, Keulen Nynke, Shashoua Yyonne, Posth Nicole.

Plastic debris exposed to coastal marine conditions displays not only the attachment of biofilm, but also a significant mineral deposition on the surface. The presence of mineral deposits and mineralcoated biofilms on the plastic surface yields new hypotheses regarding plastic fate. Metal mineral precipitation on plastic may affect several important factors influencing environmental plastic fate including transport, degradation, and biofilm development. Mineral precipitation influencing plastic mass, and for smaller particles changing aggregation behavior, would in turn influence plastic transport in water and porewater. Furthermore, precipitation could enhance abiotic degradation via processes such as metal catalysis, or form a protective layer shielding the plastics from further degradation. Additionally, community composition may be altered based on the various minerals present on plastics, which could increase the selectivity of microbes on the plastic surface. Here we discuss our current and future work on this topic and present evidence of metal mineral precipitation on plastics from SEM-EDS images of polyethylene and polystyrene exposed for 12 months in situ to the water column and sediments of Svanemøllen Marina, Copenhagen, Denmark. These plastics were further analyzed for mineral characterization using metal extraction with ICP-OES as well as for degradation using FTIR. Enhanced precipitation and degradation was observed on plastics exposed to the more oxygenated water column and upper sediments, indicating the importance of redox conditions and geochemical environment to the metal mineral precipitation process.

Keywords : biotic and abiotic loading , degradation , FTIR , in situ experiment , metals , SEM

### Session 27.2\_Ma. Chaired by François Galgani, Bastia

## Solving the non-alignment of methods and approaches used in microplastic research in order to consistently characterize risk

Koelmans Albert, Redondo Hasselerharm Paula, Mohamed Nor Nur Hazimah, Kooi Merel. Paper number 332481

## How fast, how far: method diversification for measuring microplastic in environmental samples?

*Rist Sinja, Hartmann Nanna, Welden Natalie.* Paper number 334540

## Routine analysis of samples from freshwater environments using the GEPARD software and Raman microspectroscopy

Fischer Franziska, Brandt Josef, Kanaki Elisavet, Bittrich Lars, Lenz Robin, Ivar Do Sul Juliana, Labrenz Matthias, Wick Natalie, Krause Steffen, Schaum Christian, Fischer Dieter. Paper number 334616

# Solving the non-alignment of methods and approaches used in microplastic research in order to consistently characterize risk

Koelmans Albert, Redondo Hasselerharm Paula, Mohamed Nor Nur Hazimah, Kooi Merel.

The lack of standard approaches in microplastic research caused deficiencies in the comparability of data across methods used, locations, biota and environmental compartments. This limits the development of monitoring and risk assessment frameworks, which in turn limits progress in the abatement of plastic pollution. An obvious solution is harmonization of methods, which is ongoing and crucial, however is slow and laborious. We suggest that some of the most pressing problems can also be reduced by pragmatic rescaling methods that are able to improve the alignment of methods used in microplastic research. First, we describe a method to correct for the differences in size ranges as used by studies reporting microplastic concentrations, and demonstrate how this reduces the variation in aqueous phase concentrations caused by method differences. Second, we provide a method to convert number, volume and mass concentrations into one another using probability density functions that represent environmental microplastic. Finally, we use this method to correct for the incompatibility of data as used in current species sensitivity distributions (SSDs), caused by differences in the microplastic types used in effect studies and those in nature. We derived threshold effect concentrations from such a corrected SSD for freshwater species. Comparison of the rescaled exposure concentrations and threshold effect concentrations reveals that the latter would be exceeded for 1.5% of the known surface water exposure concentrations of surface waters worldwide. Altogether, this tool set allows us to correct for the diversity of microplastic, to address it in a common language, and to assess its risks as one environmental material.

Keywords : analysis , microplastic , risk assessment , species sensitivity distributions

## How fast, how far: method diversification for measuring microplastic in environmental samples?

Rist Sinja, Hartmann Nanna, Welden Natalie.

A diverse array of methods exist to quantify microplastic (MP) in environmental samples, however, the proliferation of new methods has accelerated in recent years. This has resulted in a wide variety of complimentary but also competing approaches to MP sampling, extraction and characterisation. While there is an obvious need for a suite of methods for different purposes, for example the precision required by research and the efficiency needed in monitoring, it has repeatedly been criticised that data are not comparable. Furthermore, the degree to which new methods are acknowledged and adopted and the reasoning behind it are currently unclear. We therefore need to determine the relative usefulness of the increasing number of available methods versus an aggravation of comparability. A systematic review of method use in the MP literature was undertaken, covering five common processing steps: sampling of water for MP, MP extraction from water samples, MP extraction from sediment samples, MP extraction from biota, and MP characterisation. By approaching the literature in reverse chronological order, we identified the first occurrence of a method. New methods were categorised by their "degree of novelty" into: highly novel methods, secondary adaptations of existing methods and smaller, tertiary adaptations of existing methods. Apparent interest in new methods was determined by assessing the average number of citations per month of each publication. The proportional use of methods across all articles as well as the use of reporting units are also reported. Based on this dataset, we 1) assess the rate of method diversification, 2) identify key developments and points of diversification, and 3) analyse the level of interest in new methods and their apparent spread and adoption. The implications of method selection (accuracy, efficiency, and impacts on reporting and comparability) are highlighted and recommendations specific to both academic research and environmental monitoring are given.

Keywords : characterization , extraction , methodology , Sampling

## Routine analysis of samples from freshwater environments using the GEPARD software and Raman microspectroscopy

Fischer Franziska, Brandt Josef, Kanaki Elisavet, Bittrich Lars, Lenz Robin, Ivar Do Sul Juliana, Labrenz Matthias, Wick Natalie, Krause Steffen, Schaum Christian, Fischer Dieter.

Analysis of microplastic particles with a combination of vibrational spectroscopy and particle detection allows for obtaining the most detailed information regarding a sample's microplastic particle (MP) content. That includes MP quantification and material type identification as well as the determination of particle shapes and colors, which can be relevant for determining an MP's source, or for evaluating its biological impact. For particle-based approaches, the number of particles in a sample poses a considerable challenge. For particles ; 500 µm, common particle counts for environmental samples lie in the range of several thousand (riverine water) to hundred thousand particles (sewage sludge) per sample, which multiplies when analyzing a set of samples. This task demands automated analysis routines. To perform analyses in a comparable as well as time- and personnel-efficient way, the GEPARD (Gepard Enabled PARticle Detection) software1 was developed in our group. GEPARD is open-source and can be coupled to Raman or Fourier Transform Infrared (FTIR) microscopes, where it allows for running a highly automated combination of optical particle detection and Raman or FTIR measurements. It provides tools to handle the optical image acquisition, particle detection, spectroscopic measurement and data analysis seamlessly. The presentation will show the GEPARD-based analysis workflow, which our lab has applied to more than 100 samples from various environmental compartments over the past three years using a WITec alpha 300R Raman microscope. It covers the steps from filtering pre-treated samples onto measurement substrates in a contamination-minimizing environment to creating a summarized result output that includes the size, shape and color of all MP found. In addition, example analyses of freshwater and wastewater treatment plant samples concerning MP ¿ 50 µm will be shown. 1 Brandt, J. et al. (2020) 'High-Throughput Analyses of Microplastic Samples Using Fourier Transform Infrared and Raman Spectrometry', Applied Spectroscopy. doi: 10.1177/0003702820932926.

Keywords : GEPARD , Raman microspectroscopy , riverine water , wastewater treatment plant

### Session 27.3\_O. Chaired by Erik van Sebille, Utrecht

#### Uncertainties on plastic concentration estimates at sea

Mercier Matthieu, Ter Halle Alexandra, Simatos Florian, Poulain-Zarcos Marie, Saint-Martin Marion.

Paper number 334456

**Vertical distribution of microplastics in upper-ocean turbulence: Laboratory modelling** *Poulain-Zarcos Marie, Mercier Matthieu, Ter Halle Alexandra.* Paper number 334467

### From land to seabed: uncovering the fate of plastic in the ocean

Navarrete Fernandez Teresa Magdalena, Morales-Caselles Carmen, Viejo Josue, Hernandez Ignacio, Gonzalez-Gordillo J Ignacio, Cózar Andrés. Paper number 334492

**Fate of floating plastic debris released along the coasts in a global ocean model** *Chenillat Fanny, Huck Thierry, Maes Christophe, Grima Nicolas, Blanke Bruno.* Paper number 334565

### Uncertainties on plastic concentration estimates at sea

Mercier Matthieu, Ter Halle Alexandra, Simatos Florian, Poulain-Zarcos Marie, Saint-Martin Marion.

The large difference between the estimates of global plastic input in mass in the oceans (Jambeck et al., Science 347, 2015) and current global predictions from numerical models (van Sebille et al., Environ. Res. Lett. 10, 2015) or observations (Cózar et al., P. Natl. Acad. Sci., 111, 2014) is one of the most important issue regarding oceanic plastic litter. Yet, global predictions are based on observations, and uncertainties on the latter are rarely considered to provide error bounds on the former. We discuss here the sources of uncertainties on plastic concentrations estimates (in number and mass), based on a recent model presented in (Poulain et al., Environ. Sci. Technol. 53, 2019). The two main sources of error are the plastic rise velocity and the model for the turbulent diffusivity, although they do not have the same importance. We validated the model with controlled laboratory experiments. Applying this model to global predictions provides us with more realistic encompassing values for the mass of plastic at sea, much larger than the current estimates.

Keywords : error estimates , laboratory modelling , Microplastics , sea sampling corrections

# Vertical distribution of microplastics in upper-ocean turbulence: Laboratory modelling

Poulain-Zarcos Marie, Mercier Matthieu, Ter Halle Alexandra.

Global estimation for plastic waste in the ocean is about 200,000 pieces/km2, which represents only 1% of annual plastic inputs (Jambeck et al., 2015). Amongst the several explanations that could justify this discrepancy, the precision of current modelling tools could be one, with improvements required to better reproduce plastic concentrations at sea surface (Law et al., 2014; Brach et al., 2018) or in depth. Models for the vertical transport are based on a balance between the upward buoyant flux and the downward turbulent one due to waves and wind (Kukulka et al., 2012). Our aim is to improve these models taking into account the particles shape and size (Poulain et al., 2019), the surface turbulence properties and the fluid-particle coupling. We present here an experimental approach focused on the dynamics of nearly-buoyant particles, of different sizes (from 0.5mm to 6mm), dispersed in a turbulent flow induced by a vertically-oscillating grid (whom intensity decreases with depth). First, we characterize the turbulence, using PIV measurements, to estimate the depth profile of turbulent quantities, to ultimately get the eddy viscosity profile. Second, we study the dynamics and the transport of nearly buoyant particles in such flow. We then compare our results with previous studies on tracers (Kukulka et al., 2012) and heavy particles (Michallet & Mory, 2004); and discuss how to extend our results to global plastic pollution.

Keywords : laboratory modelling , Transport of microplastics , turbulent flow

### From land to seabed: uncovering the fate of plastic in the ocean

Navarrete Fernandez Teresa Magdalena, Morales-Caselles Carmen, Viejo Josue, Hernandez Ignacio, Gonzalez-Gordillo J Ignacio, Cózar Andrés.

From the estimated 4.8- 12.7 million MT of plastic litter that enters the ocean every year, only 0.3 million MT of it is floating on the surface, whereas the rest goes missing. The unbalances in the size distribution of floating plastics also supports the idea of an unquantified sink of litter in the ocean. The striking scarcity of millimetre-sized fragments on the sea surface has been explained by a combination of mechanisms diverting fragments into the water column and the seabed. However, the load of microplastic on the ocean floor at a large scale is unknown. In this study, we analyze the abundance of microplastics in 188 sediment samples from 45 sites in the Mediterranean Sea. By compiling a sufficiently extensive data sets, we found a sharp decrease in the microplastic concentrations with increasing distance from land sources. Although multiple environmental factors control the spatial distribution of microplastic on the seafloor, the distance to the main sources of mismanaged plastic waste was by far the main driver for the microplastic concentration at the macroscale.

Keywords : marine litter , microplastic , population , seabed , size distribution

**Fate of floating plastic debris released along the coasts in a global ocean model** Chenillat Fanny, Huck Thierry, Maes Christophe, Grima Nicolas, Blanke Bruno.

Marine pollution from plastics is a global issue that infests the ocean from coastal regions to the open sea. The pathway and fate of plastic debris in the oceans are still uncertain for many reasons, including a misperception of its sources, both in terms of quantity and distribution. Understanding the pathway and fate of plastic debris remains fundamental to better manage and reduce plastic pollution. In our study, we diagnose the fate of floating plastic pollution discharged along the coasts by comparing two different types of sources in the global ocean: one based on rivers and the other based on the population density along the coasts. We use a Lagrangian numerical analysis in a global ocean circulation model with a resolution of  $1/12^{\circ}$ . In both scenarios, approximately 6 million particles are released each month for 23 years of simulation. To study the pollution of floating plastics, we force the particles to travel only in the surface layer. Particles may experience a different fate: particles may either end up in the ocean or on the coast after traveling in the open ocean, or they may never leave the coast and instead move along the coastline. This study first underlines the importance of the input scenario on the contribution of particles to the main convergence zones. Even more interestingly, this study shows that the input scenario plays a key role on the number of beached particles that end up in several coastal areas. Beachings occur mainly locally, but a significant number of particles can travel several thousand kilometers before reaching coastal areas, allowing long-distance connectivity between two remote regions. In summary, some coastal regions contribute strongly, locally or remotely, to the pollution of specific convergence zones and coastal areas.

Keywords : Coastal pollution , Lagrangian analysis , Marine Debris , Microplastics , Numerical model , Ocean connectivity , Ocean surface pathways

### Session 27.3\_Me. Chaired by Juan Baztan, Crozon

Impact of mechanical abrasion as a main driver for (micro)plastic fragmentation in the near shore environment – Methods and first results of experiments combining wave-breaking, sediment-particle-, and particle-particle collision

Born Maximilian P., Brüll Catrina, Schüttrumpf Holger. Paper number 334191

**Microplastics on the beaches of the Jericoacoara National Park (Ceará, Brazil)** *Teixeira Luan Fokin, Prado Beatriz, Labuto Geórgia, Semensatto Décio.* Paper number 334486

## Microplastic pollution varies spatially and seasonally in water and sediment from the Guarapiranga Reservoir, São Paulo (Brazil)

Semensatto Décio, Gerolin Cristiano, Labuto Geórgia. Paper number 334490

**Distribution of microplastics in subtidal sediments at the Arrábida coast, Portugal** *Rodrigues Diana, Sobral Paula, Costa Maria Helena.* Paper number 334595

### **Impact of mechanical abrasion as a main driver for (micro)plastic fragmentation in the near shore environment – Methods and first results of experiments combining wave-breaking, sediment-particle-, and particle-particle collision** Born Maximilian P., Brüll Catrina, Schüttrumpf Holger.

Several studies have shown that the prevailing forces at a shore environment, UV-irradiation and mechanical abrasion, are largely responsible for the fragmentation of plastic waste into particles smaller than five millimetres, so-called microplastic. However, UV-irradiation varies throughout the seasons and can be neglected during winter in northern latitudes. Thus, newly littered plastic is then barely degraded but rather abraded by wave action and sediment. Since only very few studies considered wave energy in their plastic-fragmentation experiments and mostly in short termed tests, this study focuses on long term tests with low intensity wave action, sediment-plastic-particle-, and particle-particle-collision regarding the fragmentation rate of PE-HD, PET, and EPS. For the realisation of a proper testing environment a "Slosh-Box" was used consisting of three boxes with 450x300x200 mm<sup>3</sup> (L/W/H). The boxes are filled with a sediment layer of approximately ten millimetres of different grain sizes (0.25-0.5; 0.5-1.0; 1.0-2.0 [mm]) and 1.5 litres of saltwater. They are mounted onto a swivel table powered by a motor causing an oscillation of  $\pm 12^{\circ}$  and up to 26 whole movements (angular work of 48° per movement) of the swivel table per minute, resulting in waves and wave breaking, respectively. The tests are then run up to 90 days. First results have shown that especially EPS spherules fragment to a great extent already after 30 days. Up to 25% of the spherules' weight and 0.64 mm<sup>3</sup> in volume per square centimetres surface and day was abraded in this time into smaller microplastics. PE-HD and PET on the other hand are quite resistant against this mechanical influence, resulting in a weight loss of only 0.1% per 30 days for PE-HD and 0.4% per 30 days for PET. Further analytics such as MSQ and SEM will be conducted to classify the impacts more detailed once the tests are completed.

Keywords : fragmentation , long term , mechanical abrasion , microplastics , saltwater , sediment , wave breaking , waves
**Microplastics on the beaches of the Jericoacoara National Park (Ceará, Brazil)** Teixeira Luan Fokin, Prado Beatriz, Labuto Geórgia, Semensatto Décio.

The Jericoacoara National Park (Ceará, Brazil) receives thousands of tourists to visit some of its beaches and natural attractions. The park covers 8850 ha of natural dunes, lagoons, and beaches. Next to the park is the village of Jericoacoara, which is outside the protected area. The village hosts most of the tourists and offers leisure activities and local commerce. We collected superficial sediment samples (25x25 cm, 2 cm deep) at the upper limit of the intertidal zone on beaches in the village and the park to estimate microplastic pollution (<5 mm). The village beach was sampled at three points (center and extremities) and the park beaches at six points (three to the east and three to the west of the village). Each point comprises longitudinal triplicates 10 m apart. The number of microplastics ranged from  $1116 \pm 461$  to  $60196 \pm 9994$  parts/kg sed dw (praia do pescador, inside the park, and village, respectively). The volume of microplastics ranged from  $1.0 \pm 0.5$  to  $152.7 \pm 234.9$ mm3 plast/kg sed dw (duna do pôr-do-sol, inside the park, and village, respectively). The village beach showed a higher concentration of microplastics both in volume and in the number of particles than in the park beaches (F=20.48; p<0.05). The volume of microplastics in the village is 14 to 86x higher than the volume found on the park beaches. The number of particles in the village exceeds 28 to 43x registered in the park. Considering the regional prevailing currents and winds, our results indicate that the village is the main source of microplastic pollution on the beaches to the west of the park, whereas the beaches to the east receive particles from other cities. Besides, local touristic and fishing activities on the coast also contribute to the introduction of plastic waste on the beaches.

Keywords : beach pollution, Microplastics, protected areas, sediment pollution

# Microplastic pollution varies spatially and seasonally in water and sediment from the Guarapiranga Reservoir, São Paulo (Brazil)

Semensatto Décio, Gerolin Cristiano, Labuto Geórgia.

We assessed microplastic (63-1000 µm) pollution in sediments and superficial and bottom water samples during the dry and rainy seasons (Oct/2018 and Mar/2019, respectively) from the Guarapiranga Reservoir (Metropolitan Region of São Paulo, Brazil), which supplies drinking water for 3.8 million people. The concentration of microplastics varied spatially and seasonally, where the higher concentrations observed near the heavily urbanized areas and during the dry season. Water column concentrations ranged from 150-3100 particles/m3 water and 0.07-25.05 mm3 plastic/m3 water (dry season), 70-7900 particles/m3 water and 0.06-4.57 mm3 plastic/m3 water (rainy season). Sediment samples were collected only during the rainy season, and concentration ranged from 210-22999 particles/kg dw and 0.15-111.46 mm3/kg dw. Microplastics concentration was generally lower in the rainy season, but it seems that the higher reservoir level exerted a considerable influence by changing the residence time and compensating by "dilution" the higher input of microplastics during the rainy season. Therefore, we suggest investigating the relationship between microplastic flux, microplastic concentration, and residence time to advance knowledge on microplastic pollution dynamics in lentic environments. Moreover, our results may contribute to creating and improving local monitoring programs regarding microplastic pollution and adopting specific public policies.

Keywords : Microplastics , sediment quality , water quality

#### Distribution of microplastics in subtidal sediments at the Arrábida Coast, Portugal

Rodrigues Diana, Sobral Paula, Costa Maria Helena.

Understanding local distribution and diversity of microplastics (MP) in subtidal sediments is crucial to assess the availability of such particles for ingestion by marine organisms and to identify the potential pollution sources upon which is urgent to act towards the prevention of MP inputs in the region. The strong anthropogenic pressure from Sesimbra and Setúbal municipalities (Portugal), along with the multiple activities (industries, fishing and tourism) taking place on the Arrrábida coast together with the proximity of a marine protected area, makes a relevant case study for MP pollution research. Temporal and spatial distributions of MP on the seabed were assessed monthly at six stations along the Sado river estuary and the Professor Luiz Saldanha Marine Park. Sediment was collected with a Petite Ponar grab, at the 5 m isobath. Granulometry, loss on ignition and MP:meiobenthos ratio was determined for each sample. MP were extracted with a zinc chloride solution (1.5 g cm-3), after removing the organic matter with 10% hydrogen peroxide from each sample. A total of 4738 MP was registered, and 7 types of MP were identified: fibers, fragments, filaments, beads, glitter, films, and tangled ball of fibers. The majority of MP consisted of fibers (44%) and fragments (34%). Preliminary results suggest that MP concentration tends to decrease in stations further away from the Sado estuary (1347.2  $\pm$  804.1 particles kg-1 dry sediment; mean  $\pm$  SD) where sediments were finer and contained more organic matter; MP concentrations were higher in October (702.8 ± 933.4 particles kg-1 dry sediment; mean ± SD), decreased till January and increased again in February. Beads, glitter, and tangled ball of fibers were rarely found outside of the Sado estuary, suggesting that the finer estuarine sediment may act as a sink for some types of MP.

Keywords : Arrábida coast , distribution , marine protected area , microplastics , MP:meiobenthos ratio , Sado river estuary , sediments

#### Session 27.3\_Ma. Chaired by Bruno Tassin, Marne la Vallée

#### The life-story of plastic butter tubs

Delorme Astrid, Verney Vincent, Askanian Haroutioun, Roussel Erwann, Voldoire Olivier, Delor-Jestin Florence, Koumba Gaelle Bissagou, Peiry Jean-Luc. Paper number 334238

### Microplastic contamination in the ragworm *Hediste diversicolor* from the Seine Estuary (France)

Revel Messika, Chatel Amélie, Mouloud Mohammed, Perrein-Ettajani Hanane, Métais Isabelle, Bruneau Mélanie, Yakovenko Nadiia, Le Roux Romuald, Caley Timothy, Dreanno Catherine, El Rakwe Maria, Mouneyrac Catherine. Paper number 334488

**Analyzing bacterial community structures associated with different materials in freshwater** *Al-Omari Jafar, Szabo Istvan, Szerdahelyi Soma Gábor, Szoboszlay Sandor.* Paper number 334585

#### Electric clothes dryers: An underestimated source of microfiber pollution

Miller Rachael, Kapp Kirsten. Paper number 334901

#### The life-story of plastic butter tubs

Delorme Astrid, Verney Vincent, Askanian Haroutioun, Roussel Erwann, Voldoire Olivier, Delor-Jestin Florence, Koumba Gaelle Bissagou, Peiry Jean-Luc.

In this work, plastic debris has been introduced into the environment to be exposed to various environmental conditions such as moisture, heat, sunlight or microbial action. Prolonged exposure to these environmental factors action causes polymers to degrade, fragment into smaller pieces and cleaved into small molecules. We investigate the degradation pathways of five identical plastic butter tubs made of polypropylene (PP), positioned in five different locations near a French wild river, in which one was buried in sediments (after having being artificially pre-weathered in lab conditions). two were placed by the riverbank, and two in the vegetation. At each location UV radiation, moisture and temperature were monitored and samples of each plastic tub were taken frequently for 506 days. Changes in the chemical structure were followed with rheology by studying the viscoelastic properties of the polymer as a function of time. Despite, being located in different places which resulted in different UV-radiation exposure and temperature variations, the plastic butter tub followed similar trends, in that after an initial decrease in the viscosity of the polymer, we observe an increase in the overall viscosity after a longer exposition to environmental conditions (506 days) which suggests that a chain recombination mechanism is occurring within the polymer. This is a rather unusual observation since PP is known to, generally, undergo chain scission when it is aged, as indicated by a lowered viscosity. The conclusion is that such experiments in natural conditions may be very instructive to understand the fate of abandoned plastic wastes in the environment. However, a more robust methodology must be built-up with more samples to be able to assess longer times.

Keywords : degradation mechanism , freshwater plastic , plastic , polymer , rheology , SEM , weathering

# Microplastic contamination in the ragworm *Hediste diversicolor* from the Seine Estuary (France)

Revel Messika, Chatel Amélie, Mouloud Mohammed, Perrein-Ettajani Hanane, Métais Isabelle, Bruneau Mélanie, Yakovenko Nadiia, Le Roux Romuald, Caley Timothy, Dreanno Catherine, El Rakwe Maria, Mouneyrac Catherine.

The Seine estuary is a highly anthropized environment, for which it is necessary to use in situ approaches, in order to measure the level of contamination in estuarine organisms. Many reports have identified microplastics (MP) in aquatic environments and studied their accumulation and effects in various organisms. However, there is a lack of information regarding their presence in estuarine species. The aim of our study was to evaluate the presence of plastic particles in the ragworm Hediste diversicolor sampled from the mudflats of the Seine estuary (France) during April and June 2017 and 2018, on two locations characterized by high anthropogenic pressures (S1 and S2) and one of them with an important hydrodynamism (S2). For each sampling campaign, particles were analyzed after digestion of pools of two worms and two gut contents (depurated sediment) after 24h of depuration. The number of particles as well as their size, shape and color were reported and compared between sampling period and locations. Results showed the presence of particles in both worms and gut content. More particles were observed in gut content with 60.7% in pools from site S1 and 88% for S2. In worms, 40% (S1) and 66% (S2) of the pooled samples contained particles. However, a low number of particles per samples was observed. For example, in worms collected in April and June 2017,  $0.75 \pm 0.7$  and  $0.21 \pm 0.31$  particles/worms were counted. The majority of particles observed were fibers and fragments but foam, films and granules were also identified. In addition, the most polymer type observed by Raman spectroscopy was polypropylene. This study revealed that microplastics are ingested by the ragworm H. diversicolor but more research is needed to measure their effect, especially after chronical exposure in estuarine organisms such as the ragworm H. diversicolor.

Keywords : Estuary , Hediste diversicolor , ingestion , microplastic , worm

# Analyzing bacterial community structures associated with different materials in freshwater

Al-Omari Jafar, Szabo Istvan, Szerdahelyi Soma Gábor, Szoboszlay Sandor.

Microplastics are abundant in freshwater environments, it provides a new surface for bacterial colonization. Up to now only a little is known about whether the bacterial communities associated with microplastics are plastic-specific or not, even less in freshwater environments. In our study a self-designed plastic colonizers, filled with five different materials, were used in to analyze bacterial community associated with wood, glass, plastic, biodegradable plastic, and stainless steel in freshwater. The different material containing plastic colonizers installed 50cm under water surface in a fishing lake located in Hungary. The colonizers were collected after two months, and additionally, the surrounding lake water were also sampled. Microbial communities associated with the five materials and the lake water were then analyzed by 16S rRNA amplicon sequencing method. Amplicon sequencing results showed notable differences between bacterial communities associated with filling materials compared to the lake water microbial community. Verrucomicrobiales, Nostocales, Rhodobacterales, Bacteroidales, Clostridiales were dominated the bacterial community associated with degradable plastic, stainless steel, glass, plastic, and wood respectively. Furthermore, different filling materials have shown differences in terms of richness, evenness, and diversity. Comparing water bacterial community to the different materials associated ones, water showed the lowest value of species richness, evenness, and diversity. The highest species richness and diversity value were measured in case of plastic and biodegradable plastic. Aknowledgements: This research was supported by the Thematic Excellence Programme (NKFIH-831-10/2019 - Szent István University, 2019) awarded by the Ministry for Innovation and Technology.

Keywords : 16S rRNA amplicon sequencing , microplastics associated bacterial communities , self designed plastic colonizer

#### **Electric clothes dryers: An underestimated source of microfiber pollution**

Miller Rachael, Kapp Kirsten.

It is well-established that microplastics in the form of textile fibers enter the environment via washing machines and wastewater treatment effluent. Less is known about the release of microfibers from electric clothes dryers. In this study we measure microfiber emissions from home installed dryers at two different sites. At each site the distribution of fibers landing on the snow's surface outside dryer vents and the weight of lint in dryer exhaust exiting dryer vents were measured. Fibers from the pink polyester fleece blankets used in this study were found in plots throughout a 30ft (9.14m) radius from the dryer vents, with an average number across all plots of  $404 \pm 192$  (SD) (Site 1) and  $1,169 \pm 606$  (SD) (Site 2). The majority of the fibers collected were located within 5 ft (1.52m) of the vents. Averages of  $35 \pm 16$ (SD)mg (Site 1) and  $70 \pm 77$  (SD)mg (Site 2) of lint from three consecutive dry cycles were collected from dryer vent exhaust. This study establishes that electric clothes dryers emit masses of microfiber directly into the environment. Microfiber emissions vary based on dryer type, age, vent installation and lint trap characteristics. Therefore, dryers should be included in discussions when considering strategies, policies and innovations to prevent and mitigate microfiber pollution.

Keywords : dryers , emissions , microfiber , textiles

#### Session 27.4\_O. Chaired by Mateo Cordier, Guyancourt

Plasto-tarball, a sinkhole for microplastic (Croatian coast case study)

Fajković Hana, Cuculić Vlado, Cukrov Neven, Kwokal Željko, Pikelj Kristina, Huljek Laura, Marinović Slavica. Paper number 333877

#### Flocculation of microplastic and cohesive sediment in natural seawater

ersen Thorbjorn Joest, Rominikan Stiffani, Olsen Ida Stuhr, Skinnebach Kristoffer Hofer, Grube Nynne Zaza, Jedal Soren Roger, Laursen Simon Nyboe, Fruergaard Mikkel. Paper number 334271

#### **TRANSPLAS: Battling the "Known Unknowns" in aquatic plastics research**

Cundy Andrew, Paterson Harriet, Stead Jessica, Crutchett Thomas, Hovey Renae, Ford Benjamin, Speldewinde Peter, Zapata-Restrepo Lina, Yanfang Lu, Zhang Xiaoyu. Paper number 334436

### Unfortunately they have arrived: stranded pellets and mesoplastics in a beach of Fildes Peninsula (King George Island, Antarctica)

Lozoya Juan Pablo, Rossi Florencia, Rodriguez Mauricio, Pérez Andrés, Lacerot Gissell, Lenzi Javier, Teixeira De Mello Franco. Paper number 334591

#### Plasto-tarball - a sinkhole for microplastic (Croatian coast case study)

Fajković Hana, Cuculić Vlado, Cukrov Neven, Kwokal Željko, Pikelj Kristina, Huljek Laura, Marinović Slavica.

Tarballs are black, often spherical objects; when crushed, develop a strong petroleum odour, and are usually related to the oil spills. The occurrence of tarballs in nature is frequent and they are recorded in several countries, such as Malaysia, India, Iran, Cameroon, USA, etc. Aggregation of plastic debris, pellets, and fragments with tar-balls were observed during the exploration in the 80s, and the expression "plasto-tarball" was proposed. Tarballs can be found along the Croatian eastern Adriatic coast, often on the islands. Samples of plasto-tarballs were sampled at the natural beach on Žirie Island, situated in the Croatian middle Adriatic coast, 22 km from the mainland. After analyses (FTIR and GC/MS) the origin of tarballs was determined as well as a type of microplastic particles caught in it. It was concluded that analyzed tarballs are connected with crude oil, probably related to oil spill incidents, and as such the result of the human activity. Particles identified by FTIR were divided into three groups: (a) rounded particles, 3-8 mm in diameter; (b) fibers, 10 - 27 mm length, and (c) and tabular particles, 1-12 mm in diameter. If microplastic (MP) is defined as particles smaller than 5 mm, than the majority of analyzed plastic particles fall in this category. Particles were defined as PE, PP, and PVC. Due to its high-density, tarballs are considered as a sinkhole for some amount of MP particles. Based on the described occurrence, the stability of tarballs should be considered, especially plasto-tarballs as they can serve as a secondary source of afterward pollution, and/or source of remobilized MP particles. Acknowledgement: This work has been supported in part by Croatian Science Foundation under the project IP-2019-04-5832.

Keywords : Croatia , crude oil , microplastic , Plasto , tarball

#### Flocculation of microplastic and cohesive sediment in natural seawater

ersen Thorbjorn Joest, Rominikan Stiffani, Olsen Ida Stuhr, Skinnebach Kristoffer Hofer, Grube Nynne Zaza, Jedal Soren Roger, Laursen Simon Nyboe, Fruergaard Mikkel.

The flocculation of combinations of microplastic particles (MP) and natural cohesive sediment has been investigated in a laboratory setup using unfiltered seawater. The experiments were setup such that the relative number of MP particles to natural particles were environmentally realistic and both PVC, PET, Nylon and HDPE were examined, ranging in densities from about 1.4 to 0.98 g cm-3. MP particles in the size-range 63-125 µm were incubated with suspensions of local untreated seawater and untreated fine-grained sediment (; 20um) collected from a tidal mudflat. Settling experiments were carried out with both a floc-camera video equipment (PCam) and conventional settling tubes. The experiments showed significant increases in settling velocities of the MP due to flocculation with the natural sediment. This was also the case for the buoyant HDPE-particles which was settling due to the flocculation. Analysis of grain size distributions of subsamples from the settling tube experiments showed generally uniform contents of MP in all subsamples, showing that I: the MP was incorporated into flocs and not settling as individual particles and II: the MP did not alter the settling velocity of the natural sediment as these low concentrations. The exact flocculation mechanisms still remains to be revealed but the general cohesiveness of fine-grained natural particles, organic particles as well as particulate and dissolved organic polymers are believed to be responsible for the flocculation. A strong effect of salinity was also observed, confirming the classical concept of increased flocculation of fine-grained particles as they are transported from fresh-water to estuarine and marine waters. The implication of the aggregation is that primary MP from land-based sources are likely to flocculate with other suspended particles, especially in high-turbidity estuarine environments and MP from terrestrial sources are consequently unlikely to escape further offshore.

Keywords : Deposition , Flocculation , Settling velocity

#### **TRANSPLAS: Battling the "Known Unknowns" in aquatic plastics research**

Cundy Andrew, Paterson Harriet, Stead Jessica, Crutchett Thomas, Hovey Renae, Ford Benjamin, Speldewinde Peter, Zapata-Restrepo Lina, Yanfang Lu, Zhang Xiaoyu.

Plastics pollution is a global environmental and human health issue, with plastics now ubiquitous in the environment and biota. Despite significant international research, key knowledge gaps ("known unknowns") remain around ecosystem-scale and human health impacts of plastics in the environment, particularly in limnetic, coastal and marine systems. Here, as part of the multi-national TRANSPLAS project, we: (a) review aquatic plastics research and data in three contrasting geographic and cultural settings, which present a gradient of heavily urbanised to more pristing environments: China, the United Kingdom, and Australia; and (b) present data from standardised fieldwork which compares, using standard methods, plastics pollution in various coastal settings in the southern UK, southwestern Australia and eastern China. To date, development pressures and necessary responses in each nation have defined the skills developed and their research foci, and there is a bias in national research priorities driven by levels of pollution, funding, government priorities and academic areas of interest. This has resulted in aquatic plastics datasets that are hard to compare directly, supporting the need to converge on standardised sampling methods, and bioindicator species. Standardisation is critical to make geographic comparisons more reliable, and here we present initial results from simultaneous field deployments and training programmes in Australia, the UK and China, using standard methods to provide comparable aquatic plastics data between each coastal setting.

Keywords : Australia , China , Marine Litter , Microplastics , United Kingdom

### Unfortunately they have arrived: stranded pellets and mesoplastics in a beach of Fildes Peninsula (King George Island, Antarctica)

Lozoya Juan Pablo, Rossi Florencia, Rodriguez Mauricio, Pérez Andrés, Lacerot Gissell, Lenzi Javier, Teixeira De Mello Franco.

Plastic debris is transported by marine currents over long distances, and several reports have evidenced the presence of plastics in remote areas. In the Polar Regions, microplastics (MPs.<5mm) may come from local sources or be transported long distances from lower latitudes. However, reports suggest that in the Southern Hemisphere the Antarctic Circum-polar Current (ACC) may act as a natural barrier for plastic debris, such as it does for species distributions. Within the frame of the AntarPLAST project, during the austral summer of 2019, beach sur-veys were carried out on the coast of the Drake Passage, at the Fildes Peninsula (-62.1582S,-58.9391W). During those surveys the beach was characterized (slope, width, orientation) and the first 2cm of sediment from 5 quadrants (50x50cm) along 100m of the highest strand line were analyzed. The polymeric composition of MPs was analyzed by FTIR and their diameter and area using ZooScan and the Image-J software. We found 293 items of micro (188 items) and mesoplastics (105 items), with a total mean density (±SD) of 234.4±166 items/m2. Foams (124.8±72.9 items/m2), Fragments (58.4±56.0 items/m2) and Pellets (44.0±50.5 items/m2) were the most abundant types. While secondary MPs may not be a big surprise, the presence and high densities of pellets (i.e. primary MPs) it is, being this the first record for beaches in Antarctica. These results not only alert about the presence and possible consequences of these MPs in Antarctica (e.g. direct consumption by organisms, adsorbed POPs, or their associated Plastisphere). The presence of pellets would reinforce the already mentioned occasional entries through the ACC of organisms and materials from lower latitudes. Despite increasing research, knowledge of plastics dynamics and their impact in the Southern Ocean and Antarctica, is still limited but certainly necessary.

Keywords : Antarctic Circumpolar Current , Antarctica , beach , Pellets , Zooscan

#### Session 27.4\_Me. Chaired by Gunnar Gerdts, Helgoland

**Interaction of nanoplastic debris with mineral substrates: application towards their removal from aqueous systems** *Gopala Krishna Darbha, Singh Nisha, Tiwari Ekta.* Paper number 333695

Microplastic fibers in the water column do not influence fertilization and hatching success of sticklebacks

Rebelein Anja, Scharsack Jörn. Paper number 334210

**Towards monitoring of small microplastics via the rapid identification and quantification of microplastics by hyperspectral quantum cascade laser infra red (QCL-IR) imaging** *Primpke Sebastian, Godejohann Matthias, Gerdts Gunnar.* Paper number 334299

### Interaction of nanoplastic debris with mineral substrates: application towards their removal from aqueous systems

Gopala Krishna Darbha, Singh Nisha, Tiwari Ekta.

The abundance of plastic waste across the globe and its further degradation resulted in the widespread of nanoplastic debris (NPD) contaminating the majority of the water bodies. As the mechanism of separation of nanoplastics is distinct from conventional pollutants, it is challenging for the water treatment plants to supply plastic-free water. Our recent attempts to understand the interaction of nanoplastics with mineral substrates unveiled an eco-friendly methodology to successfully remove nanoplastics from freshwaters. The results showed that independent of temperature and persistent geochemical conditions (pH, ionic strength, inorganic salts), the clay particles participate in the hetero-aggregation of nanoplastics in all-natural waters[1]. The layered doubles hydroxides (Zn-Al) possessing a similar structure that of clays were then applied for the removal of NPD. Maximum sorption capacity (Qmax) of 162.62 mg/g was achieved in synthetic freshwater. However, the removal was less with an increase in hardness of water (Qmax=53 mg/g). A 100% removal of NPD was observed under acidic conditions (pH 4) and the removal significantly decreased at alkaline pH conditions (pH<sub>2</sub>9). The current results show the Zn-Al hydroxides are promising adsorbents for the removal of NPD from freshwaters[2]. Reference: 1. Singh, N., et al., Understanding the stability of nanoplastics in aqueous environments: effect of ionic strength, temperature, dissolved organic matter, clay, and heavy metals. Environmental Science: Nano, 2019. 6(10): p. 2968-2976. 2. Tiwari, E., et al., Application of Zn/Al layered double hydroxides for the removal of nano-scale plastic debris from aqueous systems. Journal of Hazardous Materials, 2020. 397: p. 122769.

Keywords : adsorption , aggregation , fate , freshwater , Nanoplastics , removal

# Microplastic fibers in the water column do not influence fertilization and hatching success of sticklebacks

Rebelein Anja, Scharsack Jörn.

Increasing global plastic production along with plastic littering leads to accumulation of microplastic particles in aquatic environments. Microplastics are detected in aquatic habitats worldwide, whereby microplastic fibers belong to the predominant plastic types. As early life stages of aquatic organisms are predicted to be especially vulnerable to microplastic pollution, we tested the impact of polyester fiber presence in the water during in-vitro fertilization and embryo development of three-spined sticklebacks (Gasterosteus aculeatus). Egg clutches (n=6) were split and one half fertilized in water containing pristine polyester fibers while the other eggs served as control treatment without fibers. Fiber concentration was set at 10 000 fibers per liter and maintained with water exchange throughout the entire experimental period. Fibers were kept in suspension by continuous agitation of the breeding bowls. Observation with a dissection microscope revealed that some polyester fibers stuck to the outside of the eggs in the fiber treatments. Yet, no significant difference in fertilization, hatching or mortality rates were observed between the divided egg clutches. Furthermore, heart rate of embryos and morphological features of hatched larvae were not affected by fiber presence in the water. The results suggest that fertilization and early development of stickleback eggs are unaffected by pristine polyester fibers in the water column, even at fiber concentration above current environmental levels.

Keywords : embryonal development , Exposure study , fish , polyester fiber

#### Towards monitoring of small microplastics via the rapid identification and quantification of microplastics by hyperspectral quantum cascade laser infra red (QCL-IR) imaging

Primpke Sebastian, Godejohann Matthias, Gerdts Gunnar.

The investigation and monitoring of microplastics (MP) is of emerging concern for legislation and societies. Especially the analysis of small MP (below 100 µm) creates additional challenges and pitfalls. To determine particle size and chemical composition in one measurement FTIR imaging provides a relatively fast method for complete filter areas. Here, modern systems measure field sizes of 12 x 12 mm<sup>2</sup> (pixel resolution =  $(5.5 - 11 \mu m)$  within 4 hours but need a permanent liquid nitrogen supply 24 hours. Here, we present a novel approach to measure such filter areas within less than 1 hour. To achieve this measurement speed a quantum cascade laser infra red (QCL-IR) based microscope, Daylight Solution Spero QT, was used. Compared to existing FTIR imaging systems it has several advantages. The use of liquid nitrogen is obsolete while 230.400 spectra are measured simultaneously on an area of  $2 \times 2 \text{ mm}^2$  using a focal plane array detector with a pixel resolution of 4.2 µm within 1 minute including data storage. A smaller spectral range of 1800 – 950 cm-1 is available compared to FTIR imaging while a larger number of spectra is collected. To investigate the performance of the instrument and to foster harmonization with existing studies, it was validated against known polymer samples, the database was optimized for automated analysis similar to FTIR imaging and finally the results compared to those achieved by FTIR imaging. The QCL-IR-based microscope achieved more detailed results with a higher sensitivity for smaller particles compared to FTIR imaging in much shorter measurement times. Here, the measurement of a 12 x 12 mm area took only 36 minutes. By combining automated analysis with hyperspectral QCL-IR based imaging, we realized a novel fast and versatile tool to identify MP allowing the measurement of large sample sets within current laboratory routine times.

Keywords : FT, FTIR, imaging, infra red, IR, microscopy, Monitoring, QCL

#### Session 27.4\_Ma. Chaired by Christophe Maes, Plouzané

#### Microplastics in urban runoff on a suburban catchment of Paris megacity

Treilles Robin, Gasperi Johnny, Gallard Anaïs, Saad Mohamed, Dris Rachid, Jérôme Breton, Rabier Alain, Tassin Bruno. Paper number 334173

### Global impact of the synergistic toxicity of microplastics and organic UV filters on phytoplankton

*Revels Brandi, Cooper Adam, Duncan Caroline, Kronquist Ray, Kurkjian Robert, Dryden Howard.* Paper number 334543

### Can the toxicity of polyethylene microplastics be enhanced in the presence of engineered nanoclays? Preliminary results of flatfish *Solea senegalensis* larvae as a case-study

M. B. M. Santana Ligia, C. M. Rodrigues Andreia, Campos Diana, Figueiredo Joana, Silva Sara, M. S. Abessa Denis, Pousão-Ferreira Pedro, Candeias-Mendes Ana, Castanho Sara, M. V. M. Soares Amadeu, M. Rocha Rui, L. Silva Ana, Martins Roberto. Paper number 334548

### Assessment of juvenile mussel growth and health following chronic exposure to anthropogenic fibres

*Walkinshaw Chris, Cole Matthew, Tolhurst Trevor, Lindeque Penelope, Thompson Richard.* Paper number 334638

#### Microplastics in urban runoff on a suburban catchment of Paris megacity

Treilles Robin, Gasperi Johnny, Gallard Anaïs, Saad Mohamed, Dris Rachid, Jérôme Breton, Rabier Alain, Tassin Bruno.

Microplastics (MPs) in stormwater are poorly investigated. In order to fill this knowledge gap, MP concentrations were investigated in stormwater at the outlet of a runoff water pipe located in Sucyen-Brie (Paris suburb, France). Stormwater from four rain events were collected (from June 2018 to May 2019). Stormwater flowrates were measured and compared. Three rain events had approximately the same intensity, and one event was more intense. During each rain event, 300 to 500 liters of water were filtered using a plankton net with 80 um mesh size. MPs are then extracted using SDS and H2O2 30 % digestion followed by NaI density separation (d=1.6 g.cm-3). Blanks were performed to quantify contamination. Samples were filtered on metallic filters with a porosity of 14 µm. Samples were then remobilized on alumina filters and were analyzed using infrared spectroscopy. Infrared mapping analysis are performed on each alumina filters. Maps were analyzed using a microplastic analysis software siMPle, developed by Aalborg University, Denmark and Alfred Wegener Institute, Germany. Preliminary results show a concentration of 3.3–7.5 MPs/L (min-max values, contamination has been considered) during rain events. PP, PE and alkyd are the most prevalent polymers found in those samples. No correlation was found between rain intensity and concentrations. Considering the runoff volumes and the impervious surface area of the studied catchment, this would imply a flux of 4\*10<sup>7</sup>–9\*10<sup>7</sup> MPs.yr-1.ha-1.

Keywords : infrared spectroscopy , Microplastic , stormwater , urban sources

# Global impact of the synergistic toxicity of microplastics and organic UV filters on phytoplankton

Revels Brandi, Cooper Adam, Duncan Caroline, Kronquist Ray, Kurkjian Robert, Dryden Howard.

Global phytoplankton populations have decreased by more than 50% since 1950 and continue to decline by 1% per year. This loss of these primary producers directly impacts the entire oceanic ecosystem and reduces the resiliency of our climate system through the removal of a critical carbon sink. Recent advances in ecotoxicology have revealed the synergistic toxicity of two emerging groups of pollutants: weathered microplastics and hydrophobic organic pollutants. Specifically, microplastics have emerged as an important vector for concentrating and transporting organic pollutants such as organic UV filters used in sunscreens, other personal care products and plastics. The COVID-19 pandemic serves as a natural experiment for the sudden removal of these pollutants in tourist communities which have seen a substantial reduction in human activity. NASA's MODIS chlorophyl a dataset reveals rebounding phytoplankton populations and marine communities during the COVID-19 pandemic when the sources of these pollutants decline. Lessons learned from these observations should lead to more detailed studies and ultimately to reductions of these pollutants in the ocean through regulation and public awareness.

Keywords : ecotoxicity , MODIS chlorophyl , oxybenzone , phytoplankton , synergistic , UV filters

# Can the toxicity of polyethylene microplastics be enhanced in the presence of engineered nanoclays? Preliminary results of flatfish *Solea senegalensis* larvae as a case-study

M. B. M. Santana Ligia, C. M. Rodrigues Andreia, Campos Diana, Figueiredo Joana, Silva Sara, M. S. Abessa Denis, Pousão-Ferreira Pedro, Candeias-Mendes Ana, Castanho Sara, M. V. M. Soares Amadeu, M. Rocha Rui, L. Silva Ana, Martins Roberto.

Microplastics are posing a threat to marine ecosystems. Recent studies demonstrated that microplastics co-exposure can alter the uptake/toxicity of hazardous chemicals. In this context, this study aimed at assessing the toxicity of polyethylene (PE; 63-125 mm, irregularly-shaped), an abundant microplastic in coastal waters, and Cu-Al layered double hydroxides (LDH), a class of twodimensional engineered nanomaterials (NM), dispersed alone or combined, in flatfish Solea senegalensis larvae. Eight-day post-hatching larvae (n=20) were exposed for 3 h to three concentrations of Cu-Al LDH (0.33, 1.0, and 3.3 mg.L-1) and PE (0.1, 1.0, and 10.0 mg.L-1) in a full factorial design (i.e., all possible combinations plus a control with only seawater), to assess histopathological and biochemical changes. The nanoclay caused no significant effects on measured endpoints. Conversely, PE ( $\geq 1$  mg.L-1) triggered the activation of antioxidant defenses (CAT), neurotransmission (AChE), and aerobic energy production (ETS), without causing, however, an activation of detoxification enzymes (GSTs) or oxidative damage (LPO). The effects of PE mostly perdured in the presence of Cu-Al LDH nanoclays. The exceptions were observed in the combination of 0.33 mg LDH.L-1 X 1 mg PE L-1, which did not activate CAT, and 1 mg LDH.L-1 X 1 mg PE.L-1, which significantly increased ETS. A significant increase of the histopathological index was observed in the mixture of 1 mg LDH.L-1 X 0.1 mg PE.L-1. Nevertheless, treatments containing PE caused some degree of histopathological alterations, namely hepatic and renal hyperemia, liver hypertrophy, hepatocyte vacuolation, and gastrointestinal space dilation. PE microplastics, combined or not with Cu-Al nanoclays, seem not to compromise the homeostasis of the flatfish larvae when considering the environmental concentrations reported so far (; 1 mg.L-1). Yet, adverse effects can be expected if the concentration of such particles continues to rise, as projections suggest (an increment of two to four-fold by 2030).

Keywords : biomarkers , coexposure , fish embryotoxicity , histopathology , layered double hydroxides , nanomaterials

### Assessment of juvenile mussel growth and health following chronic exposure to anthropogenic fibres

Walkinshaw Chris, Cole Matthew, Tolhurst Trevor, Lindeque Penelope, Thompson Richard.

Mussels are considered a valuable model species for investigating microplastic toxicity, however the majority of existing studies have focussed upon the adult life stage. Here, a chronic exposure experiment was undertaken to investigate the effect of anthropogenic microfibres (polvester or cotton) on juvenile mussels (Mytilus spp.). Juveniles were chosen for this study as effects of microfibers on juveniles are currently under-represented, and early life stages are more sensitive to external stressors. Fibres were manufactured by cryo-grinding to represent a range of sizes commonly found in the marine environment (average polyester length =  $293\mu m$ , average cotton fibre length = 171µm), and represent some of the most prevalent fibres found in oceans worldwide. Mussels (5 individuals per beaker) were maintained in 1 L glass beakers, with a continuous flowthrough of algae-enriched filtered natural seawater under controlled laboratory conditions. Mussel cohorts were exposed to one of four experimental conditions over a three-month exposure period: (1) control (algae only; n=5); (2) cotton microfibres (100 per L; n=5); (3) low microplastic concentration (10 per L; n=5); (4) high microplastic concentration (100 per L; n=5). Condition (3) simulates current marine microplastic levels, with condition (4) simulating theoretic future concentrations. Algal clearance rate, shell length and oxygen consumption rate were monitored to assess how prolonged exposure to anthropogenic fibres might affect mussel feeding, growth and metabolic rate. Scope for growth was calculated at the end of the exposure period. Further, mussels were KOH digested to quantify anthropogenic fibre ingestion. Understanding the effect of microfiber ingestion on feeding, growth and respiration in juvenile organisms is critical to understanding the effect of microplastic pollution to the value of commercially exploited marine species. Our results provide insight into how environmentally relevant concentrations of plastic and cotton microfibers have the potential to affect feeding and growth of commercially valuable bivalve shellfish.

Keywords : Anthopogenic fibres , Chronic study , Cotton microfibre , Microplastic , microplastic ingestion , Mussel , Polyester microfibre , sublethal effects

#### Online open-science edition of the MICRO 2020 international conference By the Zero Plastic working group \*

In this paper we share the main elements allowing the 3rd edition of MICRO -our biennial international conference on plastic pollution from MACRO to nano- to offer online open-access content while fostering a collaborative effort from the continuously expanding MICRO community.

This edition of the conference took place in COVID-19 lockdown conditions. Together we were more than 2000 authors with around 500 abstracts for oral presentations and posters. From the authors, speakers, chairpersons, scientific committee, core group, organizing team, and All participants, we had fruitful engagement throughout the sessions from each local node: Banyuls, Bastia, Bayreuth, Bilbao, Bouguenais, Bremerhaven, Brunswick, Crozon, Gran Canaria, Guyancourt, Helgoland, Ithaca, Lanzarote, Le Mans, Leipzig, Madrid, Mallorca, Marne la Vallée, Mazatlán, Menorca, Nantes, Ostend, Plouzané, Plymouth, Rostock, Seoul, Siena, Toronto, Trondheim, Utrecht, Vienna, Vigo and Wageningen.

This open-science process unfolded through 7 steps:

. The starting point was the CNRS platform "sciencesconf.org", which allowed abstract submissions, participant registration, and the export of the abstracts and participant registration information in ".csv"format. This was kept in the team archives in ".pdf" format, maintaining the integrity of the format and content of the authors' original submissions.

. The abstracts were shared with the scientific committee for brief review. This allowed us to achieve a peerreviewed standard and make a first step in establishing the conference session structure.

Interdisciplinarity was the key criteria for determining the set of presentations in each session.

. The ".csv" file from "sciencesconf.org" was converted to a ".bib" file using a script running SPIP\* CSM in the dedicated platform "**micro.infini.fr**". This allowed us to establish the bibliographic database "MICRO2020.bib" and import it to Zotero\*. Here you have the csv2bib script.

#### https://framagit.org/b\_b/micro/-/blob/master/formulaires/csv2bib.php#L17

. The "MICRO2020.bib" from Zotero\* was connected to the "micro.infini.fr" platform using the plugin ZotSPIP https://plugins.spip.net/zotspip.html to manage the communication informations for each abstract of the conference's programme, along with the map using mulitple OSM based APIs and Wikidata for geo-location ( photon.komoot.io, nominatim.openstreetmap.org & wikidata.org). This was adjusted manually for disambiguation. This connection was made by a bridge described in the code

#### https://framagit.org/b\_b/micro/-/blob/master/pz\_fonctions.php#L24 &

https://framagit.org/b\_b/micro/-/blob/master/pz\_fonctions.php#L106, inspired from what is done in PUMA from https://github.com/OllyButters/puma.

. Prior to the conference, each speaker sent their presentation. These included posters in ".pdf" format or a 7-minute prerecorded video for oral presentations, submitted via an infomaniak server where the data will remain archived until the 2022 edition of the conference. The prerecorded video idea was inspired by the State of the Map 2020 conference.

. Each day we shared the visioconferencing room details with the respective chairpersons and speakers; for visioconferencing our first choice was BigBlueBotton, installed in University partners' servers, but running short of time we used a proprietary visioconferencing license already available to us. We connected this via streaming to the public broadcasting interface using a video streaming provider at the Lanzarote local node.

. The Conference Proceedings have an associated ISBN as multimedia content with the text from the abstracts as the main content, including videos and posters as part of the Conference Proceedings under cc-by-nc-sa license.

By working through a collaborative open-sciences approach we managed to facilitate a conference with the highest scientific standards and no registration fee. No fee does not mean the conference was free; it means that the creation and production costs were covered by public institutions engaged in the MICRO conference series.

\* The Zero Plastic working group is an alliance between the World Network of Island and Coastal Biosphere Reserves and the Marine Sciences For Society researcher's network. For this MICRO 2020 by alphabetical order we were in the "Tech" working group: Ana Carrasco, Aquilino Miguelez, Armando Acuña, Bethany Jorgensen, Bruno Bergot, Eva Cardona, José de León, Juan Baztan, Juanje Luzardo and Raul González.

#### **Pads Section**

This section includes the Pads that were used during the MICRO 2020 conference.

They are unedited and reflect the live communications taking place during the week of the conference; the guidelines below were addressed to All participants for each pad:

*IMPORTANT: Please include your name at the beginning of your question or scientifically-relevant comments.* 

You can follow the live-stream discussion between the session speakers and the chairperson through the dedicated channel on the conference home page during the specified hours in the programme.

The chairperson will draw upon your questions to the speakers during the LIVE discussion.

Day 1/5 Pads

#### **Questions and comments for the session 23.1\_O**

Contact details for follow-up questions to the speakers: Fanny: fanny.caputo@sintef.no Symiah: barnetts@aston.ac.uk Lucian/Konstantinos: lio@civil.aau.dk, kopa@civil.aau.dk Mohammed: Mohammed.al-azzawi@tum.de

For Mohammed: Name: Mikaël Kedzierki Question: Have you tested the impact of degradation methods on aged polymers? Okay thanks

Adam Porter Hydrogen peroxide and sulphuric acid used in your fentons reaction may damage other polymers...did you see any evidence of this? Polyamide in particular (nylon) may be susceptible

Andrea: what was the size range of the first experiment where the change in size was significant?

Claudia. Fenton's reaction is very sensitive to the pH. Did you measure the pH before/during the reaction? You also mentioned the increase in temperature during the reaction. What was the highest temperature reached?

Name/Adrian López-Rosales

The Fenton's reaction could precipitate Fe (II) salts? How could remove?

Name: Sebastian Primpke Question : Did you also try to run Fenton with a temperature control to avoid the heating? Thanks!

For Konstantinos/Lucian Name: Andrea Faltynkova Q: What sort of validation do you use for the classification of MP used by the simple model? And what type of chemometric analysis is used in the simple model? Thanks !!

Andrea: I had a question for Konstantinos/Lucian: Is it possible to extract MPs from the biofilters themselves?

ForSymia

Name: Eike Esders Question: Where can we find an email address for follow up questions? Thanks:)

Name: Susanne Belz Q: What is the smallest particle size that can be investigated with your method?

Name: Cleo Stratmann

Question : What kind of Ethanol did you use? What about the impact of Ethanol on the plastics, there are some papers stat indicate Ethanol can be detrimental to plastics, too. Did you test on that in some way? thank you; Great work

Name: Oliver Knoop Q: Just sugar, water, and ethanol: Do environmental samples do not require a sample treatment? nice answer, thanks!

Name: Magdalena Mrokowska Q: Have you noticed any air bubbles attached to plastic fragments that may change buoyancy? If so, how did you approach this problem?

Name: Robby Rynek Q: How would you account for the change of density of the polymers by biofouling? Thanks for the answer.

Name Barbara Scholz-Böttcher Q: What do you think is the applicable size range in particular the minimum size? -relevance of gravity

Name / Mikaël Kedzierski I think that plastic density changes during the ageing and fouling processes. How does your methodology address these issues?

Name: Matthieu Mercier Question: How-long is the sorting process? What would it take for large numbers of samples?

Name: Matthieu Mercier Question: How-long is the sorting process? What would it take for large numbers of samples? (THANKS BUT SORRY, WRONG SPEAKER) - moved, Andy

For Fanny Name: Josef Brandt (to Caputo Fanny) Question 2: AF4 can yield mass fractions of the analytes after separation by integrating dRI detector signals. Did you try that?

Name: Barbara Scholz-Böttcher Question : How you appraise the applicability to environmental samples?-regarding aggregates...

Name: Alexandre Dehaut Question 3: What about the possibility to prepare sample for identification downstream with such approaches?

Name: Ghezali Yousra Question: how an we use oil density separation for microplastic in sediment?

#### **Questions and comments for the session 23.1\_Me**

Name: Christina Bogner Question 1: Could you give more details on the amount of MP you exposed, please?

Name:...Stephan Rohrbach Question 2:...Have you analyzed some of the rainwater properties or do you know if the rain in your area is different from other parts of the world?

Matthias Egger: Great talk! Did you also check for weight loss?

Name: Zhiyue Niu Question 1: Have you observed fragmentation of particles ?

Name: Barbora Pinlova Question: Can the amount of ozone exposure in your experiments be compared to real life situation?

Name: Walter Waldman (Brazil - Federal University of Sao Carlos) Question: Great talk and relevant theme! Not considering PE, PP, or PS, which polymers do you think are the most common in urban environments?

Name: Eike Esders Question: Do you have any clews, why the addition of rain water deepened the cracks?

Mariana Miranda: Thank you all for the great questions and suggestions. Please contact me through this email to talk more about this topic: mmiranda@fe.up.pt

Name: Alexandre Dehaut Question: Did you have the opportunity to checks which chemicals were in your extracts? (Nice presentation :-)) Barbro: Did you also check extracts of the plastics in culture medium, this may yield very different extraction than DMSO

Name: Matthias Völkl

Question: Thanks for the talk! Which concentrations did you use for the genotoxicity assay?

Christoph: very nice presentation!!! How do the tested concentrations relate to the detected particle concentrations in the seafood?

Hi Christoph, thank you. So far, it is still hard to do this relation. Soon (I hope) we will be able to know which are the predominant polymers in the organisation. But to related it to chemical concentrations, well, there is still work to do...

Winnie Courtene-Jones: Thanks for your talk, Did you analyse what chemicals were in each of the extractions from the different polymers? - you just mentioned this analysis has not yet been done. What methods will you use to identify the organic compounds and the metals? this still under dicussion actually because w would like to analyse chemicals in DMSO and this can be tricky. I would be glad to discuss it longer with you if you have some opinion about it.

#### Joao Frias

Great Presentation Florane. I was wondering whether you took into consideration the potential effects of using DMSO? Would that have an impact?

#### Alexandre Dehaut

How do you deal with the presence of bacteria from the environment too? Hi, unfortunately no. We only focused on the chemical effects extracted with DMSO in this study

#### **Boris Eyheraguibel**

Thx for the nice talk ? Did you test control with natural sample (leaves - branches) to check toxicity of other carrier ? Hi Boris, That could be a nice idea. We just perform a regular control with DMSO only but we try that next time.

#### Name: Federica Guerrini

Thank you for the presentation! Do you think that microplastics sampled from seawater could have a different effect on human cells? I read something on the very last slide, can you please tell more about it? Thanks!

#### Molly McPartland

You are assuming that the oxidative stress is linked to DNA damage? Were you able perform statistical analysis to show this could be the case? Hi Molly, yes oxidative stress could explain DNA damage observed for some of the polymers unfortunately we were not able to demonstated it statistically. But we perform the study on 2 different cell lines to strength the data Also, what are the differences in your cell lines that resulted in your different results in polymer type and outcome? At this step of the study it is still hard to answer the question. however the cell lines are different in term of bioactivation abilities for example. Hepg2 cell line have high biotransformation ability regarding PAH for example that could increase their sensiblity to such compound. Great! Thanks very much for the response!

#### Lisa Zimmermann

What chemical analysis are you planning? Hi Lisa, we are planning to perform POPs (PAH and PCB), some flame retardant and phtalates certainly as well as metals analysis

#### Zhyue Niu

Nice work!! Do you know the exact polymer type of the bio-plastic you tested?

Winnie Courtene-Jones: Thanks for your talk, i might have missed this (sorry), were all the samples 'virgin'' polymers, from the manufacturers? Were you able to get a list of the additives which were in these from the manufacturer? if so were the compositions of some of these in line with what you were expecting to find in them?

Thanks! sorry a bit of a long question :)

Ana Catarino - Hi, many thanks, very interesting. Did you pre wash the pellets to make sure there were no contaminants sorbed to the plastics?

Alicia Mateos Cárdenas. Nice presentation, thanks! How does the UV radiation selected for your experiments translate to environmental UV radiation?

Coco Cheung: Thank you for the talk! Sorry for missing it but why did you not choose HDPE as one of your sample polymer? Thanks! We randomly selected the plastics, but I expected more activities from LDPE.

Simon Wieland: Thanks a lot for your talk! Are the concentrations of chemicals in your leachcates close to environmentally relevant concentrations?

Laura Markley: Thank you for the talk! Sorry if I missed this part. How did you go about comparing your UV conditions with outdoor conditions? Were the pellets exposed in a lab using a lamp or outdoors? The plastics were weathered under laboratory conditions. Gewert et al. published one paper, where she calculated the European sun exposure and I used the formula to do the same ;-)

Barbro (b.n.melgert@rug.nl): Any idea how many fibers collect indoors by a family of 4 per year just wearing polyester clothes, assuming limited ventilation?

Florane Le Bihanic : Thank you for the nice presentation! Did you observed any change in FTIR fiber spectrum between befor and after washing the fiber

Barbora Pinlova: Thank you for the presentation. How did you check for the contamination during the movement experiments (eg. from other garments worn during the experiment)/How did your blanks look?

\_Éva Vizsolyi: Thank you for your very nice presentation! I would like to ask, how did you clean the samples for the FTIR analyses?

Yasmin Yonan: Thanks for the presentation! My question was very similar to Barbora's, I thought your answer was good. Thank you!

#### Questions and comments for the session 23.1\_Ma

Question FROM:Claudia Drago

Question FOR Michaela Miller. Thank you very much for your presentation! I wanted to ask you What was the size range of the fragments? Maybe I miss it... Thank you. Response: Thank you! The lengths (longest calliper length) for the fragments ranged from 500 μm to 2 mm, the widths (shortest calliper length) ranged from 250 μm to 1.5 mm, and the surface areas ranged from 30 μm<sup>2</sup> to 1.6mm<sup>2</sup>.

Question FROM: Ika Paul-Pont

Question FOR Michaela Miller.

QUESTION Some waste water treatment plants retain high proportion of the fibers, sometimes up to 80-85% do you know whether it is the case in your area?

Response: Thank you for your question! We've not analysed WWTP samples specifically from our area yet. However, we have preliminary data from our lab surrounding animals taken in coastal areas adjacent to the WWTP and they have found higher levels of fibres than fragments. Additionally we

have water column samples in the area showing actually more fibres than fragments. We can assume this trend is the same in WWTP here based off of that, but unfortunately we don't have actual data just yet!

Question FROM Charlene Trestrail

Question FOR: Cecilia Siri

QUESTION: Do you think exposure to these hormones via the stomach is worse for fish than exposure via the gills?

I would like complete my answer for your question here. We did not investigate the exposure via gills themselves. But if we compare the exposure of progesterone via MP ingestion compared to water ingestion, the exposure via MP is suggested to be an additional route of exposure. The model that we used is quite simplified, and this question needs to be better investigated.

Question FROM: Thomas Witzmann

Question FOR: Cecilia

QUESTION:What was the size range of the particles? Are they monodisperse and also have you investigated the surface charge of them as it will propably influence the adsorption of different steroids/micro-pollutants

The size range of particles were between 350-1000µm, depending of the MP type. We investigated the effect of pH solution on the adsorption capacity of progesterone. Since progesterone does not present any ionizable group, we didn't see any influence of the pH solution on its adsorption capacity. Therefore, surface charge interaction is not expected to play a role in the adsorption process. Nevertheless, surface charge of MPs can be measured to better characterize MPs.

#### Question FROM:Claudia Drago

Question FOR:Cecilia Siri

QUESTION:How much the size and the shape of plastic can influence the absorption and desorption? Do you think there is some size range that can be more harmful?

Thank you for your question. We did not investigate the size and shape of microplastics. We were more focused on the influence of plastic type (PE, PP, PS) than on the shape and size. But this is for sure an interesting aspect to investigate for adsorption and even more interestingly for desorption. But all plastics used have a spherical shape. The size is expecting to influence the specific surface area, and therefore potentially the adsorption capacity.

#### Question: From: Matt Cole

Question for: Beatriz

Q: Thank you for your talk. Oxidative stress is a natural process for animals responding to inflammation. Did you look at oxidative stress response over time to see if it was part of a healthy response?? (Sorry if I missed the timepoints).

#### Question: From: Kevin Tallec

Question for: Beatriz

Q: Thank you for the presentation. Some compounds can adsorb on plastic and improve ingestion of particles by organisms. Did you check the number of MP or marinated MP ingested to determine if the fluctuating toxicity is related to different ingestion between virgin and marinated MP ?

Hi Kevin. Both MPs enriched diets were composed by 10% of MPs, that means 90% of nutritional components and 10% of MPs. In addition we did not see significant differences in the amount of pellets ingested

Question from Claudia Drago Question for Beatriz Q:What was the ratio between food and plastics? Which concentration of plastic and food did you use? Thank you!! :) Hi Claudia, thank you for your question. The ratio was 90/10: 90% of nutritional components and 10% of plastics.

Question from Julia Pawlak

Question to Beatriz

Q: Do you analyse the marined particles for any absorbed chemicals/ compunds or micro-organisms? And btw, thank you for your talk!

Thank you for your question Julia. Yes, we did and results shown that after two months at sea, pellets adsorbed a total of 50 different chemicals. We find different types of antioxidants, polycyclic aromatic compounds (PAHs), fragrances, ... thus demonstrating the ability to absorb chemicals of different natures

#### **Questions and comments for the session 23.2\_O**

Name:..Contact Data of Speakers (provided by Melanie Pöhlmann / SFB1357 Coordinator); Question 1:... Ben Gilfedder: Benjamin-Silas.Gilfedder@uni-bayreuth.de;

Markus Rolf: markus.rolf@uni-koeln.de

Christian Schmidt: christian.schmidt@ufz.de

If you have questions after the session, you are warmly invited to contact the speakers directly!

Name: Eike Esders Question 2: Thanks for the talk. Do you have an idea about the precision of settlings velocities measured? Thanks for the answer

Name: Merel Kooi

Question: Thanks! I might have missed it- how did you include outflow from your lake? Since the particles sink to the loewst layer, but where does that mixed water go to? - Thanks!

Though that makes me wonder about the residence times of this lower layer? How can you calculate that when they don't go out? ;)

Name:Matthias Völkl Question Any idea why 51-150µm have the highest amount and not smaller ones? Do you assume an amount of smaller particles than 10µm?

Name: Fan

Question: Thanks for the talk, just wonder, have you performed recovery test in sample prep? is there any reason why pre-oxidation was not performed? in your sample prep. Thanks

Name: Melissa Maurer-Jones Question: Thanks for the talk! Did you test the concentration of particles in the soil outside the floodplain region? Name: Tomasz Burghardt Have you identified any particles of road markings (these most likely would be polyacrylates)? Any glass microspheres? Thanks.

Question to Christian: Matthias Egger: Great talk! Do you also have some numbers on mass of MP in your model?

Great Multitasking skills Sven!

#### Questions and comments for the session 23.2\_Me

Questions for Matthias (matthias.tamminga@uni-hamburg.de)

Name:...Winnie Courtene-Jones (winnie.courtene-jones@plymouth.ac.uk)

Question 1:... very interesting, is the water body stratified? did you measure the salinity of the water at different layers.

is Mattias online? can you briefly explain you main results?

Thank you for your talk, it is a fascinating topic! very interesting, Thank you for the answer.

Adam Porter Very quickly how did you stop your mass filters clogging? M: we used a filter cascade (1 mm, 0.63 mm, 0.3 mm and 0.063 mm)

Question 2: For Mattias

Fibers are usually hard to stained and identified. How did he manage to identify and quantify them? Did he face hard times with this? M: they are more difficult to stain, but it certainly possible. About half of all particles we found were fibers. Maybe just wright me an email, so I can give you further information. Thank you!

Questions for Olubukola (olubukola.alimi@mail.mcgill.ca)

Great presentation. How does this interact with particle size? Is the effect related to the size of the particle (Peter Vermeiren, p.vermeiren@science.ru.nl). Thanks.

What natural organic matter was used in your experiment? Thank you. In this work, we used the Suwannee river natural organic matter.

Does the freezing appear in the groundwater or in the soil above? How often do groundwaters freeze - can your results be also considered for surfacewater freezing processes in the water surface or the litthoral zones??

Walter Waldman (walter.waldman@gmail.com)

You used PS round nanoparticles functionalized with a charged chemical group. What do you expect from natural degraded nanoplastics, edgy and with other kind of chemical groups originated from photodegradation, in your (very interesting) freeze-thaw experiment?

Questions for Shaun

Trang

Do you have any suggestion to reduce the mesh size of the net, so smaller sized MPs can be monitored as well?

Richard Cross UKCEH: thank you for the talk, do you have plans for how to more systematically compare overflow events with the general background releases? how many sampling occasions do you think you would need to generate an understanding of the "background" levels prior to storm overflow events?

Thanks

Questions for Kryss (waldschlaeger@iww.rwth-aachen.de):

AVarrani: Thanks for the presentation. In your experiments, did you considered different packing of the glass spheres? (avarrani@igf.edu.pl)

-> K: It was not possible to look at different packings of glass spheres, because then the repeatability of the experiments would have suffered. But the packing directly influences the porosity and by comparing uniform and bimodal packings (which have the same average diameter but different porosities), it was shown that porosity has a strong influence on infiltration. The higher the porosity, the deeper the infiltration.

Cleo: Very nice experiment and interesting results! Great matrix.

Do you hav any idea about how upflow currents in sediments could bring MPs up again. -> K: Thank you! I would love to look at that, I can just guess right now: I think upflow currents will reduce the infiltration depth.

Sascha Müller: Very nice presentation! Did you observe water chemistry during your experiments. You believe that Sediment surface chemistry/ sediment mineralogy may interfere with your schamtic from the end? Thanks :)

-> K: Thank you for that question. I am sure that sediment mineralogy and surface properties will influence the infiltration depth. But as a first experiment, glass spheres had many advantages, especially the clearly defined diameter. I think infiltration in natural sediments will be less deep, so we are safe when using my matrix ;)

Great presentation and paper. Was there a clear relation with the polymer type (i.e. polarity of the polymer)? Also, would the results of your vertical infiltration be transferable to horizontal transport as wel. (Cynthia Munoz, c.munoz@science.ru.nl)

-> K: Thank you for that question. I did not specifically look at the influence of the type of polymer , but I looked at the influence on density (which is influenced by the polymer type) and there were no

statistical significances. So I guess, the influence of polymer type is to low to measure (at least in my experiments). I guess horizontal transport will be lower than infiltration, especially because of the lesser water flow. But we should definitely study that! Thanks.

Adam Porter: Really nice...do you think the same applies for other matrices like beach sands etc? -> K: I would love to have a look at salt water and the influence it has on the infiltration depth. I guess it is low, but who knows. Salinity is probably the biggest difference between beach sands and fluvial sediments. It would be also interesting to look at degraded environmental particles, I guess that will have a big influence as well. But other than that, I guess it should be appliable to beach sand.

Cyril Hachemi (chachemi@deakin.edu.au): Thank You Kryss, Nice talk!

How would you compare infiltration of MPs in a column filled with glass beads with real sediments? Would you predict any impact of adsorption based on the chemistry of sediments? - Same question as Sascha finally

-> Yes, please have a look at the answer I gave Sascha. But just to add something: It is quite common to use glass beads as a substitute for natural sediment in hydraulic engineering. We used it for example for looking at the infiltration of fine sediments and to get a first idea of the possible depths it proved to be quite good.

Aline: Really nice presentation, thank you. Would you expected that the water flow in sediments might interfere the accumulation and permanence of MP?

-> K: Yes I think so. The currents in the water column above as well as horizontal water flow will also influence the infiltration depths of MP particles into fluvial sediments. But I guess it will decrease the infiltration depth, so when using my calculated infiltration depth (or the matrix) as a reference for the maximum infiltration depth, you are on the safe side and should find all particles in the sampled soil or sediment.

In nature, Biofouling and biofilm formation highly affect the sedimentation of Microplastics, so are you welling to study this part? and how do you think it will affect your results? -> K: I would love to study the effect of biofouling on all transport processes. I guess it would decrease the infiltration depths, as the surface properties are more sticky. but that is just a guess. We should find out!

#### Questions and comments for the session 23.2\_Ma

email adress Elena Hengstmann: elena.hengstmann@uni-hamburg.de (for further questions, I'm happy to answer these)

Name: Ghezali Yousra

Question 1: For the plastic litter quantification did you also count the effect of storms can bring in on the shoreline !!

Elena: Thanks for your question! We didn't explicitely look for anthropogenic litter on the beaches after storm events and also within our sampling periods there were no days with great wind events. However, we of course considered wind as an influencing factor and correlated wind data and anthropogenic litter abundance at the beaches of Lake Tollense. We didn't find a significant and consistent correlation, though, for wind direction or wind velocity. But we still believe the wind to influence anthropogenic litter abundances since we observed less litter items on the beach that is

more shielded and not wind-exposed compared to beaches that are more exposed to the pre-dominant wind direction.

Name:...Charlene Trestrail

Question 2:. ..Question for Gisele: Thanks for your presentation, Gisele. Do you think the environmental impacts of cigarette butts are caused by the whole butt itself, or from the microfibers coming out of the butts?

Gisele: Dear Charlene Trestrail! Thank you. It is a good question! In addition to the microfiber present in the filter, which takes years to decompose and are extremely difficult to recycle, they also carry a large amount of toxic materials to the environment and marine and coastal biodiversity.

Name: Romain Tramoy (LEESU, Créteil, France)

Question for gisele: Thank you Gisele. What about educating cigarette producer with a better communication about the polymer used into cigarette buts (on the packaging for example like we do for human health)? Because most people do not know this is made of plastics and it poses no problem to them to litter it

Gisele: Dear, Romain Tramoy! Thank you for your question! I agree this is a great initiative! We are looking forward into that.

#### Name: Joao Frias

Question: Great presentation Gisele. I was wondering whether the use of a market-based instrument such as a higher tax or a local ban on the #1 item collected (cigarettes) would work in this region? Gisele: Dear, João Frias thank you for your question! I believe that it would have positive impacts for there are already municipal laws regarding selling of glass bottled products on the beach and straw prohibition all over the city of Rio Grande. We have samples before and after this laws came into force and we have notice a considerable decrease in the number of this items.

#### Name:...Vincenzo Donnarumma

Question 2:. ..Congrats for your presentation! Do you think runoff can have an impact on these data? Thank you!

Gisele: Thank you for your question! Yes! Runoff is not the main one but it has significative impact in our data.

#### Name : Marie Babinot (Cedre, France)

Question : Have you carried out environmental studies of these cigarette butts according to their brands (environmental degradation, impacts, etc.)? example : marlboro more polluting, degrades slower, etc...

Gisele: Thank you for your question Marie! It would be great do that but most cigarette butts brand logos are no longer visible or hard to be recognized. But It is a great a idea! I believe we could show the companies that it is their responsibilities.

#### Name: Catharina Pieper

Question: What litter items are typically recognized as domestic items on Cassino Beach? what is the difference between source other and undetermined ?

#### Name: Louise Schreyers

Questions for Bruna: Did you also analyze the polymer composition and items size? Can you expand on your sampling methodology? Thank you very much for the presentation!
Cleo (cleo.stratmann@enpc.fr) : just a note when I listened to the nice recap of your session (I joined another one) talking about cigarett butts on beaches, I remebered a beach in Chile in Punta de lobos where I had seen the greatest cigarette-beach system so far: you can see it here (I cannot insert a picture, but I have a website with pictures):

https://plasticoftheworld.wordpress.com/home/pollutiongalery/#jp-carousel-60 scroll down to Chile - southamerica, picture 2 and 3. :)

# Questions and comments for the session 23.3\_O

For any follow-up questions after the session, please contact the speakers via e-mail: Contact Data of speakers: Stephan Rohrbach stephan.rohrbach@ifmb.uni-hannover.de Julia Möller julia.moeller@uni-bayreuth.de Dimitri Seidenath Dimitri.Seidenath@uni-bayreuth.de

Name: István Szabó (szabo.istvan.temi@szie.hu) Question: The five data in the final statistics are coming from different months or five replicates? Thank you!

Name: Cyril Hachemi (chachemi@deakin.edu.au) Thanks for the talk!

Can you point out consequences based on the microbial community regarding the cycling of organic matter in the soil? (i.e. nitrification-denitrification)

Name: Sonja Oberbeckmann

Question: Thank you for the nice talk. Can you exclude the possibility that also residual monomers or additives were degraded instead of /additional to the polymer?

Name:Pascaline Francelle Question: Thank you for the very inldn't the Poteresting talk. Woulyester mesh bag have any influence on your results?

Name: Vincenzo Donnarumma

Question: Interesting talk !!! Have you tried running networks of the different microbial communities of the different polymers found? Stephan ROhrbach: Not yet, but if we get the bioinformatics running, we will include it in our dataset, thanks for the suggestion. Alright, thanks for answering here.

Julia Möller julia.moeller@uni-bayreuth.de

Name: Alexandre Dehaut What about the risk that multiple steps can bring contamination (even if experiments are carried out with caution)? Dimitri Seidenath Dimitri.Seidenath@uni-bayreuth.de

Name: Hannes Laermanns:

Julia, thanx! Great talk and great work! Do you see different effects of the purification on different types of MP? So is there for example a stronger effect on fibres etc. which could result in a underrepresentation of some kinds of MP?

Thank you Julia for your answer! :-)

Name: Kirstie

Question: Great talk - Thank you! Do you see any effect of freeze drying the samples on the mechanical intregity of the plastics? Which would therefore increase the number of plastic fragments found?

Name: konstantinos Papacharalampos which apparatus did you use for the density separation? and did you notice any zncl2 interference when scanning the samples with FTIR

Name: Kristina Klein Thanks for the talk! Do you have a solution for the detection of biodegradable plastics?

I think you can have one more questions (Melanie :-)....you still have 30 minutes and only 1 talk left :-). 1 more

Name: Nisha thanks for your interesting talk. I am intersted to know if you u have looked for the age of sample. If yes, at which wavelength of FTIR?

Name: Andrea

Question: Can you explain "aggregate dispersion" ? Ok so direct water slurry after freeze drying, thanks!

Also: You show a lot of validation for the different steps of the sample clean-up, but what about for the identification of MP by micro-FTIR? Do you have any information about the proportion of detected MP?

Name: Anna

Thanks, great talk, how do you deal with FTIR matches from the library with a low percentage, where do you draw the line for identification? For example, an oxidised HDPE particle could give a very low match from the library, like 50%? anna.winkler@unimi.it great question !!

Melanie: 160 participants ! Great :-)

Name: Nora

Question: Not sure if I missed it, is there also no effect on the eggs of the ants while they are in direct contact with the soils?

Name: Kevin Tallec

Thanks for your presentation. What was the size of the microfibres and polystyrene beads used in the study?

Info (Melanie) Anja Holzinger's talk is tomorrow in Meadows Room part of our CRC1357 Session at 10:30 UCT 11:30 CET

Name: Prasanth Babu

Thanks for the talk. What the motivation behind this study? Are there any effects with soil microbes interaction with ants egg and its effect with microplastics?

# Questions and comments for the session 23.3\_Me

Name: Susanne Belz Q: How does wethering affect the detection by NR fluorescence microscopy?

R: Possibly, at least polymer type influences staining, but we have stained weathered plastics with good results. ~Joana

Name : Cristiane Vidal...

Question 1: .Good morning! Did you combine nile red staining to spectrosopic techniques?

R: Yes, first with ATR-FTIR and now with micro-Raman spectroscopy. For environmental samples, these techniques seem to confirm that we are correctly identifying plastics. ~Joana Thanks! CVidal

Winnie Courtene-Jones: thanks for your talk, can you expand on the wavelengths you just mentioned that don't show up with FTIR

R: For FTIR we have used the following specification and could not get a Nile Red interference: "Fourier transform infrared spectroscopy attenuated total reflectance (FTIR-ATR) in a Perkin Elmer (U.S.A.) Spectrum BX FTIR instrument at a resolution of 4 cm-1, range of 4000–600 nm and 32 scans." (https://doi.org/10.1016/j.scitotenv.2019.07.060) ~Joana

Britta Baechler: Is Nile Red considered a reliable method on its own? Is it considered that you are "validating" suspected microplastics if using this technique alone?

R: Results so far led me to believe that yes, it can be a reliable method as long as sample preparation is correct. It is better than simple visual identification, and it also allows the identification of very small particles under the microscope. So far, Nile Red results seems to be confirmed by spectroscopy techniques. ~Joana

(this was answered already) Winnie Courtene-Jones: have you experimented with different illuminations, and how these influence the 'viability' of the stained particles

R: Yes! In our first test we have tested 254, 365, 395, 470, 495, 530, 625 nm with also different colored filters. The blue 470 nm with orange filter and the UV 254 nm produced the best results. (https://doi.org/10.1016/j.scitotenv.2019.07. 060) ~Joana

So Nile Red should only be used for large MPs that can be inspected with binocular?

R: No, you can actually use it coupled with microscopy to look at very small microplastics (>2 micrometers). I think this is the best application since it allows the identification of very small microplastics that could not easily be identified otherwise. ~Joana

Jordi Valls: What advantages has this technique in comparison with other IR identification and characterization methods? Thank you so much for the information No problem :)

R: Nile Red is not better than methods which allow chemical characterization in terms of validation. It's advantages are: low-cost, high-throughput, low equipment needs, allowing the identification of very small particles, and removing subjectivity from the quantification. The problems are mostly with the staining of organic matter if it is not removed during proper sample preparation. For micro-IR, the problem is the time and equipment access to characterize afilter,but it provides chemical characterization and undoubtful confirmation of plastic. ~Joana

Jyri Tirroniemi: Have you tried any other filters, green or blue?

R: Yes, we have tried several wavelengths and filters, the ones I mentioned worked best! (See above and/or https://doi.org/10.1016/j.scitotenv.2019.07.060) ~Joana

Robby Rynek: Is there any influence of the NR staining dye on IR or Raman spectra of polymers?

R: Dependes on the acquision specifications, but if done right no. ~Joana

Carolin Müller: Thanks for your interesting talk, Joana! Which Nile Red did you use for your study? There seem to be different ones on the market with varying qualities? Also, the correct ratio of Nile Red and Acetone in the stock solution and the working solution was hard to make out. We were about to set up a study using Nile Red as a rapid identification / quantification method, however preliminary trials with different wavelengths ( 360 - 380 nm ex + 415 nmm em; 400 - 415 nm ex + 450 nm em; 440 - 460 nm ex + 500 nm em; 490 - 515 nm ex + 550 nm em) were a little inconclusive. Any tipps or ideas on these? Maybe you would be up to an online meeting with our team to discuss this a little more interactive?

R: We have been using Sigma-Aldrich in ethanol, the lowest concentration which still works is 0.01 mg mL (https://doi.org/10.1016/j.scitotenv.2020.137498). I am a bit confused about the technique you describe. For visual analysis, the 440-460 nm should work well as microplastics should present a yellow to red fluorescence. If you want to talk you can contact me via email: pratajc@ua.pt. ~Joana Thank you very much! We will get back to you via mail. Best, Carolin I will be waiting :) ~Joana Name : ... Question 2:

Veronica: Hello, thank you for your presentation Joana. I would like to ask if Nile Red is an adequate technique for identifying microfibers and what are the main problems related with the identification of microfibers.

R: The main problems is that we do not always see fluorescence from fibers, but pieces of tissue can be stained (https://doi.org/10.1016/j.scitotenv.2019.07.060). This is likely the result of refraction phenomena on the surface of the fiber, with emission being lost and not detected as fluorescence. ~Joana

Amanda Dawson: Can you describe your staining process? Do you stain the filter before removing it from your filtration system?

R: We stain directly in the filtration system, after organic matter removal,for5minutes (https://doi.org/10.1016/j.scitotenv.2020.137498) and then wash with distilled water to remove unbound Nile Red. ~Joana

Richard Cross UKCEH: have you considered counter-staining to check that there is no natural organic contamination to make sure that the nile-red stain does not result in false positive results? I may have missed this as had trouble joining the talk. Thanks

R: We have checked that Nile Red can indeed stain organic matter. We are using a removal of organic matter which seems to work well for water and sediment samples, that afterwards almost no stainable organic matter remains. We have actually already used it to quantify small particles (https://doi.org/10.3390/w12041219) and our posterior testing shows that identification is mostly reliable with some rare exceptions. What we also have seen is that the paper towelsare a great a source of contamination of samples and is stainable! Ps. Richard, I think we have met before in Vienna, hi again! ~Joana

Gissell Lacerot, for Joana. Have you compared results with Nile Red and fluorescence to the results obtained with Polarized light?. What is your opinion on it as a visual method for detection?. Thank you!

R: So far, we have used only visual methods (including collecting photographs in specific conditions and characterizing in ImageJ) and also compared with IR techniques which mostly confirmed that stainable partiles are plastics indeed. ~Joana

Erik van Sebille, for Victor. Very interesting presentation! As I understand it your samples were for the surface only. Have you also taken samples at depth? Would you expect more plastics at depth too?

Thank you Erik for the question. I've tried to answer this already, but I didn't get the whole question. Unfortunately we couldn't samples at different depths. But we do expect plastics at different depths, specially in the downwelling/deep convergences parts as well as in the middle of Langmuir cells, at the retention areas. I don't really know if more or less in terms of numbers but they must be different in terms of density/shape and probably size (smaller particles should better sink with this slow water velocities).

Matthias Egger: You mention that this could be interesting for cleaning the patches. I agree. However, do you have any ideas on how to best spot these cells/foam regions in subtropical gyres? Thank you Matthias for your question. IT is something I have in my mind, although I don't have the tools to do that, but the idea is to try to estimate them from satellite images. Here is an example from foam lines associated to water masses and other coastal effects:

https://earthobservatory.nasa.gov/images/90206/lines-of-foam-on-garabogazkol

Unfortunately I don't know how common these lines are are in subtropical gyres, but there are some articles of macroalges Sargassum in the Sargassum Sea

Kirstie, for Victor: Really interesting talk thank you. Did you try to measure the microplastic contamination from the Research Vessel as well and did you see any difference in concentrations of this bias in the foam line regions compared with outside of the foam lines? Barbara Scholz-Böttcher

Q: Have you found any evidence for fibers (aeolian input) and regarding PET?

Dear Barbara, I almost skip your question now. No, we haven't found any evidence for aeolian inputs and degrading PET, at least in the fraction we were looking to. We found a plastic fiber, but to be honest, I do not know how to know if it is an aeolian input or not. Ho do you know this? In any case, I was just showing the big fragment fraction of the samples (>355µm). We still need to analyze the fraction from 100µm to 355µm.

Thank you again, Víctor

Elena: Thanks for your interesting talk! Did you also find higher concentrations of organic material within the foam, did you analyze this?

Thank you Elena, not unfortunately we got read out of the foam to continue with the sampling analyses. It would be interesting also to analyze it too. I don't know if the answer will get to you as I don't know your surname. Best, Víctor

Delphine Lobelle: Thank you for a great presentation! Do you know how common these foam lines are globally? Can they have an impact on the distribution of global surface concentrations? Hi Delphine, thank you for the very interesting question. I answer something related to van Sebille

before. I don't have the tools to do this, but the idea is to try to estimate them from satellite images. Here is an example from foam lines associated to water masses and other coastal effects:

https://earthobservatory.nasa.gov/images/90206/lines-of-foam-on-garabogazkol

They can be very variable over time, but strong windy regions would be a start targeting area to start looking for them to try to estimate these foam lines.

# Questions and comments for the session 23.3\_Ma

Name: Gerson Fernandino

Question: Hello, Felipe! Thanks for your presentation! Congratulations on your work! I wonder if there is a chance of cross-contaminating your sample as a result of cutting the PVC corer? Hello, Ju! Good to see you too! ;) Awesome. Thank you!

Name: Gerson Fernandino

Question: Thank you for your presentation, Décio! Awesome. I would like to ask what were the types of microplastics found during the 1940s. Considering that plastics entered our homes in the laste 1950s, I would expect another different source than household and sewage. Thank you!

Name: Aline Carvalho

Question: Thank you a lot for sharing your work! I was wondering about the changes in shape of microplastics. If they stayed longer in the environment could they suffered different/more intense degradation process that woul affect their overall appearance? Would we be able to distinguish between MP profiles along the time?

Name: Simone Lechthaler Question: Thank you very much for your interesting work! I might have missed it but could you explain again how you have assigned the sediment layer to a deposition in 1940? Since mass production started in 1950 this is an interesting aspect.

Name: Gerson Fernandino Question: Did you find any tire microparticles during the early decades assessed?

Will there be a way to watch the presentations later? Something went wrong with the website for me and i missed the first part of the session Hi!!! Nice to see you!! And thank you! I was waiting for your talk but it was showing the session before

Name: Laura Markley Question: What was the dating method for the core? (May have missed it - great talk!)

Name: Gerson Fernandino Question: Awesome presentation, Fernanda! Congratulations!

Name: Crislaine Question: Fernanda, did you manage to identify the polymeric composition of plastics? Excellent work, thank you!]

Hi Crislaine! Yes Which kind of polymers did you found? thanks for question. We perfomed FTIR and TGA analysis and the samples are composed mainly of polyethylene (:

Yes! Good talk, do you have any ideas what you think the best ways of identifying micro/macro plastics in the future might be? Chemical, visual (i.e, technofossils), etc? We see algenan etc. preserved over millions of years, do you think plastics could be the same?

Thanks Juliana! Excellent set of talks, i really enjoyed them :)

name: Gerson Fernandino

Question: I would like to congratulate MICRO, Juliana and all the presenters for discussing this topic. It is amazing that this somewhat novel aspect of the plastic pollution sit is being discussed by the scientific community. I am really happy for that! Thank you!

Regarding the question asked by Juliana, I believe that macroplastics and microplastic yields different possibilities for approaching the geological aspect of marine litter.

Nice presentations, guys!! Thanks for sharing such amazing works.

# Questions and comments for the session 23.4\_O

For Charlotte Lefebre. Thanks for a great presentation. Do you think it could be higher winter counts due to

particle ejection in the winter due to stronger onshore winds? Steve Allen - Thank you. I am sorry, I am not sure I understand what you mean by particle ejection ? The ejection from the sea by bubble busrt and sea spray.

We did our study in the gukf Du Gascoyne measuring atmospheric MP coming ot of the sea. It was Autumn with strong wave action. Just wondered if a lot of the winter particles could be being trasported to the beach. - Thank you for ths complementary information. I was not aware of this kind of process that is ejection of plastic particles. Here is the article. Hope it helps

https://journals.plos.org/plosone/article?id=10.1371/journal.pone.0232746 - Thanks I will read this ! It awaken my curiosity

### FOR MAURITS

Name:...Francisca Ribeiro (The University of Queensland)

Question 1:...What was the pore size of the filters you used? So, what was the minimum size of microplastics you could detect? What types of plastics can you quantify with your current method? PVC, PET, PE, PP, PC, PMMA, PS, PUR-MDI, PA6

Name:...Eleni Christoforou

Question 2:...Couldn't cryomilling the mussel tissue cut down microplastics too and thus make it harder to detect and identify? I hope I answered the question already?!

Name: Mohammad Wazne

Did you observe any physical and/or chemical differences between the same polymer extracted from year to another?

Name : Marie Babinot

Question : Have you also worked on the impact of these mPs on mussels? No

Name: Alexandre Dehaut

The LOD and LOQ are the ones described in Fischer et al. papers?

Were your polymers markers potentially altered by weathering? Yes they are. So far we havent seen a big influence by weathering on our markers (with the few test we done)

Name: Holly Nel - Could your method be better at detecting certain polymer types over others? If you look in the papers by Fischer et al. You will see that certain polymers have different LOQs and LODs. Thank you.

Microplastics analysis in environmental samples – recent pyrolysis-gas chromatography-mass spectrometry method improvements to increase the reliability of mass-related data (Fischer et al. 2019)

Name: Kristina Klein Is there any chance that the detection of e.g. PVC is a contamination based on the sampling back in the day?

I hope i answer that - you did, thank you :-) you are welcome

### FOR LUANA

#### Name: Nora

Question: For weathering of the polymers with simulated solar radiation you say that 40 hours represents 3 years of irradiation, how high was the irradiance of your UV Lamp? And how high were the temperatures reached with this lamp? For the irradiance, the actinometry method could reveal the irradiance but unfortunately this calculation i didn't have done. About the temperatures, the lamp can reach more than 40°C. I'm saying this temperature because as I said in the discussion, we cared that the temperature didn't reach more than 40°C because could cause reactions in the polymers that are not the focus of this work.

#### Name: Winnie Courtene-Jones

Question: how long were the polymers left to extract the leachate? 30 hours exposed to UV lamp.

#### Zhiyue Niu

In terms of liquid/solid ratio for leaching, are the values you used comparable to other studies? I didn't investigate this aspect in this work but it could be interesting. I'm very glad to you call my attention about this chemical aspect.

Unless I missed this information in your slide, but did you compare the 2h, 8h, and 30h with a control? Non-aged samples..? Yes! Virgin particles were used representing non-aged samples. FOR CHARLOTTE:

#### Name: Bettie

Question: Thanks Charlotte for your nice presentation. Did you take into consideration the fact that during summer, beaches are cleaned up for turistic uses of beaches? It might explain the "absence" of microplastics or isn't it linked? - Thank you Bettie, Did I answer your question?

Name: Xavier Cousin

Question: Hi Charlotte (thanks a lot for your presentation) do you have an idea on MPs origin ? oceanic or riverine/internal human activity ? Thank you Xavier, I hope I answered to your question Yes thanks ;) :D

#### Name: Winnie Courtene-Jones

Question: Thank you, very interesting. why did you decide to analyse 86% of the plastics? what made you choose 86% Thank you, I hope my answer was clear!

### Christophe Maes

Q: what could be the impact or importance of the tourism and human activity during summer? Considering MP pollution, I think that tourism generate maybe more fiber-shape particles -due to laundry for example. However, for this study that considered "big" MP that beached with the tide, tiny fiber were not considered

Marie Babinot : Thank you Charlotte! Did you not have difficulties to pass the 0.5-1mm fraction to the FTIR? Sorry Marie (two times I do not say your question, sorry!!!(Ika) ;-) Particle that have a size superior to 0.5 mm responded very well to the ATR-FTIR identification method, once you catch them with tweezers!

Winnie Courtene-Jones

Q: you looked at seasonal distribution - how many times did you sample each month? - Sampling was made once a month- did you remove all pieces of litter that you found - all particle were removed to be studied at the lab (shape, polymer,...) could it be that some of the pieces were not removed from the beach, and that they were counted in multiple surveys? - as they were removed each time, they cannot be count several times - Thank You :) - You're welcome!

Thank you for all your questions and your interest. Do not hesitate to ask more questions, or give suggestions at this mail : charlotte.lefebvre.1@u-bordeaux.fr

FOR CHRISTINE cknauss@umces.edu Question:Very interesting work! I wonder why you chose these two polymer types?These two polymers are widely used as fibers and are also easily commercially available. I purchased them both from Goodfellow. Name:Zhiyue Niu

Question: Many thanks! Any evidence of ingestion? Name Arnaud Huvet

Question :Thank you for the interesting talk. I have a methodological question: Did you produce these small fibers? And do you know if the fibers were ingested and egested? Yes I made these fibers using a modified method from Cole 2016. Name: Claudia Drago

Q: Thank you for your presentation. According to your plots, at T0 you used larvae displaying a variability in terms of size, did you check if all larvae have the same developmental stage? Thank you for your answer :) Name: Kevin Tallec

Q: Did you determined the size of your fibres during exposure, i.e did agglomoration occur? Mark Hartl

# Questions and comments for the session 23.4\_Me

Peter Vermeiren: Interesting talk. Do you have any ideas on how to calibrate the model to identify which processes and how strongly they are affected. Thanks (p.vermeiren@science.ru.nl)

Name:...Stephan Rohrbach Question 2:... Is there a correlation between MP effect on the energy flow and the size of the animal? Thanks a lot

Name: Ana Lacerda Question: Very nice talk!! I was wondering about environmental samples. How to measure the effects of microplastics (MPs) together with other effects to see the real impacts of MPs in the DEB model? Thank you so much Charlenge

Thank you so much, Charlene!

Name: Matt Cole Talk: Charlene Question: Lovely clear talk. Does the DEB model allow for perturbations from both physical and chemical effects stemming from plastic?

Name: Erik van Sebille

Question: Very cool talk. What is the key parameter that you'd need to know (and that we don;t know yet) that controls the model? How could that parameter be measured?

Name: Joao Frias Question: Brilliant talk. What kind of biomarkers would you consider using in relation to DEB model? Would this work on behavioural studies? Thanks Charlene.

Miriam Weber: Great discussion! Thanks a lot for that!

From Britta Baechler: Great presentation, thank you! Could you contextualize these results for ushow frequently are suspected vessel paint particles detected in sea surface water samples? Are they also present in sediments?

I cannot hear the presentation anymore

From Giuseppe Suaria: Thank you, very interesting presentation. How can you be sure that those paint particles were collected from the sea surface and they did not enter the net while sitting on the ship's deck?

Also airborne contamination on board from paint flakes of the vessel? What was the towing position of manta net, behind the vessel or on the side? Stjepan Budimir

From Miriam/HYDRA: Thank you for adressing paint particles! From a more general point of view what is your recommendation to detect best and most efficient other sources of paint sources in our everydays life?

Clara Leistenschneider: @ Miriam/Hydra: Maybe you could investigate rain/stormwater runoff to evaluate land based sources for paint particles and to track their way into ater bodies .

### ALESSIO PRESENTATION

Alexandre Dehaut How quite "big" MP can be found in the hepatopanceras of Nephrops norvegicus?

From Britta Baechler: What is your hypothesis for why there are more MP in the hepatopancreas than the gut? Has this been investigated in shrimps or other arthropods as well?

Stephen Kneel

What digestion protocol did you follow for this extraction of MPs?

Miriam/HYDRA: What would be the effects in the different compartments of the organism? Absolutly, very complex, very nice overview, grazie Michela!

## Questions and comments for the session 23.4\_Ma

Name: Aline Carvalho

Question 1: Thank you for your presentation, Garth.Do you think the feeding mode plays a role in the microplastic abundance in the gut content? Regarding the first question, there was a difference in MP composition between different species?

Name:Forane Le Bihanic

Question 2:Thank you very much for your presentation. Sorry if I missed it in your presentation. Did you identify polymer type of your particles? And if not, how do you think spectroscopy analysis could influence your data? Ok thanks that is clearer!

Name: Michaela Miller Question 3: I might've missed it but for determining if an item in your samples was contamination or not, what criteria did you use?

Name: Rachel Coppock Question: Would you be happy to share the protocol you used to determine the remainder of 30% of non-plastic particles? Thanks :)

Winnie Courtene-Jones (winnie.courtene-jones@plymouth.ac.uk): Question: the method to extrapolate the remaining particles sounds interesting, could you provide details or a reference for this

Andrea: I agree I would like to hear more about the extrapolation for the remaining 30%

Garth: Thanks everyone!! I can definitely share the method for extrapolation. I used a multinomial regression model via neural networks with the nnet package in R. The predictors I used in the model were whether I thought a particle looked like a microplastic or not and the size, shape, and colour of the particle, as well as what kind of sample it was in. I cross-validated the model and was able to get about a 80% success rate for predicting whether or not a particle was actually plastic on test data. Rachel: Thanks! I look forward to seeing the paper :)

Andrea: Hi Garth, this is super interesting! Curious about where you got the size and shape data. Did you image all of the plastics or measure individually? Also, greetings from a fellow Canadian (from Norway) :)

Hi Andrea!! Yep, we individually imaged and measured every potential microplastic particle. Let me just say that going through all these samples took about 8 months with a lab technician working 20-30 hours per week the whole time!!!

Hi Garth: Sounds very intensive. Did you consider using ParticleScout or something similar? Also I assume that you're predictive model could not predict polymer type. Was this excluded from the remaining 30% ?

The things was that based on how we were IDing the particles they had to be individually moved from the filter onto tape for Raman ID. Also we were using a pretty simple compound microscope so had to manually move through the sample at 100X magnification. And you're right, the model was

just for categorizing into plastic, semi-synthetic (rayon), natural, or natural anthropogenic (coloured cotton and wool). I'll be separately plotting and discussing the more specific IDs of particles. (Hope that makes sense.)

Julieta: Thank you Garth for your presentatition, it was excellent. I want to ask you, why did you choose to use bayesian GLMM and no the frequentists statistics

Garth: Hi Julieta and thank you! I used Bayesian because it allowed me to build a more complex model where I could have a hierarchical model structure with "true" concentration nested within the model. I also built in uncertainty about various things, for example the trophic position estimates and it gives a lot of flexibility in doing model predictions. Sorry, it's a bit complicated and hard to explain here, but the paper should be submitted in the next few months!

Julieta: Perfect ! and Congrats abotu the paper, can i contact you in a future to talk about the model? I wll currently study trofic transfers of microplastics !

Garth: yes, absolutely! My email is gcov@uvic.ca for anyone with any other questions as well!

Gerson Fernandino: Thank you for your presentation, Marina! Excellent work! Cheers

Marie Babinot : Thank you Marina ! What salt solution did you use for density difference separation?

Matthias Egger: Great talk, Marina! How do your results/concentrations compare to what was found in

other studies in the GBR?

Marina:L Hi Matthias, here goes the free platform with current concentrations and future predictions of mp concentrations in sea surface: https://rshiny.lifewatch.be/ng-ocean-plastic-challenge/ And their paper is: https://doi.org/10.1016/j.envpol.2020.115499

Thanks, Marina! :)

Garth: this is really cool, thanks for sharing Marina!

No worries! It's indeed very cool!! I couldn't hold my self when I first saw it hehe

Georgie savage: what species of corals did you sample and what chemical did you use to digest them? Great talk!

Hi Marina, what concentration of the nitric acid did you use?

Marina: Hi Georgie, the hardest question was the full species name! Hehe I had to change it last minute and guess I what I could not remember today?! Here goes the species name: Dipsastraea lizardensis

I used 65% HNO3 for 6h - but I tested other species (e.g. acropora millipora) that could be digested in 3h! thank you!!

Alicia Mateos Cárdenas - Marina, thanks for your presentation. It was very clear, could you please share your email address I'd like to send you an email about your coral methodology :) thanks Marina: Hi Alicia, sure I can here it goes: marina.santana@my.jcu.edu.au

Jen Jones: Hi Tainah, great talk! How do you think we can encourage more people to share photos as citizen scientists?

Tainah: Thank you, Jones! I believe the first step is to show how important it can be, since citizen science has great potential to include data from worldwide. Secondly, there are some web pages that

we can create a citizen science campaign in many different subjects, and this is certainly a great way to encourage people to share their photos!

Marina: Hi Tainah, that's fantastic! I was going to ask the same as Jen Josen but will change my question to how to have you need any limitation in using the citizen science approach and how to improve it (in case you did)?

Tainah: Hi Marina, thanks! My limitation was that I didn't create a formal campaign on those web platforms that I just told about, so I had to ask people directly through social media pages. Marina: but you could still make the info available by using them so good job! Really interesting indeed. (:

Tainah: Yesss, that's for sure! Thank you! (:

Georgie: Great talk Jen!

Hi Jen Great talk!

How do you think we can monitor transient and endangered species across what I guess is a large area?

Question from Erik van Sebille

Hi Jen. Nice talk! How do you make sure that your risk assessment does not bias towards charismatic megafauna just because more is known about these?

Hi Erik! Excellent question, there is no denying that this scoring is certainly biased to whatever species are easiest to sample and that have been previously studied. I think its important to highlight that the low scoring species are not considered low risk, more data deficient at this point. Good point. But then if you present the top-X number according to your scoring, you'd draw attention to these anyways? It's a bit of a positive feedback :-(

True, I guess it is important to prioritise the species already that are very habitat limited/threatened in other ways but also to show that we need more understanding of food web dynamics and to fill some of those invertebrate and fish gaps we have currently in the literature!

OK, I see. Thanks! Still really cool work of course!! Thanks, I am certainly learning a lot :)

Patricia: Great talk Jen!

How do you classify the impact of the entanglment (minor, major...)? It was already determinate at the literature?

Hi Patricia, thanks! We designed scoring that was linked with severity of harm e.g. injury to mortality and frequency of reporting. Ok, thanks! Great job

Marina: Hi Jen, well done! amazing work!! I might have missed but could you explain a bit more the matrix you used for the risk assignment (guess my question goes in line with Patricia's one). Also, is this work published already?! Would very much like to read more about it!

Thanks so much! The paper was submitted last week so fingers crossed for news soon! The risk assessment scoring was made of four parts - distribution (to highlight endemics), conservation status, lit evidence for entanglement and for ingestion. Happy to share more and would love to chat to anyone doing anything similar in other places including with citizen science: jj407@exeter.ac.uk

## **Questions and comments for the session 23.5\_O**

Name:...

Question 1:...

Adam Porter: Hi there; really great talk. Well done on getting all of those stakeholders signed up and engaged. I just have a question based on the bioplastics you are recommending. Have the bioplastics you are recommending a switch to been screened for their ecotoxicology and impacts as I know this field is in its relative infancy.

Name: Tim van Emmerik

Question 2: Thanks for the presentation, I was wondering if you plan to facilitate/encourage any field-based studies to better estimate the plastic sources into the Med sea?

The link to the IUCN report : https://www.beyondplasticmed.org/en/uicn-publishes-a-new-report/

Presentation S.Primpke Contact: sebastian.primpke@awi.de, jesvollertsen@build.aau.dk

Name: Alexandre Dehaut

Regarding the algorithm of siMPle. Pearson corelation are carried out on the whole spetrum or only on peaks? On a pre-treated spectrum (normalized) or raw data?

Name: Josef Brandt Question to Sebastian Primpke: Do you plan including chemometric approaches (PCA, PLS-DA, RDF, ...) for spectra evaluation, rather than spectra database matching? Great project, anyways!

Name: Alexandre Dehaut Is there a plan to have a shared database to bring our (curated) spectra and add especially atypic spectra (weathered particles, etc.)?

### **Questions and comments for the session 23.5\_Me**

For more information on microplastiX: https://www.microplastix.org/ MicroplastiX-Team: Related to the question which organisms will be used in our project: to cover different trophic levels, key fish taxa with different feeding habits, zooplankton organisms (copepods, echinoderm larvae) and jellyfish will be studied. Also, if you want to discuss more, feel free to join our poster presentation tonight: 19h30-20h, Meadows's room. Session posters\_23.7\_Me Chaired by Yannick Lerat, La Trinité sur Mer

Name : Aaron Beck Question 1: Hi Richard, you mentioned hyperspectral analysis. Is that for macroplastics (eg, from air) or microplastics?

Name:...Patrizia Ziveri

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Question 2:...Thanks Richard for the project overview. Are there any specific target regions addressed?

Matthias Egger: Great talk and project, Aaron! Will you also study (spatio-)temporal variability in fluxes (lateral & vertical)?

Name:...Cleo Question 4: Thanks for the project presentation. Will you account for riverine inputs, in what way?

Name Eike Esders Question: Is the spatial distribution of MP concentrations different/same in the whole water column?

Catharina Pieper

Q: I wonder if you are sampling the whole water column and how deep you think you sample in the open ocean? and what would be the key methods to sample vertically?

Name: Mercier Matthieu Question: I missed some of the presentations, do you plan to quantify turbulence in the ocean as well as plastic?

Name: Nia Jones Questions: Great talk, really interesting project! What hydrodynamic model will you be using?

# Questions and comments for the session 23.5\_Ma

I think the screen sharing issue is due to the fact that you just shared the single window of your folder, Bart Koelmans...

Name: Britta Baechler Question 1: Thank you Judith! I understand there is a great deal of variability/error with visual microscopy.. what are your thoughts of this method as an initial step then chemical validation (FTIR, etc) afterward?

Name: Alexandre Dehaut Thanks for your presentation. Is it possible not to feed animals taking into account animal welfare and ethics? Name:... Question 2:...You might need to activate the computer audio first

Bettie Cormier, Thanks a lot for your presentation. Just quick questions on materials, did you expose your cells to beads directly or did you do an organic extraction using DMSO or so? And is the concentration of pollutant environmentally realistic or not?

Thomas Witzmann, question to Rubin Andrey Ethan: Have you also measured the surface charge of your particles? This might influence the adsorption of Triclosan.

Zhiyue Niu

Thanks for the presentation. Sorry i was a bit lost. If im' corrected, you exposed the microalgae to beads with a concentration of 125 p/L and saw effect on growth by some polymer types? And are those beads you used are additive-free?

Presentation of Julia REICHEL

Name Alexandre Dehaut What is the risk to not extract all compounds given the huge amount of sample (100 mg) compared to Py-GC/MS? Is 200°C enough to extract the tageted samples? How did you the calibration of the quantification?

### Questions and comments for the session 23.6\_O

Name: Susanne Belz Q: Do you have info about the weathering status of fibres you found?

Name:...Marie Russell Question 1:..This presentation now running is from an earlier session.

The presentation is hardly understood. The connection is really bad.`I'm sorry the connection issue made it hard to follow! You can view my presentation here:https://vimeo.com/482713963 (Sam)

I'm having the same experience

Me too

Is it possible for someone else to host the connection for the upcoming talks in this session?

Name:..Ana Luzia. Question 2:...Firstly, congrats for your talk! The connection was not good indeead, so sorry if I missed it, but based on your results which method do you recommend to be used?

Name:..Ester Carreras

Question 3: Congrats on your talk!! Do you think that different polymers have different visual appearances? Particularly, fibres, eg. polyamide and polyester.

Anja Rebelein:

Question: Great talk! Is visual identification enough for analyzing fibers? Sam Athey - Thanks Anja! Unfortunately, visual identification is not always reliable for distinguishing natural from synthetic and semi-synthetic fibers. It's best to use some sort of analytical method for determining polymer composition, in addition microscopy could be useful to confirm spectroscopic ID for some fiber types (e.g., rayon, cotton).

Same problem... we can't understand well. Unfortunately... I'm sorry the connection issue made it hard to follow! You can view my presentation here:https://vimeo.com/482713963 (Sam)

Question for Giuseppe, Thanks for the talk, can you explain the choice of correction for contamination, would you consider using the limit of detection instead? I really enjoyed reading the publication.

#### Erik van Sebille

Very nice presentation Giuseppe! Do you think some of the difference between celluloic and synthetic fibres might be due to buoyancy differences? Do you know how buoyant celluloic fibres are?

#### Ana Lacerda

Hi Giuseppe, very interesting presentation, thank you! Where in the globe there was no fibres as you found them in 99% of samples? And also what was the composition of most black fibers? Based on FTIR do you think they are coming from tyres? I do not know if I missed due the bad connection... Perfect, thank you!!

Tim van Emmerik: Really cool work! If you can pick another ~25,000 items to analyze, and you're free to choose the size range. Would you focus on fibers or larger items?

#### Name: Jakob Cyvin

Question: Thanks for the talk! Do you have any recomendations for easy analysis to distinguise natural fibers from plastic fibers without using Raman, FT-IR or GC-MS? e.g for use in schooles/citizen science/time effective analysis. e.g. Oxidation? Hi Jacob, check this publication: https://pubs.acs.org/doi/full/10.1021/acs.est.9b05262 Zhu et al. 2019 Identification of Microfibers in the Environment Using Multiple Lines of Evidence

Patrizia Ziveri

Thanks for the excellent presentation. You hypothesise different residence time for fibers vs other MP debris. How do you explain this mecchanism?

Dan Wilson: Thanks for a really interesting presentation! How did you work out/calculate the correction for mesh net size?

Thank you, will also check out the paper!

Thank you, here's the link: https://doi.org/10.1016/j.envpol.2019.113413 but i can also send the pdf if interested! Thanks! Just used the link to download the paper! :)

Darshika: Very interesting presentation Giuseppe! based on your answer to Erik's question, fibres sink, then we should expect higher concentration in the seafloor? any studies on that? Sure, check (amongst the many others), this nice paper from Ana Sanchez-Vidal: https://doi.org/10.1371/journal.pone.0207033 The imprint of microfibres in southern European deep seas Thank you!

Name: Susanne Belz

Q: Do you have info about the weathering status of fibres you found?

Unfortunately no, but some of them seemed quite "encrusted" or "degraded", but this are just my feelings

Name: Adrián López-Rosales

Doesn't fibers "stick" (estatic electricity) in u-ATR measuring?

Yes, they tend to do that... :) we would normally put them in a tiny drop of water on a microscope slide and let them dry, so that they would preferentially stick to the glass slide rather than onto the ATR crystal

Name: Charlene Lujan

I am interested in the paper that Giuseppe mentioned about visual differentiation of natural and synthetic microfibers. Is there any way to get his email or maybe share over here the paper? Thanks Hi Charlene, here's the paper i mentioned: https://pubs.acs.org/doi/full/10.1021/acs.est.9b05262 Zhu et al. 2019 Identification of Microfibers in the Environment Using Multiple Lines of Evidence Thank you!!!

You're welcome! and also this one might be useful: https://doi.org/10.1016/j.mex.2019.11.032 Prata et al. 2020 An easy method for processing and identification of natural and synthetic microfibers and microplastics in indoor and outdoor air

There is also this interesting paper including some information of fibre id: https://journals.sagepub.com/doi/10.1177/0003702820930733 Thank you!

Questions for Lasse Rasmussen :

Name: Francisca Ribeiro

Question: Great talk! Have you tried to analyse and quantify rubber? Can I ask what type of markers? Great! Thanks

Winnie Courtene-Jones

Q: Thanks for your talk, did you mention which markers you used to ID tyre wear in the pry-GCMS? - great thank you for that

Elisabeth Rødland Interesting talk! What method of pyrolysis for tire particles have you used? For example the ISO method or similar? Thanks for the clarification!

Marco Mattonai, University of Pisa Thanks for the very interesting talk! Could you explain why you need two SPT steps during sample preparation? Thank you

Tomasz Burghardt: Good job. Have you identified any particles that could be assigned to road markings?

Reza Shiravani Question: Thanks for your great work. Did you find a relationship between concentrations in particle/m3 and mass/m3?

Winnie Courtene-Jones Have you analysed other types of 'road' other than pervious pavements to see if they differ in capture of these various particles? i was wondering what you plan to do next (if this project is ongoing)? - fascinating work, thank you for this. sounds like an interesting system to study- very best with your PhD work!

Lasse Rasmussen - Thanks for all the questions! If you have any further don't hesitate to contact me at lar@civil.aau.dk

To Mateo:

Cleo: What kind of corruptions contribute to mismanaged waste?

Miriam/HYDRA: Very intersting insights, many thanks. Form your experience, what are other (similar) factors to have such influence like corruption or education? Great insight, thank you!

Cleo: Similarly question, what about for example rising prices, or increasing taxes ? Were these included in the models?

Thanks, great interesting research;

# Questions and comments for the session 23.6\_Me

Matt Cole: Thanks for the positive feedback, if there are any further questions happy to chat via email (mcol@pml.ac.uk) or Twitter (@SciMatty)

Name: Simon Wieland

Question 1:...Thank you for your talk! How many mussels would you need to "plant" at a wastewater outlet to compensate for its microplastics? Is this realistically achievable? [Matt: We've done back of the envelope calculations, and hope to address this in the paper, thank you]

Hi Matt lovely talk and amazing system. What is the environmental impact of creating more POM (poop) in a natural system where perhaps mussels don't exist currently? [Matt: Plan is to collect the POM/poop to collect the plastic... is a good question of aquaculture though... I think I've read some papers that suggest benthic communities under mussel farms do quite well!]

Name:...Arnaud Huvet

Question 2:...

Thank you Matt! Did you test the range of microplastics size properlu filtered by mussels? from minimal up to maximal sizes? MERCI MATT [Matt; thanks Arnaud!]

Winnie Courtene-Jones:

Fantastic talk (as always), it's a really interesting concept! have you considered how mussel ropes might affect the flow dynamics of the water? could it also affect the dynamics of the river discharges etc. would this have a knock on impact to any other biogeochem processes in the 'real-world'? - excellent work, Thank you! [Matt: Thanks Winnie!]

Kevin Tallec: Thank you Matthew for this very interesting presentation. Did you assess potential effect of new mussel populations in coastal systems? [Matt: Definitely being consider in next stage... we've had a lot of M. edulis population loss in SW ENgland, so hope the new populations could help

re-seed native populations. Important to use native species]. Thank you, It is a very interesting project, I look forward to seeing your results :)

### Name: Eleni Christoforou

Questions: Thank you for your talk! As many MPs accumulate in the mussels tissue and digestive system, what would be the fate of mussels and the MPs within after they are saturated and they are not able to filter any further or die? [Matt; thanks for great question!]

#### Name: Charlene Trestrail;

Question: Such an interesting project, Matt! Do you think there would be enough food in the deployment areas to keep the mussel populations alive for a significant period of time? Would mussel density be too high for them all to be fed adequately? [Matt: Great question; in the model the food availability is accounted for, but obviously this is at river mouths where food is available... I'd imagine near to swage outputs there would be plenty of organics to keep them going, but obviously there are other pollutants that come out of wastewater (oestrogenic compounds etc) that may pose a risk too.]

#### Name: Tony Walker

Great talk Matt. what do you envision for mussels after deployment? Will they themselves be used a MP removal tool, at the end of life to prevent predation by scavangers? [Matt: Hi Tony, There was some consideration as to whether we could use them as biofuel... any suggestions?]

#### Name: Nisha

Great talk matt! I am wondering if different type of mussels may have different response towards the removal of microplastics. Any comment on this. [Matt: Important thing is to use native/local species so we don't spread non-native species... scope for different species, but mussels are particularly robust] Thank you..

### Name: Christine Knauss

Hi Matt! Thanks for a great talk. You found that mussels are concentrating microplastics in the sediment right under the mussels, which could make the microplastics easier to contain and collect. However, is this just shifting risk from one area to another? Would love to hear more about your ideal end goal is for this work (ie trying to collect the feces to collect the plastics). [Matt: Initially I was thinking we'd shift plastic to sediments where it can be "locked away"; I've come around to thinking collecting the plastic-laden poop would be better for fully removing the plastic. Thanks Matt. It seems like it! Especially since water currents dont seem to re suspend the particles in the models.

### Name: Gerardo Pulido-Reyes

Thank you for a very nice presentation. I really enjoyed it! ; ) I was wondering if you have also tested a control set up without mussels in order to check for any microplastic loss in the system (through adsorption to the walls, pumps, etc.). Many thanks! [Matt: Yes, absolutely; it took a LOT of tinkering to get the microplastic stable. We have in-line pumps helping to keep everything in momentum. The plastic levels were confirmed to be stable before and without adding mussels. Thanks for you answer! ; )

#### Name : Romain Tramoy

Hi, Will you use the national (France) platform RemedZero plastic to bankarise your data? https://www.remed-zero-plastique.org/

Please check :) Romain, isn't it just for the South of France ? Thank you for this information, I'll have a look at this platform . We are in the north west of France . Yes but if I remembered well, RemedZero is a platform for the South of France, but maybe in Brittany you can find something similar (Ifremer?). Anyway; nice talk, thanks for it! We bankarise our data in the Ifremer platform DALI at the time through OSPAR project and also with the app Fish and Click developped by IFREMER

Name: Elena Hengstmann

Question: Thanks for the presentation of your results! Did you also find other plastic particles (e.g. fragments) or did you concentrate on nurdles only? If you found plastic fragments as well, can you tell something about the relationship between nurdles and other plastic particles? Was there also a higher plastic contamination in total when you found a hotspot of nurdles? Thanks for your detailed answer!

Name: Maja Grünzner

Question: Thank you for your insightful presentation. Who do you think should be targeted to reduce the nurdles pollution? Industry who use them to produce plastic products (and are responsible for spillages) or society who consume the plastic products from nurdles?

# Questions and comments for the session 23.7\_O

Name: Fernanda Santos Question 2: Congratulations for the work! You have mapped the source of the pyroplastics?

Julius A. Ellrich: Thank you very much Fernanda. Actually, we did not map where we found the pyroplastic, but we provide the GPS coordinates where we found it in our paper. On that beach, there were some camprifre leftovers and we detected a few PET bottles. So, it could be that the detected pyroplastic resulted from such a melted PET bottle. Name:Rocío Rodríguez Torres Question 3: Could you explain briefly again what do you define as a pyroplastic?

Julius A. Ellrich: Dear Rocio, pyroplastic is burned (melted) plastic that looks like a natural piece of rockQuestions and comments for the session 23.7 Me

# Questions and comments for the session 23.7\_Me

Joao Frias presentation

Name: Alexandre Dehaut Question 1: Is there some ecotox tox studies in this project?

Ula Rozman presentation

Špela Korez:

thank you for your talk. why did you choose 13/15 week s sampling Stephan Rohrbach:

How different are your particles. Have you considered different surface areas and where do you get the particles and do you know what kind of plastic or some other properties.

Delphine Lobelle: Thank you! Were currents simulated in your freshwater samples or was the water 'static'?

Name:...

Question 2:...

Hazimah: How do you distinguish/separate between loosely attached bacteria from the suspension or the actual biofilm (strongly attached one)?

Joao Frias: Will you intend in the future to do species characterisation of the biofilm attached?

How did you rinse the particles? over filter/sieve?

# Questions and comments for the session 23.7\_Ma

Name:.Anna Winkler

Question 1:. To Antònia Solomando: Congratulations to your poster, very interesting! Could you please name the LDPE concentration and size+shape of the particles? I only saw the poster uploaded but not in the live stream... just saying

Name:...Judith Weis

Question 2:...For the Sparus poster, you gave them microplastics as10% of the diet - is that correct? Isn't that a very high concentration?

For D Silva - do you think bivales in aquaculture would have different concentrations of MPs than the same species growing wild? Do aquaculture conditions promote uptake of more MPs?

Stephen Kneel

For D Silva - How did you extract MPS from the bivalves you studied? I am working with cockles. What pore size did you use to collect MPs when you filtered digested bivalves?

# Questions and comments for the session 23.6\_Ma

IMPORTANT: Please include your name at the beginning of your question or scientifically-relevant comments.

You can follow the live-stream discussion between the session speakers and the chairperson through the dedicated channel on the conference home page during the specified hours in the programme. The chairperson will draw upon your questions to the speakers during the LIVE discussion. IMPORTANT: Any questions or comments without the commenter's name will be removed, kindly.

# **Questions and comments for the session 23.6\_Ma**

Name: Xavier Cousin

Question 1: Thanks for your nice talk ! Do you think it is possible that some differences come from the size difference ? Example fibres smaller than beads (20 µm is quite large for acartia)

Name:Zhiyue Niu Question 2:...Nice work!! Have you check the evidence of ingestion (e.g. SEM/fluorescent microscope images)

Name: Anita Jemec Question 2:...Very interesting work. Can you please explain based on what these two types infochemicals were chosen? Why is there a difference in the result between DMS and DMSP? Any biological explanation? How were the concentrations chosen? Thank you! Very good study

Name: Gissell Lacerot Question: Did you obtain your own fibers?. Could you elaborate on the methodology to obtain that size range?. Thank you very much for your talk.

Name: Valentina Fagiano Question: Very interesting work! Did you check also the influence of MPs shapes on egestion mechanisms? Or they concentration into fecal pellet? Thank you

Name: Nithin

Question: What would be the biggest challenge to do similar study in field condition? Thank you very much

Presentation Defri Yona

Name: Alexandre Dehaut Thanks for your presentation. I would have two/three questions: -What were the average weight of tested fish? Were the whole fish analysed or only subsample (like 5g or 20 g)? -How did you deal with the risk of contamination? Controls?

Name: Ciara Keating

Thanks for the very nice presentation. I might have missed it but did you notice a poor condition factor (measure of fish health) in fish with a higher microplastic load?

Name: Stephan Rohrbach

What is your assumption how fibres enter the muscle tissue? Is it by blood or rather by mechanical penetration of the tissue? Thanks for the presentation

Presentation Everaert Gert

How is it possible to aggregate all these studies that might have been conducted with different methodology? Is there any mathematical tricks to integrate such issues? (Alexandre Dehaut)

Name: Federica Guerrini

Question: Thank you for your interesting presentation! Is the risk parameter related also to the size of microplastics?

By the way, it is impressive how the Mediterranean is going to become a serious hotspot. Where is the highest risk peak located?

Name: Niels Mast Thanks for that! Question: Taking a 1 micrometer particle translates into billion nanoparticles: "How are nano-size particles incorporated in your risk concentrations?"

Delphine Lobelle: Thanks for a great presentation! How is the risk level defined as 8 particles per litre?

Stephan Rohrbach:

Great talk! It seems that seasites near enveloping countries are underrepresentated. is it due to the waste quantities or because of oceanic currents

Montserrat Compa: Very nice presentation! In the map for the Mediterranean, it seems like the Eastern Med was at higher risk, any indications why?

### Questions and comments for the session 23.8\_O

No questions for session 23.8\_O

# Questions and comments for the session 23.8\_O

Name:...Rachel Sarner Question 1:...Nice job! Thank you for your poster. Did you say 2018 was significantly different? Why was that? Any ideas?

Name:...Javier Hernández Question 2:...Nice work!! I think I missed the information. Do you have results concerning Pyr-GC-MS/MS analysis? Helen Polanco: Hi Javier, you can find the results of our 2019 analysis in this report https://hudsonriverpark.org/the-park/parks-river-project/current-research/microplastics/ where we talk about the exact findings with our collaboration with NOAA GC-MS analysis. Thank you!!!

Rachel Sarner

Any theories on how MPs are arriving to the island if the concentrations do not match the population/urbanization?

Yes, the Gulf current that beggin in México, then it up to Main and Canada and then cross the Atlántic throw to Europe. One branch of Thais current goest to the North of Europe and the o the to the south. This branch is called Canarian current. This could be the massive input of microplastic to isolated islands like La Graciosa or El Hierro

### Questions and comments for the session 23.8\_Me

IMPORTANT: Please include your name at the beginning of your question or scientifically-relevant comments.

You can follow the live-stream discussion between the session speakers and the chairperson through the dedicated channel on the conference home page during the specified hours in the programme. The chairperson will draw upon your questions to the speakers during the LIVE discussion. IMPORTANT: Any questions or comments without the commenter's name will be removed, kindly.

## Questions and comments for the session 23.8\_Me

Name: Johanna Sonnenberg

Question 1: What software are you using for automatic counting of particles?

--> CellSens software (commercial) or ImageJ but I've less experience with it. From my knowledge it is well mastered by some colleagues of the community. And there are some tutorials on the web too.

Thank you!

Name: Alexandre Dehaut Question 2:. To Denise Hart, will you develop this approach on other field related to MP/NP analysis? Is it part of the "Gymnasium" education ? Nice initiative!

Name: Vesna Teofilovic

Question 3:.How complicated it would be to use this method for night of scientists, or similar science fair? Please provide written answer, I have problem with connection.

Dear Vesna, the answer of Denise was that it is not complicated, it is simple and possible to implement

it. Dear Vesna, you'll need a seperate dark room in order to see the fluorescence of the polymers when irradiated with blue light through the dark reader. But it is very easy to conduct the experiment if you have these components.

Name:.to Denise .. Question 4:...hei di go couqjantificate fluorescence?

Name:...Christine Knauss

Question 5:...How long have you been offering this experience and have you had anyone decide to continue their education in a graduate degree because of this experience?

Name:...Alexandre Dehaut

Question 6:... What is the average volume analysed with such medium? Did you face contamination by the paddle? How samples where transfered once collected? What is the periodicity of the collection?

### Questions and comments for the session 23.8\_Ma

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No questions...

# Questions and comments for the session 23.8\_Ma

Questions for Matthias Völkl can be sent to matthias.voelkl@uni-bayreuth.de (comment by Melanie Pöhlmann /SFB1357)

Your Name: Charlene Lujan Question FOR: Matthias Volkl Could you clarify me if macrophages can ingest particles from 0.2 to 6 um that you used in your experiment?

Question from: Maja Grünzner

Question FOR: Matthias Volkl

Sascha Müller : Nice poster! You mention on the top of your poster the different factors affecting toxicological affects. Specifically shape- did you/ or are you considering to study effects of shape, charge , hydrophobicity etc. on the cell uptake?

Answer Matthias: Yes, we want to e.g. mill particles to get sharp edges or use weathered particles from other parts of the CRC

Sascha Müller: Are you considering different surface charge properties aswell? This may be relevant for adsorption behavior within the cells?

Yes definitfly. We are working here closely with the collegues in dresden to analysis surface charge and distribution.

Sascha Müller: I am just wondering weather there is a charge change once the particle migrated into the "inside" of a cell, and thus changes its behavior?

We did not yet measure inside the cells yet. But it is very likely that there might be changes. Either way, the uptake might already depend on the charge, so the charge is a very important propertie as well.

Your Name: Zhiyue Niu Question FOR:Matthias Volkl

Have you looked in to effect of mixing-sized microplastic?

Answer Matthias: Not yet, but we plan doing this as well. probably the uptake rate will depend on this

Your Name: Simona Mondellini Question FOR: Beatrice Your Name: Question FOR: Azora

Garth Covernton: Thanks Azora! Are you going to also use natural particle controls to account for difference in physical vs. chemical toxicity?

Also, have you thought about something like a Dynamic Energy Budget model to account for changes in energy consumption that might have community and food web level effects? Yes, we still think about which natural particle as control to use, silica for example. Do you have suggestions?

I haven't considered the Dynamic Energy Budget model, but it sounds very interesting to do as well! Thank you very much! Thank you! Name: Charlene Lujan For: Azora Do you know how immune responses will be evaluated in your studies? For example oxidative stress response with CAT, GST,.. and thyroid hormone levels will be interesting too.

Thank you: I just wounder what kind of biodegradable plastics are you planning to examine in your experiments?

Your Name: Zhiyue Niu Question FOR: Azora Thanks for your presentation!! i wonder how you will assess/confirm the bio degradation of these polymers then? We plan to analyse tissue and faeces samples taken after the exposure for the polymers. Thanks for the answer.

Thank you Beatrice for your talk! How were the microplastic particles prepared? Thank you ! PET-MPs were obtained by consecutive freezing (liquid nitrogen) and grinding cycles. At the end of the process, PET-MPs were passed throught a 1 mm sieve in order to obtain particles in the MPs range.

Your Name: Stephan Rohrbach

Question FOR: Beatrice

Interesting talk, thank you! Have you an explanation why you detect oxidative stress in the gills, but not in the digestive gland? (manila clam)

Thank you! I can hypotesize that gills are more sensitive and quicker to respond than digestive gland.

Your Name: Kyle Kim

Question FOR: De Felice Beatrice

Is there potential differences between the 2 species in terms of uptake and rejection of mps – virgin vs. aged particles with biofilm?

Thank you! I don't know about difference between virgin and aged MPs, this coul be something to take into account for my next experiment.

Just wondering if you tried to look at any differences between male and female of the animals studied?

No, sorry I did not take into account the difference between male and female during this study, but this could be a good suggestion for further studies, thanks!!!!

# Questions and comments for the session 23.9\_O+Me+Ma

Here Francesco, I lost the information regarding the instrument set up for metabolomics, did you use orbitrap nmr?

ok thank you :) Hello Francesco, yes it's a NHMR methods to detect metabolomics networks potentially impact

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Nina Paul: Thanks Arno for the nice poster - Did you measure oxidative stress in all tissues wrapped together? Thanks Only on digestive gland. Thank's Nina.

Thank you Arno

Danae's presentation:

Thanks, great poster. Do you have any insight why some of the biomarkers (activities) are negatively correlated with the MPs content in the fish GITs? (Ana Catarino) Thanks!

\_\_\_\_\_

Stephan Rohrbach to Inger: Could you please specify which types of natural particles are you planning to use as a control?

Ana Catarino to Inger: Hi, how important do you consider that it is to test the leachates? Should this always be considered in MP studies? A control for particle effect vs chemical effect? - Thanks!!!! Well done!

\_\_\_\_\_

Name: Ghezali Yousra

Question: thank you for this informative insights! but at which depth did you sample the sediments for microplastic analysis?

Day 2/5 Pads

# **Questions and comments for the session 24.1\_O**

### TALK 1: QUESTIONS FOR VERONICA

Name:...Hana Fajković

Question 1 Good afternoon and thank you very much for your talk. Have you measured if the particle size has an influence on the absorption and what was the size of your MP

Name: Stefan Dittmar

Question 2: Thank you for this excellent talk! One question regarding the particle size - did you also test smaller MP sizes? If the sorption is assumed as "pure adsorption" this should increase surface and thus adsorption capacity, right?

Oh, kind of doubled the first question... :) maybe they can be answered together!

Name: Cleo: Question 3: Dear Veronica, thanks for this nice talk. Did you test statistically on interactive effects of temperature and pH?

#### TALK 2: QUESTIONS FOR MAGDALENA

Name: Kryss Waldschläger (RWTH Aachen University, waldschlaeger@iww.rwth-aachen.de) Question 1: Thank you for the interesting talk! Why did you decide to focus on disks instead of other particle shapes?

Name:...Yasmin Yonan

Question 2:...Hi! I was wondering what dimensions your settling column was, and how did you make sure you didn't experience wall effects? Thank you!

Name:...Antje Wichels

Question 3:...are there already results known for transition zones e.g from a river to the marine system with more turbulence?

Winnie Courtene-Jones (university of Plymouth, winnie.courtene-jones@plymouth.ac.uk) Thank you for this presentation. very clear and important factors to account for in settling studies. Q: Did you repeat this study in non-stratified water? Did the orientation of the disk change in nonstratified water also?

- That's great, thank you for your explanation of this! very interesting and all the best with the rest of your studies.

Magdalena: Thank you a lot.

Matthias Egger (The Ocean Cleanup; matthias.egger@theoceancleanup.com): Great talk and very interesting work! I was wondering whether you also looked at settling of MPs with various degree of biofouling? If biofouling is not homogenously distributed over the plastic surface, would this impact the particle orientation during settling and thus result in a different settling velocity pattern? Magdalena: Than you for the question. I did some tests. I'll answer via e-mail. Great, thanks Magdalena! :) I would also be very interested in this answer! If possible, could you also forward it to me? d.m.a.lobelle@uu.nl And to me as well! Thank you! waldschlaeger@iww.rwth-aachen.de

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Magdalena: Yes, of course. Thank you for your interest. :)

Delphine Lobelle: Excellent talk! I was wondering if you managed to look at smaller particle sizes at any point?

Magdalena: Thank you for the question. I haven't studied smaller sizes yet, but it's duable. Ok, thank you Magdalena and looking forward to seeing what you work on next!

Marie Poulain - Zarcos : Thank you very much for the talk ! I was wondering about what happens in turbulence, because I performed some experiments in two layer fluid with turbulence and I haven't observed any accumulation in the pycnocline for buoyant particles. Do you have any literature about it ?

Magdalena: I can give you some suggestions via e-mai. Thanks for this question.

Marie : you an email me at marie.poulain@imft.fr I could also share some work of my PhD if you want to discuss about it

M: Thank you. I'd be happy to contact with you.

Marie : thank you again for you talk !

M: Thank everyone for the chat. Good luck with your studies. You can contact me via m.mrokowska@igf.edu.pl

### TALK 3: QUESTIONS FOR ROCIO

Name:...Ceri Lewis

Question 1:...Great talk, just wondering which part of the Arctic you sampled from and what time of year - were the copepods lipid rich at the start of the exposure and do you think you might have different results if they were lipid poor at the start?

Name:...amanda dawson: did you use a surfactant or solvent to suspend the particles? do you think this had any effect on your results? did you have a surfactant control? Question 2:..

Name:.Gissell Lacerot

Question Hi Rocío. Thank you for your talk!. What do you think it's the effect of food availability with MPs for the response to occur? You mention food availability was important in the response you saw.

Thank you for your answer!

### Name: Pennie Lindeque

Question: Hi Rocio, thank you for your interesting talk. Your concentration of microplastics for the "high dose" was particularly high. Do you think that this sort of concentration could impact on the individuals ability to swim and feed by the plastics adhering to the copepods appendages (and possibly impacting on their ability to create a feeding current)? Thank you

Name: Špela Korez

Question: Hi Rocio and thank you for your nice talk. Short question. You said you were collecting the fecal pellets. Were they all "intact", in one piece?

Matthias Egger: Excellent work, thanks for your talk! Did you measure the density of the fecal pellets, by any chance?

Barbara Scholz-Böttcher: Great talk! Do you thing that weathering might have an additional influence due to enhanced functionalization?

Rocio Rodriguez Torres reply: I think weathering could have a different or additional influence on this copepods. Different microplastic characteristics, different shapes, size, weathering or pollutants... would be very interesting to evaluate. It could start by affecting the ingestion rates. There is an article (Rodrigo et. al., 2020) with the same species but using another stressor that is the oil and the copepod respond different. Unfortunately, in this case, we have not done anything with weathering particles.

Thank you for the question!

### Questions and comments for the session 24.1\_Me

E-Mail Addresses of speakers:

christian.laforsch@uni-bayreuth.de; denise.mitrano@usys.ethz.ch; Anja.Ramsperger@uni-bayreuth.de; Moritz.Lehmann@uni-bayreuth.de

Name:.Barbara Scholz-Böttcher, University of Oldenburg

Question 1:.Great talk Denise, and great approach, thank you! Q: Do you see any difference in behavior between PS and PET at this small size (regarding your doped particles )? How is the ratio of MNP to metal (regarding molar and weight equivalents)- do you expect any effect on density/sinking behavior of these "heavy" metals?

Thanks for the question, Barbara! I'll answer this question again here in the chat incase there are others who missed my live answer! We don't expect there to be (too many) physiochemical differences between our metal-doped plastics and others based on the metal incorporation, since the amount of metal is quite low (approx 0.3 wt%) and the metal is inside the plastic (i.e. not on the surface). For behavioral differences between the plastics, this depended on the situation we were investigating. In the context of the WWTP, we saw the same retention of plastics in the sludge regardless of particle size. However, for transport through porous media, nano plastics were more highly mobile than microplastic fibers, likely due to size limited transport. For future studies, we will investigate these question of size/shape/polymer differences on transport in more detail!

### Name: Kennedy Bucci, University of Toronto

Question 2: Really great talk, thank you! I may have missed this, so my apologies if you already mentioned it. How did you introduce the nanoplastics and fibers in the initial settling experiments? Were they introduced in the liquid?

Hi Kennedy - now that I read your question again, maybe I can provide a more exact answer to your question than I was able to live! We had it a little easier to introduce the particles into our system, since we were not only looking at settling behavior. We first mixed the particles in the activated sludge (to assess heteroaggregation between the plastics and sludge flocs) and then just allowed settling. Therefore, we did not need to be "careful" in putting the plastics into the experiment since they would anyways be mixed first. To allow settling, then we just stopped the stirring. The situation would be different if we wanted to look at "only" settling behavior, where putting get plastic into the column would then need more care to not influence settling dynamics. Thanks for taking the time to re-answer my question! Once again, it was a really great talk.

### Name:...Ana Catarino, Flanders Marine Institute Belgium

Question 3:...Great work, Denise! If using these doped particles in biota (fate of particles in biota), what is the minimum dose we need to use to have enough material for detection? Would the detection limit be higher than environmental relevant concentrations? Many thanks!!! We have performed a number of different studies using these particles in biological systems, but it's hard to give a hard and fast rule on dosing. Ultimately, the detection limit of the ICP-MS is the relevant factor, and there we can measure very low (ppt) concentrations of metals. However, we need to also consider if we need to digest the sample (which ends up diluting the metal) and other experimental factors like sample size or weight. While collectively this may mean that some exposures could be higher than environmental concentrations, they are normally still lower exposure concentrations than other studies I have read.

Furthermore, we get a lot of benefit by being able to track the particles through the system: in this way we can make a mass balance of plastic across the system and "prove" that particles are injected/internalized/etc. instead of just showing effects or impacts to the organism. I'm happy to share our experiences more if you want to get in touch!

Name:...Víctor Hernando Morales, University of Vigo, Spain

Question 4:...Congratulations, fantastic talk. My apologies if I've might missed this in your talk, but have your synthetic metal-labelled materials different densities? What is the density compared with most common plastics? and how do you relate the experimental results you obtain in your model reactors and real reactors with what happens in nature with micro- and nanoplastics with less metals concentrations and therefore higher or neutral buoyancy?

The particle densities do differ depending not he polymers we use. Now we have 4 plastics variants: PET, PS, PLA and polyacrylonitrile. Hopefully more will be on the way in the future to have a more complete matrix of particle chemistry, size and shape! :)

Since the metal concentration is quite low (approx. 0.3 wt%), we do not anticipate that the density is largely affected to change the settling or aggregation dynamics.

Thank you Denise, Congratulations again, it is a very inspiring project! Success!

Name: Martin Mittelbach, Graz., Austria

Question 5:Dear Denise, Thanks for the great talk. Does't the metal influence the plastic properties like density?

Hey Martin - your question is a common one as you can see above (also answers above) - so I will definitely have to more clearly state this in the talk next time! :) Thanks for tuning in!

Name:Niels Mast Qustion 6: Thanks, inspiring! What MP size-ranges were used in the tests? We have a few different particles types and sizes: nano plastic particles (polystyrene and polyacrylonitrile): 150 - 250 nm, polyester and PLA fibers (endless filaments 30 um in diameter, can be cut to any length but we normally choose around 500 um since this s a common length which sheds from textiles), and PET and PLA fragments (various sizes between 50 - 250 um).

Name: Stefan Peiffer uni Bayreuth

Question 7: Have you looked into fractionation of the NP in different particles sizes of the lachate from the sludge?

We did! It seemed that the nano plastics were more mobile than the larger one. Here we only investigated nano plastic spheres and microplastic fibers, but we might expect a similar result for the microplastic fragments (i.e. size limited transport through porous media).

### Name: Holger Kress

Question 7: Thank you for this interesting presentation! How much palladium did the particles contain and do you know whether the palladium has an influence on the sedimentation speed of the particles?

Hey Holger - your question is a common one as you can see above (also answers above) - so I will definitely have to more clearly state this in the talk next time! :) Thanks for tuning in!

### Question 7

Name:Steve Allen, Awesome work and a great presentation Moritz. So the oblique angle transfers the energy to a smaller part of the spray. Is that why it goes higher? Great stuff thanks At least the momentum pf the impact is focussed into one side of the crown after impact, so one side has a larger vertical velocity component and ejection height is larger.

### Question 8:

### Name:Juliana Ivar do Sul

Question 8: Inspiring work! what was the motivation to do it? In terms of the environmental consequences of it.

Hello Juliana! Moritz here. Main motivation is to understand how exactly microplastics get from the ocean into the air and then spread all around the globe. We want to identify and quantify processes that may explain the detection of marine microplastic particles in the air at the coastline. The processes in detail are very complicated and difficult to access in experiments, so we look at these systems with computer simulations.

Excellent! do you think marine sources of fibers and fragments that are prone to be transported by air are significant then? That is something I did not think about before. Seems scaring.

I haven't yet done estimations on the global amount. There are probably large local variations in how much litter is on the ocean surface and also temporal variations when wheather allows for the transition across the water-air interface and sufficient wind to pick up the droplets. So far we have only shown that there is transport from water to air and that raindrop impacts are one possible mechanism. But it is known already that sea salt aerosol, which is also produced by spray during wave action, bubble bursting and raindrop impacts, is in amounts large enough to affect concrete construction along the coastlines.

### Name: Julia Möller:

Question 1 for Moritz: Great talk, thank you very much! What Microplastic particles did you use and do you think the polymer type and particle shape will affect the particles behaviour? Our particles have 108µm diameter. The only particle property that we have control of is density. We tested the entire possible density range for plastics - 920-1270kg/m<sup>3</sup> - but during the short 10ms time after impact, bouyancy effects are negligible and the particles are just passively advected with the water. As long as the particles are small enough, their shape does not have any impact on their advection during impact.

Name: Holger Kress

Q: Thank you for this interesting presentation! Is it possible to take different physico-chemical particle properties into account to model for example particles that prefer to be in the bulk water or particles that prefer to be at the water-air interface?

We cannot directly simulate particle surface properties, but we could place the particles at the interface in the beginning of the simulations. However we know already that there is a certain volumetric region around the impact site - including parts of the surface - in which particles are going to be ejected.

Sascha Müller: nice talk Moritz! how would surface roughness of the water surface impact your simulation? Now you simulate a flat surface only, right?

Hi Sascha! Moritz here. In our simulations we use a flat water surface, although we did wome tests with a rough water surface to trigger different (random) crown breakup. Surface roughness doesn't have large impact on the dynamics overall as long as the scale of the bumps is small. With large-scale roughness, there will be some differences, but this also adds more complexity and less reproducibility to the model.

Presentation of Anja Ramsperger

Name: Nevena H

Q: Did you use other types of polymers too which are more prevalent in the food-chain thus would simulate a particle of specific concern for human ingestion?

Name Alexandre Dehaut

Q: How pristine Particles can be tranlocated inside cells? Diffusion throughout the membrane?

### Name Barbro Melgert

Q: Did you check the corona of the pristine particles after being incubated in the cell culture? What proteins absorb to the particles? Any differences between the weathered and pristine particles? All particles were incubated in the media together with the cells and therefore it seems (at least in comparison to the ecocorona particles) seem to play a minor role. But you're right, there will deifinetly being additionally coated with proteins from the surrounding media.

that is only when you assume the different surfaces absorb the same proteins.... thanks! Very interesting study!

Name Christoph Rummel

Q: Could you test if different polymer types display different affinity to eco-corona biomolecules? Thats a very good point. I assume that particle with different pristine properties (eg surface charge or hydrophocity) will show different affinities to biomolecules for instance, This is definitely an important factor to investigate. We are currently working on that usinfg different polymer types with dufferent initi<l surface properties.

Name: Susanne Belz

Q: Maybe I missed that info... What was the size of thean particles?

--> 3 µm

and the material/polymer?

---> PS (if I remember correctly) yes ;)

Many thanks!
# Questions and comments for the session 24.1\_Ma

Mikaël Kedzierski's presentation

mikael.kedzierski@univ-ubs.fr (https://www.sciencedirect.com/science/article/pii/S2214289419306738?via%3Dihub)

#### Name:...Alexandre Dehaut

Question 1:...Thanks for your presentation Mikael. Did you have time to check additives? Specific ones (eg gamma-HBCD) or others more broadly used.

Which advise when unpacking meat?

It would be interesting to have a partner with these skills to be able to go further ;)

We do not have any specific recommendations. Especially since the contamination probably occurs during the manufacturing process of the product. The consumer can therefore only notice the contamination.

#### Name: Bettie Cormier

Thanks for your presentation Mikael, I have a question regarding M&M, how did you take out the meat? Isn't it possible that when you took out the meat from the packaging, you added some MPs? If not, then it is a lot a MPs that we are able to eat by eating meat...!!! Thanks!

### Name: Natalie Hernandez

Thank you for the presentation! Will you test whether rinsing of the meat removes any microplastics? In other words, if it was recommended that consumers rinse the meat thoroughly, would this remove most of the contamination? Or only decrease it slightly? We are not even sure that rinsing the meat will reduce the amount of microplastic. It was a real surprise during the rinsing extraction phase to have so much difficulty in extracting the microplastics.

Amanda Dawson: Awesome talk!. Microplastics are sensitive to high temperatures, do you think the microplastics will survive the cooking process?

We propose an hypothesis that the microplastics are in facts cooked. In this case, and as a function of the cooking process, some molecules will probably be released. We propose some overwiew on what could happen and possible consequences for human health. But its mainly a hypothesis.

Chandrasekaran Natarajan's presentation

Name:... Question 2:... Name: Claudia Drago Question: Thank you for your presentation! Did you find some nanoplastic in the eggs? They were translocated from the adults to the eggs?

Irene Brandts' presentation

Name Bettie Cormier

Question 3 Nice presentation, thanks a lot. If the survival of zebrafish is not a good endpoint to assess the toxicity of NPs on the immune challenge, which endpoint would you like to follow?

Is the paper published? Can you give us the DOI please :) Thanks :)

Hi Bettie! Thank you for your interest, =).

Yes, it was published in Environmental Science Nano, here is the doi: DOI: 10.1039/d0en00553c Thanks a lot! Your presentation was really interesting, the combination of the use of ZFL and zebrafish larvae! Thanks, if I have more questions, I will get back to you :)

I will be very happy to answer any questions you might have! irene.brandts@e-campus.uab.cat

Michael Strum's presentation

Name:Cleo

Question 1: What are the potential environmental risks of the silanes ? And how do you further treat the removes agglomerates/particles? How are silanes produced? Name:...anita tirkey Question 2:.can this organosilane be us Name:...Éva Vizsolyi Question 3:. When you used PE-X, your removal was good, except in case of PE. What will happen with this particles when you use this technology for wastewater treatment, since PE is one of the most common polymer?

# Questions and comments for the session 24.2\_O

Name:...for Lisa Rocher

Question 1:...You say that the fibres you found were almost exclusively polypropylene. Did you not find fibres such as polyester and nylon in your samples?

MY EMAIL: lisa.roscher@awi.de

Robby Rynek: Thanks for the nice presentation. Were the small particles also sampled from the water surface?

Aline Carvalho: Thank for your clear presentation. I maybe have missed it, but did you perform a contamination control?

Catharina Pieper: do you have any idea where the foamed plastics could come from? thank you!

Marie Babinot : Thank you very much! Why put the low size limit at 2mm and not 1mm like many other studies? Do you also take samples of microplastics buried in the sediment?

## Alexandra:

Hi, thanks for your question! We only sampled in spring and winter last year, and observed great differences with maximum ccumulation in spring. We will continue this monitoring more frequently ti get the idea of the variability. Thank you for your answer and good luck with further research!

Catharina Pieper: question for Daniela - do you have any industries that could justify pellet presence on the island? and do you find pellets with different degradation states?

Cristiane Vidal: To Daniela: Wonderful presentation! Will you characterize type of polymers of pellets? Thanks!

do you plan to show your results to the industry that might be leaking the pellets?

Marie Babinot : Do you practice separation by density difference ?

Cristiane Vidal: Daniela: Could you please share your e-mail?

My Email: gadens.d@gmail.com

# Questions and comments for the session 24.2\_Me

E-mail address of speakers:

seema.agarwal@uni-bayreuth.de; sven.frei@uni-bayreuth.de; anja1.holzinger@uni-bayreuth.de

Name:Liliya Khatmullina,Shirshov Institute of Oceanology, Russian Academy of Sciences Question 1:Now we have plastic waste which is roughly speaking Carbon that is stuck in the environment. If this waste become biodegradable we will have a lot of easily degradable organic material coming into nature. Is this fact and its possible effects on the ecosystems also considered in your studies? And let me add to this: And these polymes (biodegradable polymers) become a huge source for carbon dioxide that is released into the athmosphere! (Sven Frei)

Name: Eleni Christoforou, University of Glasgow

Question 2: What is the different between biodegradable and compostable packaging and bags? Can both go in the compost bin then?

Question: Are there plastics that can be made from organic materials (e.g. shrimp shells) and when they break-down they can be used as fertilizer to the soil?

Name: Teresa Menzel, Uni Bayreuth

Question 3: Isn't the degradation of PET via enzymatic PETase also a process of biodegradation? Why is the biodegradability of PET discussed so controversially?

Enzymatic degradation not always biodegradation. If there is a proof of conversion to carbondioxide then only can be called biodegradable.

That makes sense, thank you!

Name: Jakob Cyvin, University NTNU, Trondheim

Question: Thanks for an interesting talk! I'm wondering what you think about the use of biodegradable polymers when it comes to plastics entering the arctic oceans (e.g. fishing equipment) - the conditions of cold deep waters - is it the right use of resources to implement it for these purposes, or should we use the resources on other applications or e.g. better infrastructure for delivering old fishing equipment: Cost vs. benefit? Thanks alot!

Name: Nisha Singh, IISER Kolkata, India

Question: Thank you for your talk! Are the additives used in the biodegradable plastics similar to normal plastics and do they leach as plastic gets degraded?

## Name: Zhiyue Niu, Flanders Marine Institute

Q: Very interesting talk!! For biodegradable polymers, they will be degraded both by organism and abiotic stressors. Do you think how important the biodegredation compared to abiotic degradation. Does it mean that biodegradable polymers have higher potency of forming microplastic than non-biodegradable polymers?

This can not be generalised like this. Like I gave the example of leaves, we hope that Thanks!

Name: Alicia Mateos Cárdenas

Question for Seema: Hi Seema, thanks a lot for your keynote. Did I get it correctly that bioplastics can degrade better in soil than in water (but there are not many degradability studies under real conditions)? is that for all of them?

Based on whatever is known, one can roughly conclude it.

## Sarah Stevens, NTNU Trondheim

Question: Thanks for the very intersting talk! When biodegradable and non-biodegradable plastics enter the same waste stream, can they be (easily) separated for recycling purpose? Very relevant question. At present this is one of the major problems. Therefore, instead of making water bottles or cola bottles biodegradable, it makes sense to be going for more specific applications. In agricultural fields or when they go to compost plants together with bio waste surely will bring some positive effects.

Thanks!

## Name: Mohammad Wazne:

Question to Fei: Did you compare your results in this work or previous work with sedimentation zones where there is a water stagnant state? And thank you for this work it is great!! We published this work in 2019 and compared our results of MP abundances with published values. By now there are much more studies avaiable. We plan to do such a in detail comparison in future when we have our results from the flume experiments.

Q about details of lab experiment - how do you deal with the water (with these micrometere scale particles) from the flume after the experiment. Does it go simply to the wastewater or you accomplish some kind of filtration to get rid of the plastic particles?

It is a recirculating flume so it is a closet circut. If we have to change the water we filter the particles. I see, cool, thank you for the answer! )

PRUVOST Jean, Lehna-IPE, Lyon to Sven Frei

Question : How did you manage to obtained the video you showed us in the end of your presentation ? Is there a risk that your staining protocoloe mostly result in water staining in stead of MPs ? By the way, thank you for the super talk

The fluorescence coating is only on the particles we do not produce the particles ourselves they are commerically avaiable. The water does not contain the dye. We detect the particles with a fluorescence camera.

Thanks

Name:Camila Vidal, University of the República, Uruguay

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Question to Anja: did you try different colors of microplastics, of did you find a preference from the animals?? or was this not evaluated? Thank you!

Name: Kennedy, University of Toronto Question: What dose did you use? Thank you!

Name : Claudia Drago

Question: Thank you for your nice presentation!! 📥 Did you test for feeding rate? It was altered? Do you think the inflammation response are due to a decrease in feeding rate or the microplastics per se? Thank you Claudia, for the fibers we did not test feeding rate. For the fragments we did not find an effect. I think it might be due to the shape. Fibers are known to cause gut damage in other organisms.

Kristina Klein:

Thanks Anja for the nice presentation. Maybe I've missed it: what was your explanation again for the GST activities for fragments, but not for fibers? As the fibers were bigger in size/volume than the fragments we thought that they might be beneficial for binding toxins that are in the soil. Therefore the worms do not produce as much toxic metabolites themselves and do not need as much GST.

# Questions and comments for the session 24.2\_Ma

Questions for CAROLINA ROCHA: Name: Alexandre Dehaut Question 1: Thanks for your presentation You mentionned the use of Fume Hood, was the air flow switched on or off? May be I missed it, bhos did you take controls into account? --> Thanks

Name:...Andy Booth Question 2:...Could some of the aquaculture infrastructure (e.g. ropes) be contributing to the high load of fibres that you observed?

Questions for MARTA SILVA:

Name:...Matt Cole

Question 1:...Really interesting results. Polystyrene can contain monomers and high levels of additives. Was there any chemical analysis? Do you think the effects stemmed from physical damage, chemical pathways, or food replacement, or other?

Thank you for your answer. If made in lab, you may want to consider monomer (styrene) leakage? Styrene v toxic.

Name:...Andy Booth

Question 2:...Can you comment on the relevance of the spherical test materials you used? For example, did they have any surfactants associated with them? You conclude with biochemical level effects, but what mechanisms can you provide to support these toxicological responses? Name: Andreia Rodrigues

Question: How do you explain the decreases in lipid peroxidation?

Name Miguel Tamayo

Question: Thanks!! :) Did you sudy the aggregation at the different concentrations? Did you find any correlation with the toxicity?

Great thenks! Name: Yuanhu Zhang Question: Thanks for the nice talk, how did you synthesize the PS NPs, and how do you purify them?

Questions for LAURA STUTZINGER:

Name:...Amanda Dawson: Super interesting talk. Do you know if nile red has any impact on the spectra obtained using ftir? What is your typical match %? when comparing to reference standards? no worries if i don't make any sense

Question 1:...

Name:Elena Hengstmann

Question 2: Were environmental samples purified before staining with Nile Red?

Name: Holly Nel - Are you counting everything that has a pixel brightness/fluorescence? Or do you have a threshold/cut-off? Thank you, I think I was misunderstood. I meant do you exclude particles with a lower fluorescent intensity?

Name Alexandre Dehaut

Then, which chemical to be used for orientation test (discriminating synthetic vs. non synthetic particles)?

Peter Vermeiren

Thank you for your talk. Do you have an idea of which organic materials are most similar in their fluorescence to polymers? Ok, thanks, perhaps we can find ways to remove these kinds of materials.

Questions for ALESSIO GOMIERO:

Name: Alexandre Dehaut

Question 1: Dear Alessio, thanks for this presentation. Just a "Fashion" question, where did you find these MP suits weared by theperson on the picture? Is it sold by a supplier? Is it 100% cotton? Name:...Matt Cole

Question 2:...Hi Alessio, great talk. (1) Did you validate your methods using spiked tissues with known quantities of plastics? (2) What biological pathways would explain the presence of microplastics into muscle/flesh?

-- Thanks Alessio :)

Peter Vermeiren

Thanks for the presentation. What was the motivation for chosing the liver as a tissue? Did you have a hypothesis on accumulation in this tissue. Thanks p.vermeiren@science.ru.nl Florane Le Bihanic :

Thank you for the great presentation. What do you consider as the detection limit in term of MP size for you whole study?

Amanda Dawson: Thanks for the talk, did you also find microplastics in the GIT in the same fish where you found microplastics in the fillet?

Andy Booth: What mechanisms do you propose are resulting in the MP in the sizes you have identified (up to 241 µm) entering the tissues of the fish species studied? Covered already :) Carolin Philipp: Thank you for this very informative talk! Is this study already published? If yes, can you maybe send us the DOI? Ok, great! :)

Stjepan Budimir<> Is this the answer to old question "is there MP in fish fillet". no need to discuss is there, but how much?

# Questions and comments for the session 24.3\_O

If you have any further questions or data that you think should be considered in our river catchment scale models, please feel free to contact me: elke.brandes@thuenen.de

For the German listeners: There is a new paper from E. Brandes, S. Gholamreza, S. Cieplik, M. Henseler, et al. (2020): Modellbasierte Forschung zu Mikroplastik in der Umwelt - Synthesepapier. Link: https://bmbf-plastik.de/sites/default/files/2020-11/Modellbasierte%20Forschung%20zu%20Mikroplastik%20in%20der%20Umwelt\_Synthesepapier\_Brandes%20et%20al\_2020.pdf

Name: Erik van Sebille, Utrecht University

Question 1: Nice overview presentation of the project! How representative do you think that the models you are developing for Germany would be for catchments/basins in other parts of the world? What is similar, what is different?

Name:...Prado Domercq, Stockholm University

Question 1:...Will these models be publicly available and how do you plan to parameterize them in terms of MPs fate processes at the different levels/compartments? Thanks! great overview and clear talk!

Name:...Batdulam Battulga (Tokyo Metropolitan University) Question 1:...Will you consider the plastic aggregation in the models? Thanks for the interesting talk!

Questions for Reza:

Reza, do you have an email contact? I'd like to discuss more about the answer of your first question. Thanks, Nia Jones. Thank you very much, I'll send an email shortly. :)

Gholamreza.Shiravani@nlwkn-ny.niedersachsen.de

Reza, i missed this talk :( i really wanted to see but messed up what room i was in. is there any way i could view your recording (i will not circulate!) my email is winnie.courtene-jones@plymouth.ac.uk

Name: Maximilian Born (RWTH Aachen University) Question Thank you for your nice presentation! Do you think it's crutial to implement fragmentation data as well, since MP is transported differntly with changing size? The aggregation is more important for nano plastics, and it was not yet considered in our model.

Questions for Stephanie:

Name: Erik van Sebille (Utrecht University) Question for Stephanie: has the model and report been peer-reviewed? I mean independently verified/validated by independent experts OK, thank you

Name: Astrid Delorme (Sigma-Clermont)

\_MICRO 2020. Fate and Impact of Microplastics: Knowledge and Responsibilities. 23-27 November 2020\_\_\_\_

How do you differentiate between secondary and primary microplastics? ok thank you, Yes I agree it is not easy!

If you have further questions, please fell free to contact me: stephanie.cieplik@bkv-gmbh.de

The report is available on our website: www.bkv-gmbh.de/infothek/studien.html www.bkv-gmbh.de/en/info-zone/studies.html

Questions for Martin:

Link to the synthesis paper (in German language): https://bmbf-plastik.de/de/node/355

# Questions and comments for the session 24.3\_Me

E-mail address of speakers:

Hannes Laermanns: h.laermanns@uni-koeln.de; Nora.Meides@uni-bayreuth.de;

Martin.Loeder@uni-bayreuth.de; Gerasimos1.Gkoutselis@uni-bayreuth.de

Name: Stefan Dittmar, TU Berlin

Question 1: Thank you very much! Can you say something about the properties of the particles used, were they spherical? To me, it (slightly) looked as if particle size also had an effect on path length in your example video. What do you think? What's the minimum particle size, you can still detect?

Name:...Arianna Varrani, Institute of Geophysics, Polish Academy of Sciences Question 2:...Did you consider the effect of the pressure drop in the rainfall simulator as possible cause of your high asymmetrical path areas? Thanks for the interesting presentation!!! Pressure drop= pressure decrease in the pipe just before the spray.

Hannes: Thank you Arianna for that input. I will consider that!!!

Name: Andrea

Question 3:.. Are you interested in how the particles are moving in a three dimensional space or just over the surface of soils on slopes? I'm thinking about migration of particles down into the soil rather than across the surface

Name:Johanna Sonnenberg

Question 4: Have you observed the burying of particles in the sand? Ok, that also answered my question, thank you!

Hannes @ Andrea and Johanna: That would be interesting, indead. Could be tricky to achive subterrain particle tracking but I think a colleage works with some radiation scans... That could be possible.

Name:...Hana Fajković, University of Zagreb

Question 5: Thank you, great talk! Do you plan to use a similar approach to the water body?

Name:...Lucrecia, University of Hull

Question 2:...All the particles where from the same type of plastics ? What is the density of the plastic ? Details of the flow ?

Hannes: PMMA: The are quite light weight particles. The densitiy is not much more than 1 point something. So that is also something we have to consider: Are the particles pushed, elevated by a water film or how are they set in motion!?

## FOR NORA (nora.meides@uni-bayreuth.de)

Name:...Matthieu Mercier, Toulouse (IMFT, CNRS)

Question 6: Can you more details on the stirring (nature, intensity)? Also how concentrated were your plastic suspensions (very dilute or not)? Finally, did you track the creation of nanoplastic all along the fragmentation process? Great talk !

300 rpm with a PTFE magnetic stirring devide to avoid contamination and 20 g of PS particles within 600 ml DI water. The water level was constantly adjusted due to evaporation taking place, however temperatures were held at a constant level. Nanoparticles were observed within SEM images, however for particle size analysis (and due to agglomeration at very small sizes) it was challenging to quantify within particle size measurements.

## Name:...Cristiane Vidal (Unicamp, Brazil)

Question 7: What wavelengths did you use for irradiation? (Thanks!! Nice presentation!) The irradiance of 60 W/m2 is set to a wavelength between 300-400 nm, however the entire spectrum ranges from 290 - higher wavelengths, according to solar radiation. The area between 300-400 nm ist most important, as the energy of this radiation is sufficient to cleave C-H and C-C bonds.

Name:...Sanye Soroldoni - FURG (Brazil)

Question 8: How did you calculate the equivalence of the accelerated time to outdoor weathering? Thank you! Great talk!

We took the total irradiance within the chamber (~600 W/m2) and using also the total irradiance from Central Europe we were able to calculate the acceleration factor of ca. 5. So the acceleration is based on the irradiance.

## Name:...Christina Bogner (Cologne, Germany)

Question 9: Thank you for this very interesting talk! Do you have any idea of the size distribution of the daughter particles? Thank you :-)

Yes, daughter particles after 3200 h of simluated solar radiation and mechanical stirring (mechanical stress) range from nanoparticles up to  $<100 \mu$ m, however small particles start to agglomerate and the smaller the particles the more challenging size analysis is.

Name:... Andrea NTNU, Norway

Question 10 Thanks for an excellent presentation! Do you think the production of polar end groups is high enough to show changes in for example IR spectra?

Yes, but NMR is more accurate and allows us to differ between different functional groups I was thinking specifically about FTIR, as it is a common method for identifying environmental samples. It would be interesting to see if there are any characteristic peaks that appear to possibly "age" the plastic simultaneously with polymer type and particle #, shape, etc.

Thank you for this interesting input! We will check this up. Until now, we only did NMR measurements.

Really great work, thanks for sharing!

Thank you :-)

Name: Matthias Egger (The Ocean Cleanup, Netherlands)

Q11: Exciting work, thanks for sharing! Any chance you could provide an overall fragmentation rate? I am thinking of %mass loss per year (or similar). Thank you! We do not observe mass loss, only fragmentation :-)

Name: Susanne Belz Q: Sorry I had problems to join and thus came late. What type of particles did you use (size, shape, source)?

We used milled PS particles (125-200  $\mu$ m) made out of granules with mainly rounded shape but varying

Name: coco cheung (hong Kong) Q13: thanks for the talk. why did you choose PS as your target sample? And also how do

Name: Amedeo Boldrini, University of Siena

Question 14: Thank you Nora for this interesting work. During the experiment is possible that also temperature played a role in degradation?

This might be a point. During the experiment, temperature was set constant. We will check degradation at different temperatures as well. It might accelerate the degradation process (Chain scission, radical reactions etc.)

Name: Johanna Sonnenberg

Question: Do you published your results already?

Hey Johanna, yes, the paper including the data from my talk is submitted, but not published yet. Please keep an eye on our microplastics page for upcoming publications! :-)Thank you, I am looking foreward to read it. :)

I am also happy to answer any further questions! Please don't hestitate to contact me: nora.meides@uni-bayreuth.de

Martin Löder's presentation

Name: Alexandre Dehaut , Anses France (Boulogne sur Mer :-)) Question : Any idea to know how to transfer such nice techniques to field samples?

What is the hazard of BaSO4 for the manipulator?

Name:Barbro Melgert (groningen the netherlands) Question: have you ever tried bigger animals, like mice? Or isolated organs from mice (for instance lungs?)

Name:Gerasimos Gkoutselis Question:Could we use the fluorescence imaging for nanoplastics in hyaline fungal cells?

Name:Susanne Belz (JRC)

Question: What is the source of your particles? In-house produktion? Supplier? Which size(s) did you use? shape? What is the relevance to a realistic environmental setup?

Name:Flora Rendell-Bhatti, University of Stirling, Scotland

Jafar Alomari: how did you determined the pathogenecity of the detected Fungi

Jafar Alomari

Thanks for the interesting ppt, do you publish this study? If yes please provide the DOI, I am interested in detailed methodology

Question: Would sediment particles or food particles in the gut of animals interact or disrupt the image for quantifying microplastic particles?

Name: Alexandre Dehaut

Question : What is the limit of detection (or resolution? ) of such technique?

Questions to Gerasimos Gkoutselis Talk:

Name: Diego Lelis (Federal University of Rio de Janeiro)

Question:Did you have difficulty trying to view the microplastics in SEM? What voltage did you use to be able to visualize the microplastics without distorting the sample? This is for Makis , right yes? --> coated plastic fragments were visualized and imaged using a Quanta 200 electron microscope (FEI, Hillsboro, Oregon, USA) electron microscope with a 10 kV electron beam and varying magnifications

--> the SEM approach was really suitable, no real problems

Remark: If you want to improve the images of the SEM sample from biological samples you can apply an Alcian blue protocoll which helps to preserve biological structures, at least for bacterial samples, but it might be suitable for fungi as well.(Stephan Rohrbach)

Name:Ana Lacerda, FURG-Brazil

Question Very interesting talk, thanks for sharing! Do you think the occurence of potential human pathogens really implies in their pathogenicity? And are you/your research group seaking to analyze their "real" pathogenicity in humans? How it would be done?

--> a lot of fungi are only opportunistic pathogens of animals and humans, but assuming MP increase the probability of coming in contact with those potential pathogens or possibly increasing the carrying capacity of an ecosystem for those microbes, per definition, increases infection probability

Yeap, I totally ttagree!

I'm asking because I'm in the same situation.. I've found many pothential pathogens in environmental samples from the open ocean, but we cannot affirm they are really pathogenic!

Check it out our recent paper https://doi.org/10.1111/mec.15444 and I'll contact you by email, as I have more questions. Thank you so much for your presentation!

--> I will check it and would be very happy to hearing from you via mail. Thanks for the very inspiring questions.

Ana, if you are using DNA you can also look for pathogenicity genes with PICRUST. --> very good point.

In fact I have used the MGRast and found some human pathogens, but they are only "potential", right? This is a big issue actually. Anyway, I'll be presenting tomorrow morning.

Yes, potential. Another option is to use a more curated database, maybe. But of course, based in DNA will always be potential. Looking forward to see your presentation.

During the MICRO 2018 and also the review process of our fungi paper some Plastisphere experts raised up this point of talking about pathogens...

--> there are only few primary human pathogens existing. So in the case of fungal organisms, I would only talk about human pathogens, when encountering some of these.

Name: Tallita Tavares (Marine Sciences Institute, Brazil)

Question: Congratulations on the excellent talk and results. Considering the DNA extraction from MP samples, I wonder about the difficults with contaminants and concentration of samples. Thank you!

--> certainly, plastic particles (e.g. nanoplastic) could not be completely removed from the soil and vice versa. Persumably, considering this, the plastisphere is even more distinct from soil communities.

Name: Jafar Alomari

Question: how did you investigate the pathogenecity of detected fungi,

--> we classified only genera and species as potentially pathogenic, which reportedly can occur as human pathogens. No test for pathogenicity was performed.

Name: Jafar Alomari

Question: Thanks for the interesting ppt, do you publish this study? If yes please provide the DOI, I am interested in detailed methodology

I am also interested in reading the paper! Very interesting stuff :)

--> we're in the final steps, it will be submitted very soon. Please check the CRC SFB 1357 Website of the University of Bayreuth for the DOI in a few months for information. Or scholar it :-) will you be first author? (i.e. whos name should I keep an eye out for?)

--> yes, G. Gkoutselis :-)thanks!

good Luck

Thanks and all the best from Germany

Kennedy (she/her): Did you or your lab investigate implications for harm in wildlife? Do you have plans to?

Thank you!

--> yes, we have plans in this direction, but have not designed a study yet. This will follow as soon as possible.

# Questions and comments for the session 24.3\_Ma

Questions for Andrea Binelli

Name: Ksenia Groh

Question 1: Thank you for the very interesting presentation! I have a question about your findings of particles in the eye and neuromast. In how many fish have you observed this (statistics), and also, in how many neuromasts per fish?

Name:...Ana Lacerda, Question 2:...Very interesting talk, Andrea! How do you see the same kind of analysis being applied to humans?

Name: Julia Pawlak

Question to Andrea: Thanks for your interesting talk! Maybe i missed that point. Did you used environmental relevant concentrations of Triclosan?

Question for Eric Ben-David

Name: Konstantinos

Question:Nice work! How did you filter your samples through 0,45µm mesh? How did you define your fibers?

Name: Sam Athey Question: Great talk! What types of dyes/pigments did you find associated with fibers?

Name: Vianey Landeros Question: Great work! How often were you taken the samples? i.e every 3 months, or every month?

Name: Maria Kazour Question: How many filters were used for each sample? Since a 0.45 µm mesh size is really small. And was the whole volume of 24H composite sample analyzed? or just a sub-sample?

Question for Maaike Goudriaan Name: Ouestion:

Name: Stephan Rohrbach

Question: Thanks for your great talk. Have you synthesized the 13C PE by yourself and is there a chance that there are also residual isotopic labelled components within the PE?

Matthias Egger: Great work, thanks for sharing Maaike! Any idea why 13C-CO2 levels off after ~15 days?

Hey Matthias, well we have some speculations. Could be that we reach the stationary phase of the growth, although the start amount of biomass was very low. Oxygen should not be the problem since we add this and the dissolved oxygen concentration seems sufficient. Another option could be that the microbe starts CO2 fixation.

Stephan Rohrbach: Have you gained already some information which kinds of enzymes are responsible for the detectable degradation?

Ghezali Yousra: Thank you for your presentation! did you determine any specific genre of bacteria that specifically degrade the PE?

Hi Ghezali, thanks for your question! In this case it was a specific strain of R ruber that were isolated in a lab. So we know that it was this microbe specifically in this experiment.

Questions for Tania Alajo

Name: Alexandre Dehaut

Question : Why did you choose the PP trimer as a marker (instead of 2,4 dimethyl-1-heptene) (in the M&M)?

Did you noticed any change in Py profile before and after weathering for other polymers?

At 350°C don't you fear to have signals from additives of other molecules? Did you use specific markers ?

Name: Stephen Burrows Question: Great talk! Do you think temperature would be a significant factor in your PP degradation?

Name: Francesco Saliu

Question: did you already think about how to calibrate the system? If every particle in an evironmental sample display a different degree of weathering (and it should be like this in the "real world") and the peak used for quantification is affected by the degradation of the polymer (as you demonstrated), is still possible to provide quantitative data by pyrolisis-GC-MS?

# Questions and comments for the session 24.4\_O

Speaker: Klein Malin Microplastic particles in atmospheric deposition of Northern Germany - Klein Malin, Brecht Torben, Fischer Elke.

Name:...Eva C. Question 1:.Why do you think that smaller particles are more abundant in the samples ?..

You mention on the poster that laboratory analyses hasn't been complete due to Covid-restrictions, however, were you able to continue to sample during the "lockdown"? If so, do you expect an effect on the MP concentration in the atmospheric fallout?

Name:...Eike Esders Question 2:...Nice poster presentation. I am not sure if i missed it. Did you also did some footprint analysis for potential sources?

Speaker: Oster Jakob Towards revealing sources of atmospheric microplastic pollution using an innovative wind-drift sampling device - Oster Jakob, Loeder Martin, Babel Wolfgang, Georgi Christoph, Laforsch Christian.

Name:.. Alex C.

Question 1:...Is there a maximum wind speed at which the device can work with or be exposed to? Thinking on that maybe sometimes stronger winds will bring debris from further away and if it's possible to study with this same device when there are strong winds. Thank you!

Name:...Eike Esders Question 2:...Nice poster and presentation! Any clues why more fragments than fibres were found?

Speaker: C. Prata Joana Sampling airborne microplastics and fibers: lessons learned from Aveiro, Portugal - C. Prata Joana, L. Castro Joana, P. Da Costa João, Duarte Carlos, Cerqueira Mário, Rocha-Santos Teresa.

Name:...konstantinos

Question 1:...for jakob,

Very nice work! and personally I am not surprised that you have found more fragments than fibers. Most of the quantification studies indicate the same .

Anyways, How did you take your blanks? did you find any contamination from your lab procedures? and Couldn't you try to go a bit lower in size with changing the settings of your instrument?

# Questions and comments for the session 24.4\_Me

Name:Tanja Kögel Question 1: Interesting findings! What was the size of the MPs the plants were exposed to?

Name:...Stephan Rohrbach Question 2:...Do you think other plants/ grasses will be affected in a similar way by Microplastic?

## Questions and comments for the session 24.4\_Ma

Name: Amedeo Boldrini, University of Siena

Question 1: Thank you Thomas for your interesting poster. Do you know what is the distance (not the exact one but maybe a range) at which P-MPP particles exhibithed actractive forces and if is possible the formation of aggregates?

Hello Amedeo, thank your for your question. P-MPP particles showed attractive forces at around 10 nm separation. But as they are highly charged they don't aggregate. So the suspension as we recieved it by the manufacturer is stabilized by charged. However the supension for M-MPP seems to be stabilized by steric repulsion so outreaching polymer chains.

fell free to reach out @schuerc on twitter or schuer@bio.uni-frankfurt.de

Name:...Alexandre Dehaut

Question 1:...Thank for your presentation. (I'm far from being an expert in this field) On the "road" from lab studies to field likelihood, where do you place your study (for the comperehension of the toxicity mechanisms)? (I hope it is clear)

Name:... Alice Horton Question 2:... Why are there no data for kaolin at 10000 concentration after generation 0? Or did all individuals die? None died, we just discontinued this treatment aT MAKES SENSET MAKES SENSE! oops :)

name: Charlene Trestrail Question: Thanks for the interesting talk, Christoph. Do daphnia eat the kaolin?

Name: Anita Jemec K

Question: Did you analyze kaolin for any other phys-chem properties than size. Could there be any other potentially hazardous chemicals present? Do you find it a suitable "natural " control. Would you have any suggestion for natural control in soil tox testing.

Name: Claudia Drago Thanks for your interesting talk!! Did you notice aggregation of particles when you expose them to wastewater?

Name: Alicia Mateos Cárdenas Q: Why did you choose Kaolin (high density so it sinks) to test toxicity for Daphnia? very nice talk as always :)

Name: Inger Lyngstad Question: Why do you think the wastewater treated MP gave lower toxicity?

Name: Xavier Cousin Question : regarding food this can be one limiting factor in the wild so eliminating this prevent evaluating the influence of food limitation in "toxicity expression" (and nice talk by the way ;) )

may be overlooked if not considered

Name: Lukas Kruckenfellner Question: Nice presentation, thank you! :) Did you notice the formation of winter eggs (Ephippium )? No, interestingly we had none! Thank you for your answer!:) Thanks for asking!

Merel Kooi 's presentation - merel.kooi@wur.nl Name: Reza Shiravani Question: Thanks for your great presentation. Elongation value of one could be referred to a cubic as well as to a sphere, how do you distinuish them?

Name:Gissell Lacerot Question: Very interesting presentation Merel!. Quick question. Which species are included in your analysis? Oh, great. Already answered. Thank you!

Name: Garth Covernton (University of Victoria, Canada)

Question: This is very interesting. I may not be fully understanding the power law aspect of this for particle size, but it seems deterministic. Why go this route rather than assuming stochastic distributions? For example, exponential or log-normal distributions with different parameter values for different environmental compartments, etc.

[Thank you for your answer! Super cool work. It would be really cool to build in a stochastic component in addition to the deterministic fragmentation part and then build that into various microplastics models. Your model is essentially a linear model so could use various GLMMs fit to real data to then make predictions. I'm a Bayesian stats nerd and this would be a great Bayesian modeling project!!!] Hi Garth, how about a meeting after MICRO, since I'm definately not a stats nerd, but am very interested in your suggestions! Can I contact you? Absolutely! gcov@uvic.ca is my email.

Name: Anja Rebelein, Thünen Institute

Question: Great talk!! Did you include microplastic fibers? Or are there limitations concerning analyzing fibers?

Name: Winnie Courtene-Jones

Thanks for your presentation. do you think that the shape of the polymer influences the size distributions you report, e.g. I noticed polyester had a very elongated distribution, polyester would often be in the form of fibres (I'd imagine), with the length reported which can be highly variable - I did not have time to notice if this was the same for other polymers.

- maybe a factor to analyse and look into this with your further study that you mentioned looking into this. could help tease apart some of the polymer specific distributions in different compartments that you mentioned. all the best with your future work! Thanks for this suggestion, I will definately look into it!

Name: Josef Brandt

Do you explore chemometric clustering methods to observe patterns or correlations in your datasets? So, PCA in the simplest form or other models...? Very interesting approach, for sure!

Questions and comments for the session 24.5\_Me

Name:...Nur Hazimah

Question 3:...Moving forward, are there any other criteria you would consider to improve toxicity studies?

Name:...Kristina Klein Question 4:...Is this (high) number of criteria realistic? I guess not every lab has all of the recommended methods/devices.

## Name:...Stephan Rohrbach

Question 5:...Do you think it is more likely to find MP in salt then in other groceries in general? Especially considering that one eats only a few grams of salt each day, but eats far more from other foods. So whats your opinion about the resulting risk? Thank you for your talk.

R:I think it will depend on how the product is extracted, processed, packaged and distributed. There are many variables, but the more steps you have and the lower the quality and cleanliness of the steps, the greater the risk of contamination. It is difficult to measure the risk. If we insist that all food may be contaminated by microplastics. The risk will be proportional to the amount of food we consume, especially if it is a food that may contain a large amount of microplastic.

# **Questions and comments for the session 24.5\_Ma**

Name:...Carolin Philipp Question 1:...Thank you very much Sonja, for this great talk. Do you have explanations for the fact, that the amount of microplastics is similiar in the Mediterranean and in the North Sea? Ok, thank you!

Name:...Judith Weis Question 2:...Is it possible that the technology in 2009 was not as good as this year and so overestimated the number of microplastics?

Name:.Cleo Question 1: Thanks for the nice overview. Ho would you calibrate the model? thank you

Name: Ana Liria Question: Are you measuring the impact of aquaculture on wild fauna? By entanglement for example. Thank you for your reply.

Name: Winnie Courtene-Jones did you find microplastics actually ingested by Velella? or where they just adhered to the surface? how were they sampled? - yes, very interesting!! thank you

Concerning Velella also, which microplastic shapes were found in this species? for example fibers, etc

Name:.Cleo

Question 3 : It is a pity, it seemed on my laptop that the slides are jumping too fast ahead, missing parts of presentation? Or maybe the video was cut like this? But it seemed very very intersting, thank you. My question is: you mentioned plastic sighting as method, did you also do net sampling or how did you assess the microplatics? thank you, I am sorry I missed this;

Name: Claudia Drago

Very exciting project! Which biomarker do you analyze in correlation with microplastic or macroplastic ingestion in cetacean? Maybe I missed it.. Thanks

## Questions and comments for the session 24.6\_O

FOR PRESENTERS: please insert below your email for any additional questions later on:

- Stefanie: s.l.ypma@uu.nl
- Mikael: m.l.a.kaandorp@uu.nl
- Victor/Charlotte: victor.onink@climate.unibe.ch
- Delphine: delphine.dobler@ird.fr

For Stefanie Name:...Steve Allen, Question 1:... Great work in a nice area :-) Just wondering if you had considered ejection from the sea into the air?

Georgie: are you taking into account land sources of plastic also in your model, as well as the marine sources such as fishing activity you mentioned? Thanks for the talk.

Astrid Delorme: Very nice presentation! how frequent were the sampling done on the beaches?

Name: Federica Guerrini

Question: Thank you for the interesting talk, great topic! I am curious about how are you going to implement machine learning. Thanks

hi Frederica, thanks for your question, We're still in the early stages of the machine learning, so any tips are very welcome. At the moment I'm mainly using multivariate linear regression to connect hydrodynamic fields to the beaching patterns given by the model simulations.

Matthias Egger: Very exciting work, thanks for sharing! I was wondering whether you also looked into the effect of windage? Do you see different beaching patterns dependent on windage?

### Name: Winnie Courtene-Jones

really interesting talk, thank you. interesting to see the seasonal differences. could you also use any colonising epibionts to also look at where plastics may have come from/how long they have been circulating in the ocean? (not sure if this is in scope, or if you are working with a biologists on this)

hi Winnie, this would definitely be possible and for sure we have some biologists involved, so I will keep this in mind!

Question: Hi Stephanie, nice talk. What coastal/beaching processes did you consider/neglect? Is the effect of onshore/offshore wind taken into account?

For Mikael

Name: Louise Schreyers...

Name: Isabel Jalón-Rojas

Question 1: Thanks Michael for this great presentation. Can you expand a bit on the calibration/observation data that you used? Looking at your question I might have misinterpreted it; if you mean the observational data, we used neuston net/manta trawl samples taken at the surface of the water to calibrate the model

Federica Guerrini

Very nice work! Does fragmentation also affect particle sinking time in the model? If so, how? Thank you!

When fragmentation breaks down the particles, I would expect some differences in the sinking time scales, as biofouling can have a different effect on different particle sizes. Delphine Lobelle will present a biofouling model in the 17.00-18.00 session in Ostrom's room.

Matthias Egger: Excellent work! Do you still get the 50/50 beaching/sinking distribution when including fragmentation? (sorry, haven't read your pre-print yet) Good question, it's definitely a good idea to look at this sensitivity in future studies!

Marie Poulain-Zarcos : thanks a lot for your talk !

For Victor/Charlotte Name:... Question 1:...

Astrid Delorme: Thank you for the interesting talk, did you only consider 'surface' beached plastics? Is it possible to consider buried plastics in your model?

Louise Schreyers: thank you very much Victor and Charlotte! How do you explain the overestimation that you noticed in Southern Africa between model and observations? Thanks for the reply btw

Federica Guerrini

Another very nice presentation! What is the fraction of released particles (thus mass of plastics) that is retained in the coastal zone + beaches? Thank you!

Matthias Egger: Great work! Did you also run a simulation where you turn off the tap (no more inputs from land)? If so, what is the fraction actually beaching vs. remaining afloat? Yes :)

For Delphine

Name:...Cleo Jongedijk

Question 1:...your grid resolution of 9km compared to your beaching region of 10m is quite large, do you know how well the hydrodynamics in the cells adjacent to coasts represents the actual currents there? Can you see a difference between 10m and say 100m or 1km?

Astrid Delorme; thank you for the presentation: when you say beaching of the plastic, how long does the plastic have to be on the beach before you classify it as 'beached'? or is it just as soon as the plastic arrives at the beach from the ocean that it is then 'beached'?

Name: Matthieu Mercier

Question 1: Does this type of modeling suggests where (and when) to collect plastic debris that leaked to the ocean through rivers? A cleaning strategy ?

## Questions and comments for the session 24.6\_Me

Name:.Katrin Wendt-Potthoff..

Question 2: Thanks for the comprehensive presentation! Regarding freshwater, may the contact to different materials be more crucial than time? ..

@ Thorsten

Michaela Meyns

Thank you for the interesting talk! How do you measure the surface area of the samples? Hey Michaela, thank you! We have so far measured SSA using BET with N2 or CO2, looking for different pore sizes.

Thanks!

### Name:.Reza Shiravani..

Question 2:.Thanks for your informative presentation. Could you parametrize the sorption as a mathematical function/relationship of the influencing variables (size, polymer-type,...)?.. Hey Reza, I thank you! We have tried to do so with respect to bioavailability using Deep Learnig, For absorbers, the most important parameters were sorbate MW and crystallinity. For adsorbers, this was actually not size but pore volume and O/C ratio.

### To Thorsten:

Sascha Müller, Copenhagen University: I really enjoyed your talk. I wonder how some of your sorption results translate into flowdynamic environments. Particularily, are those sorbents sorbed irreversible or is there the possibility of a reversible process where flow velocity determines sorption capacity/re-release

of sorbants?

### Name: Anita Jemec Kokalj

Based on your extensive research in this field would you be able to make an assumption if the final concentration of adsorbed organic chemical on microplastics when exposed simultaneously in soil would be different for example for polyester fibers or tire wear particles? What would be the main driver for difference her- the surface area or polymer type assuming the same mass conc. ?

### Name: Laura Markley

Question: Fantastic talk! On your last point concerning the low residence time in freshwater and additive leaching, what about particles that remain in freshwater systems, such as those that end up in sediment or otherwise in a "sink" in a freshwater system?

Hey Laura, thanks a lot, and sorry for adressing your question late. Excellent point! If particles "endup" in the sediment they of course continue to release addititives. I assume under these "more calm" conditions the release would even be slowed down. At least this is what we have learnt from our recent experiment. More to come, hopefully soon...;)

Jes Vollertsen's presentation

Name: Alexandre Dehaut. Thanks for this interesting presentation and question I would say.

-Quickly, isn't the matter that results are presented as a number of particle and not a mass ?

-To you knowledge, is there a way to estimate the forgotten cherries (mathematical approach)? Like counting approach of the most probable number in microbiology?

### Name: Ludovic Hermabessiere

Q: thank you for your interesting presentation, big topic! If you size fraction your samples, can you use quantification method such as Py-GC/MS? Here you will have a mass per different size fraction

Name: Sabine Pahl. Thank you for the interesting perspective. I'm intrigued that you said human bias was a problem in the more detailed analysis, can you explain this a bit more - in which way or directon were the researchers biased? Thank you, fascinating!

#### Name: Alice Horton

Q. Thank you for an excellent talk! You didn't mention much about spike recoveries - these can go some way in helping us to understand the effectiveness and limitations of our recovery and detection?

Thanks for the great talk. Ilka Peeken Why do you think the vision method is still recommended for monitoring e.g. GESAMP

## Questions and comments for the session 24.6\_Ma

Questions to Lorena Mendoza (don't hesitatie to add your email address)

Name:Melissa Question 1:Thank you for the talk. What mass did you use for extracting the POPs?

Name:...Juliana Ivar do Sul Question 2:...Its a pleasure to see you here Lorena. I know you have been working in the field for many years, how do you see we have evolved from 10 years ago to now?

Name: Sam Athey Question 3: Great talk! How did concentrations of POPs associated with plastic compare to surrounding sediments?

#### Name: Joanna Gonçalves

Question 4: Thank-you for a great presentation. My question is in regards to the collection of samples. From the pictures in your presentation it seemed that the vials/containers for sample collection were plastic. How did you rule out any plastic that may have leached from the containers ?

Questions to Simone Lechthaler (lechthaler@iww.rwth-aachen.de)

#### Winnie Courtene-Jones

What method/tracer did you use for the sediment chronology?

i am really interested by the polymer composition with sediment record can you elaborate more on this - i know there are several studies now looking at this in marine sediment cores with very different results (some show multidecadal trends, some don't)

- yes it's hard with the dynamic processes, and bioturbation. Thanks, all the best with your studies just a comment - do you involve a cleaning step after the canola oil extraction (i have used this method too - so can speak from experience), the canola residue interferes with the FTIR spectum, cleaning with a solvent, e.g. ethanol/reagent alcohol will help obtain better spectral data - excellent

Dear Winnie, thanks for your comments and suggestions! A further step will definitely be to use common sediment dating methods to validate our idea of dating recent sediments with microplastics. To prevent the influence of bioturbation and infiltration we decided to sample layers of 10 cm but, of course, there might be less dynamic processes within marine or lacustrine environments. Maybe we can get in contact on more information, the related publication is in review right now and I will send it to you as soon as it is published. :) sure my email is winnie.courtene-jones@plymouth.ac.uk - please drop me a line to chat more :)

#### Pruvost Jean

What are your hypothesis for explaining the 1930s microplastics ? Thank you very much for the presentation

Dear Jean, thank you for you comment. Since plastics are definitely older than 1950 there could also be an accumulation in the environment. We just used 1950 as 'zero point' as mass production started then and thus to make the method more applicable.

André-Marie Dendievel Hi Simone, thank you for presentation! You showed a nice map of the spatial distribution of MPs. Did you try to track MP sources along the river? Ok thanks :) Thank you André-Marie for your comment - this is an interesting connection and a good suggestion for further work! :)

### Name:...Juliana Ivar do Sul

Question 1:...Super cool presentation Simone! Thank you for getting in contact! what I think is very cool is to use the plastics as markers of past events. This idea has a big avenue to be explored in Mp research field! Thank you very much, Juliana, I also think this could be a great application for MP research in the future! :)

Questions to Lisa Zimmermann (l.zimmermann@bio.uni-frankfurt.de)

Comment to Bruno: please move your pointer from the screen, thanks!

Name:...Judith Weis

Question 1:...For Lisa Zimmermann - are you going to do similar experiments with more environmentally realistic concentrations?

Boris eyheraguibel

Thx for the talk ! Do you consider biodegradable in your experiment ? Biodegradation of oligomer Do you think biodegradation could by micro organism could modify the results

#### Name:...Stephan Rohrbach

Question 2:...Great talk! Are you using durable PLA or could it be that you have some PLA degradation (hydrolysis) causing an increase of lactic acid and pH reduction in Daphnia? Yes you answered it already before. Thanks a lot.

MIRIAM/HYDRA: PLA is know to be biodegradable under indrustrial composting conditions which cannot be transferred to the open environment. Come to my session, its the next one :) GREAT :))) Stephan: I read something about surgery material made of PLA which is self-degrading due to the body humidity...But maybe its a different production method.

and increased temperature (body). You need increased temperature, acidity, humidity to trigger hydrolysis before real biodegradation can kick in. You have hardly such conditions in the open environment, therfore the PLA tests do not work in the environment. Very tricky and would be great to have a PLA which is biodegradable in soil, freshwater and marine ecosystems, however it needs clever material scientists to develop it.

But I will definetly join your talk too ^^

EXACTLY :)))

Name: Inger Lyngstad

Question: Do you think that the fact that chemical toxicity is greater than the physical toxicity in some plastics is due to the polymer type or that the findings will differ among plastics of the same polymer type as well?

Name: Charlene Trestrail Question: For the PVC, why do you think there were no physical effects? [thanks, you've just answered it]

# Questions and comments for the session 24.7\_O

FOR PRESENTERS, please insert below your email for any additional questions later on:

- Lowenna: lbj203@exeter.ac.uk
- Matthias: matthias.egger@theoceancleanup.com
- Erik: E.vanSebille@uu.nl
- Delphine: d.m.a.lobelle@uu.nl

QUESTIONS TO Delphine Lobelle below

#### Name:Matthieu Mercier

Question 1: When particles transit from floating to sinking, they have a nearly zero 'sinking/floating' velocity and then are transported vertically by the oceanic flows. Does your model have this aspect in it? Because vertical transport can alter growth/respiration/etc....

#### Name:Magdalena Mrokowska

Q: Have you accounted for the physical parameters of biofouling material in the model i.e. denisty? Thank you for very interesting talk and answer to the question!

Astrid Delorme: Interesting talk! have you considered that the density of the plastic material itself could possibly be altered as it is degraded?

Federica Guerrini

Interesting talk and topic! Does the model account for biomass removal from particles? I mean, can modelled particles be resuspended if biomass dies and detaches from particle surface?

Matthias Egger: Could you use your results to see how the relative abundance-size distribution varies spatially in the global ocean for plastics of different sizes? Different sizes lead to different sinking timescales, and thus different ratios...

Stephen Kneel

Have you considered modelling MPs in rivers using this method? or are there too many variables? Great talk by the way very interesting. Thanks for your question, Stephen. I have not looked at rivers yet, no!

#### QUESTIONS TO Erik van Sebille below

Name: C. Maes Question 1: Can you tell more about the total surface currents as measured from Space (not the case at the moment) ?

Name: Reza Shiravani

Question: Why did you apply the Lagrangian approach for your model instead of Eulerian. The lagrangian approach is highly expensive and can not reproduce the depth dimension.

From Britta Baechler: Great talk Eric, Thank you! Do you consider subsurface currents in your work? Do subsurface currents concentrate plastics in polar zones or in gyres? victor martinez: in terms of observations from remote sensing can you give 1 to 4 most to less important physical processes to target? No pressure :-)

Federica Guerrini

Thank you for the interesting talk! Did you look into the effect of wind on plastic distribution in the water column, or it can be already accounted by using 3D models for ocean currents?

QUESTIONS TO Matthias Egger below

Name: Matthieu Mercier

Question 1: A "balanced" model for fragmentation based on one lengthscale is a bit strange... Small pieces from a large pieces is not properly described by solely one lengthscale. Did you do a measure of more than one dimension of the objects collected?

Name: Holly Nel - We often try fill in the missing gap? Could the model be slightly off - I see you are answering this at the moment. Thank you for a interesting talk.

Astrid Delorme: What polymer(s) do you use for your models? Great talk, and very interesting work!

Matthias: Regarding the model questions: There is a good description of the model in the Cozar et al. (2014) PNAS paper. https://www.pnas.org/content/111/28/10239.long We didn't develop the model ourselves, but used theirs.

Robby Rynek: Thanks for the interesting talk. Could you provide the DOI of the publication you just mentioned?

https://iopscience.iop.org/article/10.1088/1748-9326/abbb4f Thank you! You're welcome :)

Reza Shiravani: The tourbulence model for water surface could be one of reasons of size gap. How does the model consider the turbulence?

We corrected all concentrations for wind-induced mixing at the ocean surface. So all values reported are the integrated concentrations for the upper 5m of the water column. The correction is based on the model by Kukulka et al. (2012); GRL

https://agupubs.onlinelibrary.wiley.com/doi/full/10.1029/2012GL051116. Note that there is a great explanation on how this correction is done in the Supplementary Materials of the study by Lebreton et al. (2018), Sci. Rep. https://www.nature.com/articles/s41598-018-22939-w

Victor martinez: could another explanation to the particle size distribution a lack of methods to measure the smallest fractions?

QUESTIONS TO Lowenna Jones below

Name: Erik

Question 1: Some of the samples (e.g. near Svalbard) were taken much closer to the coast than others. Do you think that has an impact on the interpretation of your results?

Name: Holly Nel

Question 1: How feasible is it to collect a sample by a citizen or sailor out at sea? Especially as you may need a special net? Thank you!

## Name: Cleo

You used a custom made net, which I presume you distributed to the sailors that volunteered to sample. How do you feel about automated sampling system on sailing boats as such they performed during most of the legs of the volvo ocean race on a couple of competing sailing boats? (This is a nice perspective: we have currently on the Vendee Pace some boats equiped with MPs

(This is a nice perspective; we have currently on the Vendee Race some boats equiped with MPs sensor - Christophe Maes)

Hi Cleo - thank you for your question. So I actually collected the samples myself but our aim was to predominantly focus on floating debris at the surface. From my understanding the samples collected during the Volvo and Vendee races were obtained using sub-surface pumps. I think this is fab and if you have cooperation from sailors then there is definitely a space for it but it was our aim to ensure our methodology could be utilised by everyday users.

Name:...Isabel Goßmann

Question 2:...What about possible contamination during the sampling? Do you have some kind of "ship blanks" ? Thank you! :)

# Questions and comments for the session 24.7\_Me

Maja Grünzner: Here the link for all my plastic experts :) https://plymouth.onlinesurveys.ac.uk/mpperceptions For further questions: email: maja.gruenzner@univie.ac.at, twitter: maja\_gruen

Name: Giorgia Question: Should we prefer, in such situation of scientific uncertainty, soft or hard law instruments at the international level?

Name:... Vesna Teofilovic Question 1:...Is there a way to influence governance to make such laws? How scientists and public can "force it", to make it priority?

Name: Ryan Shum

Question 2:... Thank you so much for the presentation, I was wondering what role the precautionary principle plays ( currently and in the future) in the governance of microplastics?

Name: Elisabeth Schmiedel

Question 3: What are the barriers responsible why the government in e.g. Germany was not able yet to address the micro plastics problem in a way that citizens are aware of release of microplacstic in the household? E.g. from plastic nets from food or flushed packaging?

Maja Grünzner: Thank you for this great presentation. What do you think, how (in what settings or with which tools) can we bring scientists and politicians together and make them talk to each other?

Name: Sabine Pahl. I was surprised to see the evaluation category was lower for MP than energy transition, any idea why that was (hope I'm remembering the data correctly) Very nice, thank you - I wonder if there's another level of analysis, maybe distinguishing between positive and negative evaluations? We can discuss more...:) Thank you for your question and looking forward to it Sabine! I will say that in the original study, Böhm et al did distinguish positive and negative evaluations, as it was one of the sub-categories, about which i did not talk in this presentation due to time limitation!

### Vesna Teofilovic

Do you think that labeling of products (about microplastics content or negative effect to environment, etc.) could affect behavior of consumers. What do you think that this label should contain to be effective?

### Elisabeth Schmiedel

Do you think that the relationship to the environment is more or less important than labeling or incentives from the government? And why so? Maja: I think both is really important, esspecially because citizens need to accept the incentives from the government. This could be easier, if people care about the cause. As microplastics are a systems problem, we should tackle it systematically from every possible direction.

Laura Markley: Will you be looking at any differences between expert opinion and non-expert perceptions of the microplastic issue? Maja: As Kai mentioned that concern is high for MP when asking lay people, but experts have no clear results yet, I am highly interested in looking at experts and non-experts opinions. Moreover, it is highly relevant for communication. I hope to get more insights from Marcos research as he is looking at lay peoples mental models. For now no study looking particular at non-experts opinion is planned. Hope that makes sense.

Sabine Pahl: please share the link for the new survey, so that people can start filling it in - we need everyone's input!

# **Questions and comments for the session 24.7\_Ma**

## WELLCOME TO 'BIODEGRADATION - CHALLENGES & OPTIONS'

Please do not hesitate to also write to the speakers by e-mail katharina.a.schlegel@basf.com c.lott@hydramarinesciences.com m.

Speaker: Katharina Schlegel

Name: Xavier Cousin Question 1: Is such normative work in progress for marine environment at EU level ? At national level ? (where)

Name: Matt Cole. Question: Should there be better descriptors /quality standards of what "bioplastic", "biodegradable", "compostable" mean? Would this not help public understand how to dispose of waste.

### Lisa Zimmermann

What does BASF do in the field of biodegradable polymers? What is you opinion on (toxic) chemicals in biodegradables? Should they not be avoided since prone to end up in the environment? How can that be implemented? The certification EN 13432 only prescribes examinating the effect of resultant compost on plant growth (agronomic test). This is not sufficient to guarantee absence of toxicity for animals!

MIRIAM/HYDRA: Very good point. We need to support adaptation of current standard test methods and specifications in case of gaps asap. Or support development of new ones.

### Stefan Dittmar

In my eyes, biodegradability and suitability for the desired application always (as you also mentioned) recquire a compromise regarding the material choice. This compromise is probably quite specific depending on the application. With that in mind, do you think it makes sense at all, to employ (a variety of) biodegradable plastics in consumer products and thus having to integrate them into the waste (separation) system challenging both consumer behaviour as well as the technical systems involved (e.g. composting conditions/times)?

Alexandre Dehaut

Any clues for the research on the amount / names of the non-toxic chemical added in biodegradables?

Xavier Cousin One issue with some regulatory tests used is that they are acute tests not necessarily suitable for assessing plastic toxicity... Miriam/HYDRA: Absolutely right, we have some gaps to fill asap.

Speaker: Lott Christian

Name:...Judith Weis Question 1:... What happens when an intermediate breakdown product is quite toxic?

Name: István Szabó

Thank you Christian, for the interesting presentation! Is the microbial community in the lab and tank tests are controled (known)? Or is it just originated (coming) from the sea water? Thank you for the answer! May I ask for the mentioned article's DOI?

Name: Ana Lacerda, FURG- Brazil Thanks for the very interesting talk, Christian! Please, could you state what were all the "relevant conditions" you considered?

Name: Stephan Rohrbach Great talk, To which extent can fragmentation or abrasion due to abiotic processes contribute to your detected biodegradation data?

Name/ Boris Eyheraguibel Q: to calculate the half life , you consider désintégration of material as biodegradation/mineralisation ? Julia Taylor

How can you know where the plastic ends up? Plastic in the marine env move to different areas so if you know the situation for one certain marine env, how do you know that the plastic won't move to a different one? Thank you for a very interesting talk!

Speaker: Weber Miriam

Name:Zhiyue Niu Question 1:Will you also look at the role of biodegradation onto plastic fragmentation (into microplastic/nanoplastic)? Thanks!

# Questions and comments for the session 24.8\_O

Joao Frias: Full details of this work in the open-access paper "Floating microplastics in a coastal embayment: A multifaceted issue": https://doi.org/10.1016/j.marpolbul.2020.111361. More details about the project can be found at https://www.joaofrias.com/impact-project And about our other projects in GMIT https://mfrc-gmit.ie/meet-the-team/dr-joao-frias/

Name:...Catharina Pieper Question 1:...great talk joão! I was wondering if these wind measurements were taken only during sampling days? thanks!

Name Joao Frias:... Question 2:...You mentioned that one of your sites had 658 items kg-1. How do your results compare to similar studies around the Mediterranean? Are they higher, lower?

# Questions and comments for the session 24.8\_Me

Speaker: Fischer Elke Microplastics in marine species within the Wadden Sea along the coastline of Lower Saxony, Germany - Schwarz Matthias, Polt Laura, Dau Kirsten, Fischer Elke.

Name: M Question 1: why do you think it is that there are more fragments compared to fibers found in the samples? why do you choose Arenicola marina to focus on for the further studies?

Name:...Carolin Philipp Question 2:...Thank you Elke and Sedat for your nice and informative presentations!!! :)

Speaker: Gundogdu Sedat Occurrence of microplastics in the gastrointestinal tracts of Chelon saliens along the Turkish coast -Gundogdu Sedat, Çevik Cem, Temiz Ataş Nihan.

Name:M

Question 1: how do you think this knowledge can be applied to management and regulations?

\_MICRO 2020. Fate and Impact of Microplastics: Knowledge and Responsibilities. 23-27 November 2020\_\_\_\_

## Name:...Camila Vidal, University of the republica, Uruguay

Question 3:Question: You spoke about types of microplastics from the gastrointestinal tract of the fish, but did you analyze by color and shape? specially because you mencioned having analized other species. Great presentation, thanks!

# Questions and comments for the session 24.8\_Ma

Name:...Stephan Rohrbach

Question 2:...Great work. Sorry if I missed it. Have you performed also some sort of comparison between plastic fibres and cotton fibres or other types of fibres?

Name: Gerardo Pulido-Reyes

Question 3: Great poster, thanks! Could you please say more about how strong is the interaction between the particle and the metal? No leaching at all? Great! Thanks

# No questions or comments for the Session 24.9\_O

# Questions and comments for the session 24.9\_Ma

Name: Garth Covernton Question 1: Sorry if I missed this, but how did you collect the feces?

Name: Àlex Cortada

Question 2: Could be some interactions with the content of plastics also of its prey (like bioacumulation)? Independently of the direct microplastics present on the water that they could swallow during ingestion Thank you!

Name: Stephan Rohrbach: Do you see a difference in the amount of bacterial and fungal biofilm on the particles?

Are you also adressing archaea? Thank you

Remark: I think there is an updated SILVA database :) [should be SILVA 138] But it might not affect your results at all

Name: Àlex Cortada

The change in bacterial communities after the 14 days could be somehow also relatedc with competence between bacterial species such as a colonization process---> ecological succession? Thank you!

Answer: hi Alex. Yes, there's a lot of other processes we are keen to understand. We used the pilot study to help us better understand the sampling time points we needed for the large scale experiment. we intended on doing more frequent early sampling in the big experiment but sadly we got the structures in the water 3 days before New Zealand went in to a national lockdown and we couldn't get back to them for 3 months. We are hoping to capture thone missed early time points when we redeploy in March 2021.

For Nia

Name: Matthieu Mercier (IMFT, Toulouse)

Question : Do you plan to repeat the observations in the future? Do you record more information on the water column (stratification, currents, etc.)? Thanks!

# Questions and comments for the session 24.10\_O

Name:...Karolina

Question 1:... Hi there, interesting work! Are you planning to develop this study in the future? Eg by using a bed made out of natural grains?

Name:...Stephan Rohrbach

Question 2:...Awesome poster: I don't know the cost of this process and if you can upscale it, but wouldn't it be a great technique to recycle PET waste in general? I mean 100% percent degradation is amazing.

Name: Camila Vidal, University of the republica, Uruguay

Question: You spoke about types of microplastics from the gastrointestinal tract of the fish, but did you analize by color and shape? specially because you mencioned having analized other species. Great presentation, thanks!

# Questions and comments for the session 24.10\_Me

Name: Christina Bogner

@Lea: Question 1: Thank you for the presentation. What are the species feeding on? Thank you @Christina Bogner: Thank you for your question. They are mainly feeding on zooplankton (I found several remains of Antarctic krill (Euphausia superba) in P. antarcticum's stomachs), but also on fish, when nothing else is available.

Did you have access to their food or examples of it? Does it contain any MPs?

Unfortunately not, I only had the prey items I could find in the stomachs. I have found fibres that were trapped in the krill remain (chitin carapace), but I could only identify these after the enzymatic digestion, and these fibres are most of them of natural origin. Thanks :-)

You're welcome, thank you for your interest! :)

Name: Christina Bogner

@Aghathe: Question 2: A question from a non-zoologist :-) How did you get the fish to ingest the same amount of food? :-)

@Christina: Thank you for your question. That's one good advantage of working with trophic chains and especially live preys: the fish prey on moving preys (contrarily to inert food) so you can be sure that they will eat everything. Every fish was given the same number of prey and all the prey were ingested, so we know precisely how many were ingested in total.

Sometimes it is just straight forward :-) Thanks.

You're welcome, thanks for your interest!

Name: Nina Paul

@ Agathe: Thanks, very interesting study. What was your MP concentration that you used? (maybe I missed it :) )

@Nina: Thank you Nina! we used a quite high concentration initially (~1mg/ml) to contaminate the artemia, then we checked how much they ingested and on average each artemia ingested ~200 particles. So that's (on average) a total of ~10 000 particle/fish over a month.

Thank you very much, Agathe!

You're welcome! Thank you for your interest!

These kind of findings are actually very fruitful with respect to discussions about MPs being vectors for contaminants etc. Thanks :)

## Questions and comments for the session 24.10\_Ma

Name:Sam Athey Question 1:Great talk Alicia!! Which chemical contaminants will you be analyzing? (Sorry if I missed it!).

Name:...Nicole Posth

Question 2:...nice talk, both! Alicia, I think that I missed it. What are you sampling for with your landers over the course the year?

Day 3/5 Pads

# **Questions and comments for the session 25.1\_O**

### Name:Elena

Question 1:Thanks for your nice talk! Which method did you use to identify your "suspected" microplastics? Were all particles analyzed by Raman spectroscopy? Yes definitely, thanks for your answer!

### Name:...Sonja Ehlers

Question 2:...Thank you for this really interesting talk, Carolin! Do you know a reason why there were more MPs in the porpoises when compared to the seals? Because of the feeding type/position in the food web? Is the accumulation in their bodies different? -Thank you very much!

### Name: Juliana Ivar do Sul

Question: So cool work! thank you. How did you digest the fecal samples? did you have also notes on what they ingested? (natural food items)

Excellent! I was wondering if the stomachs were empty but you already answer that. thank you.

### Name: Carmen González-Fernández

Question 1: I was the first presenter today in Margulis' Room. If you have more questions you can write me an email. I will be happy to answer all your questions. My apologies for the hurries on the presentation... this is my e-mail: carmen.gonzalez1@um.es. Thank you all! hi Carmen, this is Ostroms room ;-)

#### Kristina Klein

Nice work! Regarding the 24-h growth inhibition, how do you explain the higher impact of oligomers compared to the NP + oligomers?

-- Dear Kristina, thanks a lot. What do you means about oligomers? Actually, we only detected growth inhibition upon PS-NH2 nanoparticles and that can be associated with the charge of cell's lipid membranes (which are neutral/negative) and react more to positive charges as we have previously observed in other cells types ;)

MIRIAM/HYDRA: Very nice talk and important work. Thank you! Yesterday Lisa Zimmermann showed nicely that MP impact is mostly physical by the particle. Can you comment your study in respect to Lisas finding? Did you use also, lets say, pure particles to understand if we phase such physical impacts by any MP and NP?

#### Miguel Tamayo:

Thanks Miriam, unfortunately yesterday i couldn't attend Lisa's presentation but i' d love to ask it for her, i alway follow her interesting works :) (please @Lisa could you also share me the video). In any way, i did'nt use any referent particle to compare its effects with those we observed with the polycaprolacton secondary nanoplastics; our aproach aims to elucidate wheather among the debris (generated on te same way and from the same material) those nanoparticled worth special attention base on their effects; for that reaseon, we compare bothe the mode of action of the nanoplastics by measuring several physiological parameters under the exposure towards nanoplastics vs free oligomers. Regarding the phisical impact, biodegradable plastics present more reactive surface than traditional ones, and, at least in the case of nanoplarticles, the physical effects may be triggered by different ways (block essential channels, altering the membrane shape or potential, making nanoholes...) but in any way probabely we should includ any inhert nanomaterial al reference to compar, thanks a lot!

They were commercial particles of polystyrene and they were checked by Raman microspectroscopy analysis. The culture media is Gibco<sup>™</sup> Leibovitz's L-15 Medium and was pH stabilized with HEPES solution. Nevertheless I didn't check changes in pH solution. I will consider it for future experiments.

I'm performing now tests with high resolution microscopes to assess if particles are internalized in cell's membranes and it seems that they transfer easily lipid membranes keeping in cell'cytoplasm Thank you Michgel!

A question Lisa got might be intersting for your study too: Did you measure th pH in the solution you exposed? The degradation processes can change the conditions, expecially in small volumes quite a lot.

Yes, we measured the pH at the beginning and at the end of the degradation assay; we performed some preliminar degradation essays and we concluded that the best and more realistic condition would be to do it in phosohate buffer at neutral pH (at 1 mM the pH little acidify during the degradation but it don't vary more than 1 pH unit)

Lisa Zimmermann: @Miriam: I actually measured the pH in exposure suspensions, also in that with PLA, and it didn't change during the experiments (missed to mention that yesterday). Besides, what I wanted to point out is that it might be either the chemicals or the physical properties that drive MP toxicity. This strongly depends on the individual microplastics and its chemical and physical properties.

@Miguel: Due to time shifts I, unfortunately, missed your talk but i'm very interested in the topic and how you are proceeding. Any chance that you can share your talk (and maybe your thoughts) with me?! l.zimmermann@bio.uni-frankfurt.de

Annegret: Thanks! How could you distinguish between Nanoplastic & free oligomers and free oligomers only? this is a preliminary study, we will deal on all of that soon.

Questions for Winnie Courtene-Jones (winnie.courtene-jones@plymouth.ac.uk) Thank you so much for your questions and interest, i will answer below.

Name: Catharina Pieper

Question: Thank you for your presentation. I was wondering if you have any idea why there might be so many more pellets in one trawl and almost none in the other locations ? thank you very much!

Name Steve Allen for Winnie, very interesting talk and looks like a great sail. Did you stop at San Blas? We stopped at islands on the Pacific side and saw council rubbish dumps were just a cliff edge over a tidal beach. I wonder if San Blas has the same system. Might explain the high counts.

Name: Sonja Ehlers

Thank you for your interesting talk! If I understood it correctly, fibres were excluded from the analysis as they might have been introduced to the manta trawls as contamination. As fibres usually make up a great proportion of MPs in the environment, could you think of a way to control for such airborne contamination so that not all fibres have to be excluded from the analysis?

-Thank you, Winnie.

Delphine Lobelle- very interesting results! Could you explain your thoughts on why there are negatively buoyant particles found on the sea surface? Thank you very much for your response!

### Erik van Sebille

Nice presentation Winnie! Do you know what could have explain the variability in amount of plastic between the trawls and regions?

### Tim van Emmerik

Thanks for this tropical talk! Do you know anything about the main sources of marine plastics in the Caribbean?

### Marie Russell

Hi Winnie - nice talk - did you relate your polymer differences to any differences in current/tides Thanks Marie - we are working on this at the moment, i can't say too much at the moment as the analysis is not complete, but i aim to bring in data regarding the tides/currents in the area to consider the data on plastics more fully.

### Marie Babinot :

Such a great talk! In the sediments it is possible to find mostly fibers, but not in the surface water. How to explain this difference?

Thanks for the question, the results i presented today were only from surface waters. We have also sampled sediments and are working through those - these results have been a little delayed (due to COVID-related lab closure over the summer- as i am sure many of us encountered). from experience analysing other sediments, we usually find a mix of types of plastics. it will be interesting to see how they relate to surface waters.

Thanks Winnie !

Matthias Egger: Great talk, thanks for sharing Winnie! You mention country specific differences. Do you think these are due to local differences in the composition of plastic waste, or maybe the result of oceanic circulation patterns?

Mariella Siña: Have you tried taking subsurface samples? what do you think it would be expected? Thanks for sharing your work Winnie

yes we have, we have collected sub-surface water, bulk samples, with NISKIN bottles. i am working through that data, these results have been a little delayed (due to COVID-related lab closure over the summer- as i am sure many of us encountered). but i look forward to relate these datasets. as i mentioned, bulk samples, e.g. NISKIN bottles are better for considering fibers, as these can pass through the manta trawl net because of their size.

## Name : Prasun Goswami

Nice talk Winnie, Seeing your data, I feel very interested to know how is the effects of local currents in the distribution of the MPs in the Carribean. Can you comment in this?

i think i answered this in the discussion, but i think it's the big question, and very likely a combination of both. some of the analysis i mentioned above, which we intend to carry out will hopefully provide some more information on this.

## Questions and comments for the session 25.1\_Me

Speaker: Thibault Masset
Name:... Matt Cole

Question 1:... Great talk. Is pH the main factor in enhanced metal desorption in biological fluids compared with control? Or are there other parts of the gastro/intestinal fluid that might enhance desorption?

Name: Lukas Kruckenfellner Question: Thank you for your great talk! How do you plan to conduct (which set-up and scale) the mesocosm experiments?

Tomasz Burghardt: Quite interesting indeed. Good job. Do you know what was the source of lead?

Anita Jemec Kokalj

There is a new review paper on the solubilisation of nanomaterials in digestive fluids, might be useful to compare to these particles.

van der Zande, M., The gut barrier and the fate of engineered nanomaterials: a view from comparative physiology. Environmental Science: Nano.

Thibault Masset : Indeed, it would be interesting to look at this paper, thansk much for the link !

Stephan Wagner

Great talk! Based on the dissolved Zn conc. you observed would you expect any effects based on literature values?

Hello Stephan, thank you for assisting to the presentation. I did not yet looked much in the litterature regarding the effects of the dissolved concentration obtained here. The main issue is that we obtain a value in mg.L of fluids which is a metric rarely presented in the litterature. Which impact the concentrations measured here will have on organisms is the main question to answer, and we will have some answers with the toxicological testing planned in ou projects !

Also, obviously the amount of TRWP put in contact with the fluids will impact the solubilized amount and need to be kept in mind...

thanks! I am really curious about the results of your study :-)

Hazimah:

Nice study :) Would you investigate what type of metal complexes form with the compounds in the gut fluids?

Barbro Melgert: you focus on soil and water, but these particles are also in the air and may be inhaled, any plans on testing this with lung fluid?

Bettie Cormier:

Great talk, thanks a lot! Can you tell us a bit more about the composition of both biological fluids you used? Is your work published already? We did some similar "pre-experiments" with fish, and so I am interested in your M&M. Is it possible to exhange about it? Also interested in your PhD thesis :) Thanks in advance!

Thanks Bettie for your comment ! The SGF was composed of various salts, Pepsin and adjusted to a pH = 2 to mimic gastric fluid composition of our model species. The SIF was composed of natural Bile extract, Pancreatin and the non-adjusted pH was 7,4. We can exchange about it if you want, here

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my email address: thibault.masset@epfl.ch. Thanks a lot! I will mail you for sure :) Many thanks and again, great talk!

Speaker: Isabel Goßmann

Gabor Bordos:

Interesting study, thanks! In which format did you use the tyre particles to calibrate your Py-GCMS system? What was the limit detection? Hey Gabor, thank you for your question. I "shred" the tires, weighed them in different concentrations and then performed several measurements. The LOD was 20  $\mu$ g for car tires indicated by SB, for truck tires the smallest calibration point (1  $\mu$ g) still had a S/N of 12.

Thanks Isabel! How did you shred the tyres, what was the smallest amount that you could measure gravimetrically?

Isabel, we can have this conversation after Ana's talk :)

Elisabeth Rødland Great talk. Did you see a variation in the SB marker in your 15 reference tires?

Stephan Wagner

Nice talk, Isabel. I was wondering what is the reason for the difference in truck and passenger tire concentration you detected? any idea?

Speaker: Ana Luzia Lacerda

Name: Hazimah Question 1: Interesting study. Did you compare the diversity in the plastic with the diversity in the surrounding water?

Name Stephan Rohrbach

Very interesting talk. Sorry if I missed it firstly I want to know how long have you incubated the particles in the ocean and have you noticed a reduction of archaeal abundance on the plastic particles in comparison to the community in the free ocean?

Jafar Alomari , Szent Istvan university , Hungary very intersesting PPT, was the community DNA isolated from single plastic piece enough for amplicon sequencing?

Name: Prasanth Babu

Question: Interesting study. I wonder are there any multispecies biofilm community found in a particular microplastic type. If so, could you name few class of such microorganims based on their DNA datasets.

Yes, in terms of functional taxanomical level. Thanks Ana for an elaborate answer, I would like to discuss about over to your mail. Yeap, so please send me an email at analuzialacerda@gmail.com I'll be very happy to discuss it with you :)

StephanROhrbach remark: Hey Ana you could have a look on the poster from yesterday presented by Olga Pantos it might help you to get a better understanding of how the MP type impact the community. Hey Stephan,thanks for the suggestion! I already saw it and it seems that established \_MICRO 2020. Fate and Impact of Microplastics: Knowledge and Responsibilities. 23-27 November 2020\_\_\_\_

communities in plastics from environmental samples are composed by generalist organisms, and the plastic type do not drive their composition...

Name: Gloria Fackelmann Question 1: Have the fastq files also been made available?

Name Stefan Lips:

Question: When is the paper ready for reading? That is a good question!! haha But I hope it'll be published soon. You can add me on Research Gate to see the updates. Would be a nice christmas gift :) Definitely!!

# Questions and comments for the session 25.1\_Ma

SPEAKER e-mail ADDRESSES: carmen.gonzalez1@um.es Marte Haave: maha@norceresearch.no /marte.haave@uib.no Rawil Fakhrullin kazanbio@gmail.com

Name: Carmen González-Fernández

Question 1: I was the first presenter today in Margulis' Room. If you have more questions you can write me an email. I will be happy to answer all your questions. My apologies for the hurries on the presentation... this is my e-mail: carmen.gonzalez1@um.es. Thank you all!

Name:...Marte Haave

Question 1:...Thanks a lot, great presnetaton. Sorry, didn't get this detail: Controls, was that any other type of Nanoparticle, or no exposure?

Name:...Charlene Trestrail

Question 2:...Great presentation! What do you think it means for the brain as a whole tissue, to have increased oxidative stress? Will it affect the fish development?

Name: Amedeo Boldrini, University of Siena Question 3: Thank you for this interesting presentation. From where did you get the particles? Were they already functionalized?

Name : Bénédicte Morin Have you checked if you have internalisation of the nanoparticles into the cells? Thanks for the great presentation !!

Name: Špela Korez, AWI

Question 5: Hi thank you for your talk. What were the units in the graphs you presented? Were they normalized by wet weight or protein content? Perfect, thanks!

## Name: Nina Paul, AWI Bremerhaven

Question: Thanks for this interesting talk! Do you think that the increased size of pristine PS may have an effect compared to the smaller sizes of the functionalized PS? I need to deep in this data, it is a preliminary study, but in general, bigger particles are not easily internalized so we can expect

higher impact of < 100nm particles. If you want information regarding environmental impact, plastics in natural environments get negative charges more than positive ;) Thanks, Carmen :)

Hi, Thanks for your questions. I was the second speker today, presenting a small study on MP in wild coastal animals. Please email me at Marte Haave: maha@norceresearch.no /marte.haave@uib.no if you have any further questions.

Name: Holger Kress, Univ. Bayreuth

Q: Very nice presentation! How do you expect that the particles are transported into the muscles and the liver?

### NAME: Marte Haave:

Q: RAWIL: Thanks for the presentation, very interesting method. Do you expect to be able to use this method on sections of tissues and animals thicker than a C. elegans?.. or more specifically, do you think it can be used in slices of tissues of fish and mammals? How thick sections if so.

### Name: Andrea

Q: It looked like you were working mostly in visible light. How far into the infrared can this microscope go? and what data analysis methods are you using to approach the large amount of data?

Name: Andreas Eich Q: How does a biofilm affect the identification of plastic?

Name: Josef Brandt

Q: The spectra you showed are often quite noisy.. What are the acquisition times of the hyperspectral camera and, would it help to increase the acquisition times for better spectra? Great work!

Name: Joana Prata

Q: Have you tested environmental samples where microplastics are weathered or having contaminants in their surface? Can you tell microplastics appart from different contaminants (e.g. minerals, organic matter)? How time consuming is this technique?

Name: Svenja

Q: (during the time we didn't hear you): It looked like you were using an environmental samples? How well does the identification work when so many particles are on the slide?

Name: Andrea

Q: It seems like the sample holder and this whole set up is targeted towards very small particles. Is there a maximum particle size? In that vein, what surface area can be analyzed in what amount of time?

Alessio Montarsolo: Are the spectra obtained comparable with those of FTIR ? thx you

Name: Robby Rynek

Q: How does the system perform with particles in very diverse sizes in terms of the focus?

Name: Joana Prata

Q: When you mention 2 hours analysis, what filter area and particle number are you taking this estimation from?

# **Questions and comments for the session 25.2\_O**

István Szabó: Thank you for the presentation Sergio! Have you published your results yet? Sergio: Yes of course I published this result the last week. This is the reference: Martínez-Campos, S., González-Pleiter, M., Fernández-Piñas, F., Rosal, R., & Leganés, F. (2020). Early and differential bacterial colonization on microplastics deployed into the effluents of wastewater treatment plants. Science of The Total Environment, 143832.

Name:...Antje Wichels Question 1:...thank you Sergio! How would you explain the higher diversity on MP material as compared to the water column?

Name: Stefan Dittmar (to Sergio) Question 2: Thanks for the talk! Did you inspect the biofilms via microscopical methods as well? If yes, what did you see? Thank you!

Name: Prasanth Babu (to Sergio)

Question: f Thanks for the talk, Does ARG function of discovered microplastics are persistent with different set of samples? What are the source microbial samples had been extracted at water treatment system, are they biofilms or biofouled particles or anyother source?

Name; Sergio Martínez-Campos Gutierrez Thank you for your attention if you have more question you could write to me: sergio.martinezcg@uah.es

Name: Brittan Scales

Question for Ciara:

Thank you for your great talk, such meta-analyzes are so important to getting an overview of a field. You used a 85% cutoff to define your core. This seems a little low, as the cutoff for defining a core genome is typically 95%. How did you come to this percentage cutoff?

Question to Keating Ciara Name: Katrin Wendt-Potthoff Question: Ciara, is your nice work published? I would like to compare it with cultures (from marine and freshwater)? (katrin.wendt-potthoff@ufz.de)

Ciara - great I will be in touch - we're still collecting sequences but we can share anything that is useful - have noted your email mine is ciara.keating@glasgow.ac.uk

Name: Camille Richon Question: Hi Ciara, Thanks for your talk! Did you observe different communities colonizing different plastic types? Or did I interpret your last figure wrong... ? Cheers!

# Question: Jean-François Ghiglione (to Ciara)

Very interesting talk ! Geographical location looks to be more important that microplastic composition. There is an increasing literature showing that environment matters very much to shape the plastisphere, but we are missing metadata to better describe the environmental conditions. Did you manage to get metadata from the authors ? We should all make an effort on this aspect. And we have data to share with you !

Thanks so much Jean-François we appreciate you sharing your data with us my email is ciara.keating@glasgow.ac.uk

### Antje Wichels:

just a comment:we could offer raw data although we have deposited them of course, you are wellcome!

That's excellent Antje thanks very much my email is ciara.keating@glasgow.ac.uk

## Name: Vincenzo Donnarumma

Question: Hello Ciara! Very nice talk! I have found those core microbiome families myself as well in my data, but I have never found the family "Francisellaceae", is there a specific region or polymers where you found this family?

### Name: Jean-François Ghiglione

We are many time looking at the plastic properties (crystallinity, hydrophobicity, oxidation levels, ...) but rarely looking at the quantity and quality of the organic matter covering the plastic, which I believe in also crucial for the conditioning film.

Did you finally got an idea of how much DOM quality/quantity drive the conditioning film compared to the influence of plastic properties ? How long was your incubation and Do you thing DOM is important is driving the communities even after the conditioning film when the biofilm become mature ?

thanks for your interest, incubation for DOM was 1h in 0.2µm filtrated stream water, then another identical batch was incubated in microcosms in unfiltrated stream water. generally we see that different biofilm communities on different substrates converge with time. so I think the observed specific OM layer at the very beginning becomes less important with time, since secondary colonizers may not even attach to the surface anymore but moreover getting trapped in sticky EPS a I love these session talks ! Thank you for sharing these results and Congratulations to all the speaker !

Great answer. thank you !

### Name: Jessica Song

Question: Hi Christoph. Thank you for the very cool presentation! I am sorry if I missed it but I wonder if you looked deeper into how the selective sorption of the DOM relative to the different materials? If so, what kind of changes did you observe over time? Thanks!

I did not investigate the changes over time, however after 1h sampling we saw different DOM fingerprint for different substrates which is a result of fractionation of the DOM present in the original stream water. glass did not show clear differences between aged and non-aged materials but we detected different OM fingerprints between aged and non-aged PET and PS

Name: Stephan Rohrbach

Great presentation. What type of mesocosm have you used for incubation and do you expect to find same findings in different mesocosms?

We used microcosms or aquaria. I would expect similar trends when re-doing the experiments in different areas, so the underlying processes to be general, but with specific differences in dependece of the incubation DOM and inoculating community.

# Questions and comments for the session 25.2\_Me

For K Amrutha. From Conrad Sparks. What QA was done to assess potential contamination

### Presentation 1

Fluxes of Plastic Debris to a choked coastal lagoon: a top-down approach based on socio-economic data from south Brazil - Santos Itele E. Dos, Fernandes Elisa H., Pinho Grasiela L. L., Abdallah Patrízia R..

### Name: Johnny Gasperi - Chairman

Question 1: What is the quantity of plastic consumed per year and per inhabitant? For a comparison, in France, we consumed about 70 kg of plastic per year and per inhabitant

Question 2: You found between 6 and 30 g/inhab/day. Do you compare this order of magnitude to results based on Jambeck's approch, i.e. by consideriong 2% of mismanaged wastes and 15-40% reaching the sea?

### Name:...Romain Tramoy

Question:...Nice presentation. You said your results are in the same order of magnitude than another recent study. But what method was used for this recent study? Did they use a similar statistical approach?

### Name:...Juliana Ivar do Sul

Question:...Hi Itele, nice work! What about the exportation of plastics to the adjacent marine environment? do you have the idea to include that in your calculations (yes, I agree; and there is fragmentation as well. Thank you Itele! Thank you Johnny!

Presentation 2

Spatial distribution of microplastics in the riverine waters of southwest India - Amrutha K., Kumar Warrier Anish.

### Name: Johnny Gasperi - Chairman

Question 1: How do you collect your water?

Question 2: Considering your limit of observation and your smallest cutt-off (300  $\mu$ m), you fund very high concentrations of plastic per m3. You compare to the littertaure and to other data but do you have the same limit of observation to really compare

### Name: Tenzin

Question: Would you consider to study lower size of microplastics in the future of the same river? What do you think about the concentration of the lower size microplastics? What are the quality control measures considered in the study?

### Name: Aline Carvalho

Question: Thank you for your nice presentation. Why this high proportion of PET, do you have any idea?

### Name: Prasanth

Question: Thanks for the talk, What kind of separation methods you adopted for screening microplastics from river water samples? Does the separation methods influence the quantitative measurement of microplastics?

### Stephen Kneel

Nice Presentation, What did you use to identify the MPs found in these rivers? Were the river samples sediment rich (silt, sand, mud etc)? Also what volume of water did you sample?

Marie Babinot : Thanks for your great and very interesting presentation! Have you taken into account the potential contamination of the laboratory?

Liz Atwood: Great work, thank you. My question is similar to Stephen's, did you also take additional water quality samples to the microplastic sampling?

**Conrad Sparks** 

Question:... Interesting presentation. 1) What QA was done to assess contamination? 2) Were recovery tests done to assess methodolgy? do you think river speed data could be included?

Simone Lechthaler: Many thanks for the presentation! Especially the connection to possible sources was very interesting! So would other sources, such as illegal wastewater discharges, be possible as well? Maybe something like that was visible during sampling?

### Presentation 3

PLASTICOPYR PROJECT. Guidelines for monitoring plastic litter in Mountain riverine systems: from macro- to microplastic sizes

Name: Johnny Gasperi - Chairman

Question 1: For Macroplastic litter, which surface do you investigate for each station? Question 2: For microplastic, what do you plan to do for athmospheric compartment? Which elevation of the mountain station considered Comments: In France, a team in Grenoble is also working on contamination of alpine lakes, whih

Comments: In France, a team in Grenoble is also working on contamination of alpine lakes, whih high elevation. I can give you contact.

Name:...Conrad Sparks Interesting talk. What were the methods for micro & meso plastics and litter?

Name: Tim van Emmerik

Question: Great to see more macroplastic observations! Are you also looking at transport processes? And can you hypothesize what the dominant transport processes in your study areas would be?

Thanks! I agree:)

Stjepan Budimir. Do you have plans for future work with riverine macroplastic in the studied area. Thank you, great work!

## Presentation 4

Microplastics in stormwater runoff and stormwater treating systems - Adyel Tanveer M..

### Name: Johnny Gasperi - Chairman

Question 1: Based on your reveiw of the litterature, can you imagine that a trend/correlation could be drawn between Microplastic concentration in stromwater runoff and land use of the cathment? Question 2: You have significant diffrence between inlet and outlet in your nature base solution site and slight change of size distribution. Do you have any idea of teh contamination of sediment in NBS? What is the hydrualic retention time in this NBS?

### Name: Delphine Dobler

Question:...Thank you very much for your talk. How do you explain that there are more small microplastics at outlet location compared to inlet location in NBS ? Do you think it is linked to larger microplastics fragmentation ?

Name:...Marte Haave

Question:...Adyel Tanveer M: Thanks for the comprehensive talk. In the particle numbers given for different land-use types in Mexico, Denmark etc: do you know the size classes investigated? 300 n/L in Mexico of particles down to x µm..

Name: Trang Nguyen Question: Thanks for your presentation! What kind of MPs used for the exposure testing, pristine or particles collected from stormwater/run-off water? Name: Tanveer Advel: Thanks Trang! We used pristine one

Name: Romain Tramoy

Question: Thanks for the presentation. Can you establish a correlation between the level of microplastic exposure and the physiological response of macrophytes across a range of microplastic concentration? If yes, is it linear or is there any threshold? Ok, too early :). Thank you Name: Tanveer Adyel: Thanks Roman! We are working on it

Name: Stephan Wagner Question: thanks a lot for your work. I am wondering if you plan to determine the overall efffect of MP on the performance of the NBS for your stromwater? Name: Tanveer Adyel: Thanks Stephan! That is the ultimate goal :)

# Questions and comments for the session 25.2\_Ma

For Cátia

Name: Kevin Tallec

Q1: Thanks for the talk! Do you have the EC50 value to compare the sensitivity with other species reported in the litterature? Ok thank you :)

## For ANDY

## Name:Alex Gulizia

Question 1Question about your GC-MS data, you suggest ~2000 different components across all the plastic products. As consumer plastic products undergo a variety of different proprietary reactions during manufacturing, do you think that there could be a lot of shorter chain polymer groups of different lengths that would have contributed to the different GC-MS peaks?

Barbro Melgert: Hi Andy, great talk, can we talk about what is released from nylon? We found mostly cyclic oligomers that in pure form were not toxic in our assays, while the leachate from nylon was itself was toxic, so we are looking for other leachates. You can reach me at b.n.melgert@rug.nl Hi Barbro, I would be very happy to discuss this with you in more detail. I'll send you an email shortly.

## Boris Eyheraguibel

Q : thank you Andy for the nice talk . Did you consider any other solvants to extract the différent compounds? non polar ones for example. did you consider some oligomers (as degradation products from aging ) in your studies

do the chemical would leach out in the water ? in the environment

Do you think some of the chemicals could be biodegraded by biofilm in the environment ? Hi Boris, I think I answered some of your questions already, but not this one. We certainly think that some of these chemicals leaching from plastic products will be readily biodegradable and would therefore have limited persistence potential. However, others are less biodegradable.

### Name: Natascha Schmidt

Question: Nice talk, Andy! Did you test different solvents (and if, were the number of chemicals detected very different?) or how/why did you choose Methanol for the extraction?

Lisa Zimmermann: Great talk and great approach, Andy! Did you observe polymer type specific toxicity and chemical composition (e.g. number of peaks)? What products did you analyze (FCMs?) - I might have missed that.

--> Thanks for answering! I'm looking forward to further results! Thanks Lisa! As I said this something we will now focus on when we have finalised all the chemical and toxicity data. :)

### Name Thomas Maes

Great talk, lots of toys Andy! Did you also check tyres? If so, did the results help explaining some of the findings of the previous speaker? Thanks Thomas, we have characterised the leachates derived from car tyre particles (rubber granulate from end of life tyres) and found a wide range of organic and inorganic chemicals present. In particular, they were dominated by benzothiazole and zinc, but many others were present in lower concentrations: https://doi.org/10.3389/fenvs.2020.00125 (Car Tire Crumb Rubber: Does Leaching Produce a Toxic Chemical Cocktail in Coastal Marine Systems?)

### Name: Kennedy Bucci

Thanks for the great and informative talk! Are you able to correlate complexity of chemical cocktail with toxicity? i.e. did more complex chemical cocktails (plastic products that you identified as having more "features") induce higher toxicity, while products with fewer features induced lower toxicity?

- COOL! very interesting. thanks!

## Name: Bettie Cormier

Thanks for your interesting work and presentation. Regarding the extraction, you used methanol, but we know that methanol extraction is not representative of an "environmental" extraction such as sea water or biological fluids. With a methanol extraction you will get a lot of additives released in the leachate as well as monomers, but what do you think about the "real" release of additives in environmental conditions?

ANOTHER QUESTION: Will you have a post-doc position in 2021 ;) (SINTEF; NILU; NIVA) Thanks Bettie, please feel free to send me an email directly (andy.booth@sintef.no) ==> Thanks a lot, I will send you an email soon!

## Name: Nina Paul

Question: Thanks for this interesting talk, Andy! Did you discover any correlation of numbers of additives to certain plastic types/compositions? (kind of meant in the same direction what Kennedy is asking for). Stay tuned, we hope to present a more complete assessment of the data at SETAC Europe 2021! Thanks, I definitely will :)

## Kristina Klein:

Nice talk! Probably missed it, but what about the 2-3 cryomilled materials? Were they more toxic? Hi Kristina, that is the second stage of the experimental work that we have not yet conducted. We will select the 2-3 materials based on the outcomes of the preliminary chemical and toxicity screening :) thanks ;-)

## For Ula:

Name: Winnie Courtene-Jones

Question 1: Hi, thanks for this, i am sorry, i think i missed what your control was - did you use a different particle, i.e. natural?

also, how long were the particles exposed, to obtain the leachate? could it not have been long enough to obtain very high concentrations ?

- Thank you for your response.

Name: Natascha Schmidt

Question: Thanks for the nice presentation! Did you check what kind of chemicals were in the leachates?

Ula: thank you! No, we have not analyzed the leachates yet. Hope we would do this in the future.

Name: Alex Gulizia, James Cook University, Australia

Question: Did you test if any additives/plastic monomers were present in the leachate prior to doing exposure studies? How did you determine if anything leached out during your time period? Thanks!

Lisa Zimmemrann: Nice talk! Did you perform chemical analysis on leachates? If already published can you send me the publication, Ula?! -> l.zimmermann@bio.uni-frankfurt.de Ula: we did not (not yet, but meybe in the future :)). This research is under the review at the moment, hope it will be published soon. --> Thanks for answerting! Good luck!

Alicia Mateos Cárdenas: thanks for the presentation, I think I missed this, were the MPs also floating in the media and if so how did they get in contact with L. minor roots? I also agree that it's nice to see more studies on freshwater plants :) - thanks for your response Ula: Thank you!

## Name: Isabel Goßmann

Question: Thank you for the interesting talk! What kind of leachates did you expect? What kind of additives did you check for?

Ula: Thank you! We did not (not yet) analyse leachates. But we expect leaching of some chemicals (like heavy metals), as these particles were from an old tyre that was used for some years.

## Name Boris Eyheraguibel

Q: Hi Ula, Thx for the talk . I was wondering what kind of analysis did you do on the TWP. Did you identify any chemicals , or inorganic compounds

Does the original material was aged ? did you evaluate the level of oxidation? In the environment , TRP will be a mix between road particles and rubbers and should sink .

Ula: Thank you! We did perform FTIR of the material, which was similar to FTIR from other study, where they perform FTIR on new tyre material. As particles were from an old tyre it is possible that also some other road particles. Most of the particles were floating on the surface (but there were no mixing during the exposure).

## Name: Charlene Trestrail

Question: Is it possible you had leachate, but they degraded over the leaching/exposure period? Ula: yes, it is possible, but I think unlikely. For being sure, we should perform an analysis before and after the exposure. But pH did not changed during this 7 days.

## Name: Christina Bogner

@Ula: Thank you for your talk. When you compare essays with particles vs. essays with lichates, how can you be sure that the particles containing the chemicals are not leaching during tests? This coulc be physical + chemical. Thank you :-)

Ula: thank you! Yes, I already mentioned, we can just say that if something is being leached, it's not toxic to Lemna. We can not be sure, but if impact was the same when only leachates were in test medium, we concluded that impact was due to chemical leaching. For example, with other mps that we also tested, impact was seen when leachates were tested and impact was similar to the one when particles was tested (so particles+leachates).

Amanda Dawson: Awesome talk. Do you know much about the source of your tyres? Are your particles from a single tyre or a mixture of tyres? Do you think they may have been weathered or leached before you received them? thank you!

Ula: thank you! It was mixture of tyres (as already mentioned). There were weathered during its usage (there were from an old tyre). :-)

# For Ton

Name:...Andy

Question 1:...Do you really think that MP are going to be more toxic than NP? If so, what do you think is driving this difference between the two particle types? I appreciate you can only model the data available in the literature!

Tong: I think it is the size range, MPs is more or less within the range of the ingestion of test species while nanoplastics seems not to be so.

Anita Jemec Kokalj

Regarding the fact that almost all studies currently available on nanoplastics are done for polystyrene NPs- how relevant is this review to make general conclusions about nanoplastics? We found only 4 out of total 200.

Tong: we have 8 entries in our paper that are non-PS vs PS (6 after the removal of sodium azide). this is of course no strong enough. Also, most common plastics such as PET, PE and PP are not present. this has its limnitation~but I think it is good to start the analysis in this direction.

Name: Yuanhu Zhang

Questiong: Thanks Tong, I missed your presentation, but there is one point that is really interested for me. are you still here? You found PS NPs are more toxic than other type of polymer NPs, do you know what the reasons behind this? and did you analyze these investigated Polymer NPs in terms of the purity, addtives ect. thanks

Tong:

regarding additives, yes, we tested the difference after the removal of NaN3 (a common biocide additive in nanoplastic suspension), this is still a difference than PS than non-PS nanoplastics tested in previous studies. you can add my wechat if you have more questions: 18811313852 Yuanhu: Thanks, I do have more questions. see you on wechat

# Questions and comments for the session 25.3\_O

Name:...Charlene Trestrail

Question 2:...Very interesting results. Any thoughts about why biomass is decreased by microplastics? Did you hear the response live?

Name: Stephen Burrows

Question: Lovely talk! Were you able to characterise the microplastics you used at all? Physically and chemically in terms of any additives?

Hey, thanks! Yes. If you want, you can take a look at the paper, we have all the characterisation there. You have the link here: https://www.sciencedirect.com/science/article/abs/pii/S0043135420309064. But shortly, we used POP-free (washed with hexane) irregular shaped polystyrene and PMMA microplastics. All the MPs were under 100 um. We used electron microscopy to characterise the shape and general size, and they were mostly between 40-60/70-80 um.

Stephan Rohrbach to Cesar Cunha: As an idea: You could think about comparing a conventional algal production plant with a plant which is using filtered media. It would be interesting if you see an increase biomass/ fuel production. (Or first in lab-scale :)

Thanks for the idea! Actually, measuring biomass was not in our initial plans. I guess this type of stuff happens when you least expect it to. We were aiming to do the most complete biochemical characterisation possible. But absolutely, now we're looking forward to understand if there are changes in specific cell size, as well as carbohydrates and lipids metabolism. And since we have the means to, we really want to push industrial samples to try to understand this further.

To Francisca Ribeiro Stephen Kneel Hi great talk, did you have any issues with clogging of filters? I am working with cockles and there is a lot of sand remaining post digestion with KOH. I think i answered that during the presentation. Was it clear? :)

## Alex Gulizia

Have you considered how chemical digestion may impact the chemical properties of the MP polymer and skew identification using FT-IR? Yes. So we decided to digest our samples with KOH 10% because it has been proven as one of the most efficient methods for digesting biological samples with minimal impact on the plastic particles. For Py-GC/MS that is not an issue. Considering FTIR, we chose this method to make sure the surface of the particles was minimal impacted. However, the biggest problem with our FTIR analysis is the weathering of the particles surface, rather than the processing methods.

## Maurits Halbach:

How would you explane the high loads in the deploit oysters? 2. Wouldnt we expect lower masses at nanoparticle size? Yes good question. Well we think that the high plastic content in the field deployed oysters is due to the surrounding environment where they were deployed (a maritime port with all the boat traffic associated), but of course this is only a guess. Regarding the fraction < 1um ("nano") in a way we can consider it a surprising result, but there is evidence in the literature that suggests that the nano fraction is probably the most abundant in the environment but is being missed in most of microplastics studies assessing plastic in seafood. Also, it is likely that smaller particles are easily bioaccumulated by bivalves.

Would be very interesting!

Thank you for your explonations :) No problem! Thanks :)

Stephan Rohrbach to Marina: Interesting results! I am not a physician, but I thought that highlyunsaturated fatty acids are healthier than saturated, so why you conclude that the fatty acid quality decreases? Ok sorry maybe I misunderstood on your slide ok ;-)

## Charlene Trestrail

Question: Sorry if I missed it, but do you think these changes have any effect on the overall health of the bivalves?

I don't know if I answered. Please, if not, my email is azulmarinita@gmail.com Yes, we have results on bovalves health, and physiological status.

You mention this in your conclusions - Did you measure the oxidative stress in the anemones? Thanks :)

Georgie for Anna: did you quantify the ingestion in those anemone at all, following their exposures ?

# **Questions and comments for the session 25.3\_Me**

Name: Giorgia C.

QUestion: Do you think a potential regulation on microplastics should target this issue focusing on environmental risks or rather risks for human health? In your opinion, which approach would be more efficient in the perception of the public? Thank you fot the interesting inputs.

## Name:..Nevena H.

Question 1:...The intentionally added microplastics to personal care products are decreasing amount in Europe. The question is how to deal with the microplastics produced from reusing and recycling plastics in the food chain? Was more a question of what regulation on the packaging and production of food you need to create to reduce/remove microplastics that come in through the food chain. Should all chemical types of MP be regulated the same way?

## Name:...Carla Elliff

Question 2:...Thank you all for your presentations! Considering that microplastics are a transdisciplinary issue and that they affect multiple UN Sustainable Development Goals, I would like to know your views about using the SDGs framework to act upon microplastic pollution.

Ryan Shum: Thanks so much for all the presentations. This question is a follow-up on Sarah's response, I was wondering when you suggest that we need to rethink our practices (not suggesting that this is not important) doesn't that once again reinforce the individualisation of responsibility you spoke about which we need to move beyond?

To Greet (and thimc): What would be sufficient regulatory approaches to tackle the problem of flushing items like non-woven fabrics, sanitary towls / pads / tampons etc. down toilets, in your opinion? Do you think extended producer responsibility schemes would work with these particular products? But how would you measure if labelling works?

### Name: Cleo:

Question/ comment:

Very intersting discussion, thank you all. I would like to add, concerning the topic of women and motherhood, I think we should not get too much into men and women, but to start sharing responsability, strenghtening parenthood. And we should always include cultural, regional, political differences and conditions and that we need to be so careful when saying, for example: "mothers use plastic products because it is more confortable or practical, because sometimes available alternatives are missing as well. For example, I travelled in Panama and in the supermarkets you often had limited choices, big companies dominate the choices to be sold. There is, hence, a big problem, that is complex. and again, we need to share responsability. And we have to use alternatives if we have them, using our "power" as users and pressurising producers.

So... what do you think, policies could do right now to create improvements, what are tools?

### Name: Julia Taylor

Question: Very interesting, thank you! A problem that we are facing is that we don't have any clear answers on the risks of MP. Therefore we find it hard to regulate it. Any thoughts on this?

### Name: Susanne Belz

Q: Do you think there is relevant microplastic test material available for exposure and tox testing? If not, what is needed?

Katrin Wendt-Potthoff: the material you can buy is often not relevant for environmental settings, and the testing itself is problematic (suspension, homogeneous exposure,...)

### Name: Andrea

Question: Taking a more environmental perspective, lots of presenters have been asked how realistic their research is when using so called "virgin" MP in toxicity studies. How can we make plastics

available which have been through weathering, degradation etc. for researchers to study the effects of REAL microplastics/plastics as opposed to virgin analogs? Indeed! Who could provide such 'real' material or at least material that resembles reality?

I see an opportunity here to connect to beach clean-ups and local communities who can provide a large volume of material in only a couple hours of cleaning for example. This material could be further handled to create MP from macroplastics. Yes, why not? Good idea. (I am currently doing this and its hard to find the amount of material I need when I am alone! ) Connecting with NGOs or other such groups could facilitate a chain to research and simultaneously educate the public around these issues.

Well, I can tell you that in Brussels - the heart of the European legislators, I can only recycle plastic bottles and very hard plastics that go into dump yards (e.g. toys, storage containers etc). ALL THE OTHER plastic food packaging goes in the same bin as my regular waste and that's A LOT.

# Questions and comments for the session 25.3\_Ma

Richard Cross UKCEH riccro@ceh.ac.uk Lisbet Sørensen: lisbet.sorensen@sintef.no

Mateo - the sound is really poor on this presentation I have a problem with the sound too, it is really fast in some parts

Seems to have 'settled' a bit now. It is too speedy!

We had the same problems in previous sessions with Mateo....:-(

Here it works regularly!

Matt Cole: Hello to my old desk neighbour! You said polymer needs to be accounted for in blank correction, but does shape also need to be considered in same way?

Rich Cross: Hi Mat! Old desk buddy :) just for prosperity, yes the polymer type needs to be accounted for in the blank correction (if you don't find a polymer in your blanks, no need to correct for in your samples), but this blank must be representative of your entire processing workflow so any introduction of contamination in your samples would be matched in your blanks. I don't believe shape would need to be considered in the same way except if in conjunction with polymer. For example, if you saw PET fibres and PET fragments in your blanks and in your samples, it would make sense to apply the correction to each individually, not to correct for PET overall.

Anna Winkler: What do you mean by separating polymers in blanks? Shall I not subtract the microplastic particle number from samples ("blank corrected") if the polymer in the blank is another polymer/not represented in my sample? Can you explain this argumentation better? Okay thank you!

Rich cross: thanks for the question Anna, as I've mentioned to Mat above, blank correction on a polymer by polymer basis is essential. Yes it absolutely makes sense... So, just to clarify: If I have, for example, only one microplastic (a PET fibre) in my blank, but no PET fibre (or fragment) in my

sample (only few microplastics in general), I should NOT subtract the 1 number of the total number of microplastics in my sample just because cannot assign them to the polymer found in my blank? Great talk, as always, btw.

Name:...Irina Pucić, Croatia

Question 1:...for Sørensen Lisbet what about viscose fibrers, also synthetic polymer? Why di you choose wool that contains keratin, slik would be better, pure protein TNX!

Question for Lisbeth from Giuseppe Suaria: very interesting talk, was just wondering if you are planning to perform similar experiments with cellulosics fibers such as cotton or rayon since they are often the most common fibers in the natural environment?

Name: Nora Meides, University of Bayreuth

Question: Very interesting talk! Did you determine fragmentation rates or were you able to quantify the amount of fragments produced from one fiber? And did you do the same experiments without seawater, so DI water or no water at all? Thank you!

Name Boris

Q: hi Lisbeth , thank you for the nice talk . what kind of device you use for uv irradiation what is the range of wavelength ?

Name: Kristina Klein

Question: Great talk, thank you! Based on the chemicals from wool fibers, what would you recommend to use as a natural, rather chemical-free fibre in the future?

Name: Alimi Olubukola

Question: Great presentation Sorensen. What type of UV lights was used for the exposure? Wavelength?

Name:... Sam Athey

Question 2:... I had trouble following because of the connection. Could you summarize the conclusions regarding recovery?

Rich Cross: Yes, we took a worst case approach to recoveries, using polyamide as a worst case polymer based on its higher density than other polymers we detected. It will also be important to also understand recoveries not only based on the recovery in the sample, but also recoveries towards the analytical limits of your detection methods. Thanks Rich! Great talk :)

Name: Erika Cedillo

Question: Dear Sørensen Lisbet, which analytical technique did you use to measure the substances present in the leachates?

Name:... Rich Cross

Question 2:. Did you say that all fibres natural and synthetic all degraded to similar sizes fragments? Do we know anything about the rigidity of these smaller fragments and if there is any proposed mechanism of physical effects based on high aspect ratio, rigid fibres

Name: Bethanie Carney Almroth @ UGOT, Sweden

Question for Lisbeth: Thanks for some really nice work! Did you do analyses of the chemicals in the animals following fiber ingestion?

Name: Barbora Pinlova Question: where did you source your fibers for the research? Are they cut yarn?

# **Questions and comments for the session 25.4\_O**

Name:Marta Barone

Question 1: Dear Macarena, thank you for the presentation. Is it assessed weather this method is applicable for WWTPs (larger scale than experimental assessment)? Would it be financially advantageous for WWTPs? How would the separated fallout be disposed of?

Name:...Christine Knauss

Question 2:...Interesting work! Sorry I missed the beginning because of technical issues so you might have addressed this. Is the adhesion of minerals specific to plastic particles or will they adhere to particulate organic matter and other minerals?

Vesna Teofilovic Did you get any response from government?

# Questions and comments for the session 25.4\_Me

## QUESTIONS TO POSTER 3 (Microplastic pollution in the Arctic seas; Irina Makeeva)

Name:...Nico M.

Question 1:...Nice poster, thank you! From what depth was the water pumped and how much water were you able to pump along the whole cruise track with the shipboard pump? How did you make sure, that you don't sample for example the waste water from the ship?

# Questions and comments for the session 25.4\_Ma

Ana Catarino: is there a problem with the streaming? Stjepan: I can't see it either Name:...Stjepan Budimir Question 1:...Regarding PLUXIN poster: have you been looking into floating macroplastic, if yes which methods did you use, and if not do you plan to do it?

Name:..Louise Schreyers Question 2:..Can you tell us more about the detection with Remote Sensing and hyperspectral information that you plan to use?

Super interesting! We will also be doing something similar at WUR (Wageningen University).

# Questions and comments for the session 25.5\_O

There is no sound maximilian Maybe you have to unplug your earphones We cant hear anything

For further questions you can also contact me via: eike.esders@uni-bayreuth.de

Name:Magdalena Mrokowska Question 1: How did you calculated mean velocity of the air flow? Have you done it using 3D velocity data? Answer: Yes, I calculated the mean horizontal velocity from the 3D data.

Name:...Melissa

Question 2:...Could the polydispersity of the samples you used could have contributed to the variance of the percentage moved at the different wind speeds?

Answer: Hello Melissa, sorry I got your question wrong in the talk. The sizes of the particles definetly plays a role, if you think about a minimum force needed to suspend them, or move them. The range of particle diameters I used so far was 53-63µm. The size range between these two sizes is normally distributed in our samples. The size histograms do not show that for example particles with bigger diameters are preferentially suspended.

Name: Reza Shiravani Question:

Great work! Thank you. Did you find a relationship between flow eddy and particle diffusivity?

What were the Reynolds number range during the experiments?

Answer: Hello Reza, the Reynolds numbers in the experiments ranged between 54000 - 160000. If you like I can send you more details via mail. Just send me an email.

Name: Trang Nguyen

Question: How you can be sure that the particles were plastic because in other studies they showed that not only plastic material 'reflect' fluorescent light

Answer: So in the experiments only fluorescent particles were used. They were placed on a glass plate to be analysed. We didn't use samples from the environment.

Name: Johanna Sonnenberg

Question: Is this transferable to behaviour in the water, e.g. on the seabed? Answer: Hello Johanna, if you like send me an email and I can probably make a contact to the PhD Student working on this. Also here is a link to the projects worked on in the CRC 1357: https://www.sfb-mikroplastik.uni-bayreuth.de/en/projects/b-projects/index.html take a special look at B02

Moritz Lehmann, SFB B04: Are particles suspended in air with turbulent flow with mean velocity less than the vertical laminar flow velocity required to keep particles floating? In other words, is turbulence beneficial for particle suspension? Hello Moritz, turbulence is kind of necessary to suspend the particles. If you like we can talk in more detail later.

Questions to Andrea: further Q can be sent to andrea.faltynkova@ntnu.no

Name: Andreas Eich Question: How does biofilm on the plastic surface affect the identification?

Name: Stephan Rohrbach to Andrea:

Is it possible to automate the analysis of your hyperspectral imaging using ImageJ /FIJI software?. Thank youw

Name : Thomas Witzmann to Andrea How do you want to tackle the issue with the resolution gap between  $\mu$ HIS and HIS? Thanks for he nice talk!

Name: Matthias Egger Question: Thanks for this exciting talk, Andrea! Do you think HIS could be a suitable tool to study the ageing of plastics too?

Questions to Irina: Name: Juliana Ivar do Sul Hello Francois, good to see you here! I am wondering if she trying to do the same correlations but with fragments relatively lower and relatively higher polymer types.

and did you check whether deep regions have more plastics in relation to coastal regions? maybe these fibers are more common in deep regions, like the anoxic basins

Maurits Halbach Have you correlated against TOC? total organic content.

Questions to Samuel: Hamsun Chan How do you think the result of your study will be different if treatment is done with a more urban/contaminated water source?

# **Questions and comments for the session 25.5\_Me**

Name:... Peter Vermeiren Question 2:...Thanks, Did you also compare the gut content, with natural materials, to see if microplastics reduce the ingestion of natural materials (p.vermeiren@science.ru.nl)

Name:...Ester Carreras Question 3:..Great talk, and very nice presentation :) Do you think the formation of tangles could act as a trigger for moult?

Miriam/HYDRA: what kind of stakeholder envolment would be in action during the project?

Great, when is the first occation to engange with the project? Hi Miriam, thanks for the question, here is the link to our newsletter sign up: https://email.ilvo.vlaanderen.be/h/t/9D19DD88BE32F0B1

# Questions and comments for the session 25.5\_Ma

1. Carlie Herring - Overview of the NOAA marine debris program's funded research

carlie.herring@noaa.gov

https://marinedebris.noaa.gov/

https://marinedebris.noaa.gov/current-efforts/research

Name: Erik van Sebille

Question 1: How do you think that the NOAA funding strategies and priorities in marine plastic differ from those in e.g. EU or other regions of the world? What do you think has worked particularly well in the NOAA funding strategy; and what could have been done better, compared to other schemes?

Carlie = We have also recently released our new strategic plan (2021 - 2025), you can see our priorities and focal areas in the new plan here: https://marinedebris.noaa.gov/who-we-are/2021-2025-strategic-plan

Name:.Tim van Emmerik

Question 2: Most projects you discussed focused on microplastics. Are you also supporting any projects focused on the effects/risks/quantities of macroplastics?

Name: Christine Knauss

Question: From one of your slides, it is clear that interest, proposals, and funding has increased over the years. Do you expect that to continue for the Marine Debris Program?

Carlie = Hi Christine, I missed the 'funding' component of your question earlier. Funding is dependent on appropriated funds to our program. It is hard to predict what this might look like in the future.

2. Romain Tramoy - Endless journey of macroplastics in rivers

Name: Tim v Emmerik

Question 1: Really cool work, Romain. DO you think we may be considerably overestimating the plastic emissions from rivers into the ocean (by models)?

Name:...Stjepan Budimir

Question 2:...great work! Would this aprach be needed when considering river clean up efforts and "litter interception" Actions

Johnny Gasperi = This kind of approcah can be very usefull to identy litter accumulation zones and then to launch cleaning operation

Any suggestions for others who want to use GPS trackers for studying macroplastic transport in other rivers? (TvE)

Name: Simone Lechthaler

Question: Really great work! I might have missed it but was the bottle that was tracked closed or open? If it was closed, maybe the air inclusion could have influenced the transport? Johnny Gasperi : bottles are of course closed, but the GPS is in the upper part of the bottle to communicate

Romain Tramoy: We used half submerged bottles filled with sand and floating bottles full of air to get neutral and high bouyancy. Bottles were closed to keep the GPS inside and always up the water surface in order to enable telecommunication

Name: Erik van Sebille

Really cool talk! How many GPS trackers do you at least need for statistically significant results, in your study?

Johnny Gasperi : It's depend on the objectives. To study dynamic, a few are required. For statitsic and fluxes, more are required

And how do you get authorities to accept you putting plastic into the river?

JG : collaboration with local authories which help to discharges" litter". Note plese taht these litter were prevsouly collected along the Seine river banks and we do not add additionnal pollution :\_) Romain Tramoy. The marked litter released were in complement with the GPS tracker. The whole experiementation was set to quantify mass flows of plastics from the Seine River to the Sea based on daily cleaning of riverbanks (to get a probability of collection). This is the coming paper, waiting for reviewers since July :)

Name: Sarah Nelms Great talk! Is this published yet? Johnny : Yes, https://www.sciencedirect.com/science/article/abs/pii/S0025326X20306846 Thanks! :)

3. Amaia Mendoza - DISTRIBUTION, COMPOSITION AND ABUNDANCE OF MICROPLASTICS IN THE BAY OF BISCAY amendoza001@ehu.eus

Name:C. Maes Question 1 Any ideas about the main sources if you want to model the observed dispersion?

Name: Erik van Sebille

Question 2: Nice talk and impressive collection of data from different sources in the review article! Do you also plan to use these data in an integrated model approach to 'interpolate between your data points'?

4. Maria Kazour - Effect of exposure duration on caged blue mussels (Mytilus edulis) microplastics bioaccumulation

maria.kazour@univ-littoral.fr

## Name:...Thibault MASSET

Question 1:...How did you deal with the influence of tidal coefficient on the plastic concentration in water ? Your bioaccumulation test was therefore performed in no homogenous envrironments in terms of concentration of MPs. It could have had an impact on the bioaccumulated MPs ?

## Name:... Kevin Tallec

Question 2:... Do you have an idea to explain why the majority of MP are higher than 200  $\mu$ m in water while in mussel the majority of MP are smaller then 200  $\mu$ m?

Name: Camille Détrée Question3: How do you explain the very low abundance of microfibers while an increasing ammount of study is showing that microfibers abundance in the environement is higher than Mp fragments?

NAME: Marie Russell Question: did you depurate mussels after exposure in the cages

# Questions and comments for the session 25.6\_O

Name:...Isabel Goßmann

Question 1:...Sorry I missed the beginning of the talk due to technical issues. Did you metion what the road markings were mostly made of? How did you characterize the road markings? Thank you! Sorry my internet connection is bad at the moment.

## Name:...MIRIAM/HYDRA

Question 2:...DId you already consider to use paint which is biodegradable in soil next to roads and in freshwater ways nearby?

Thank you for the update. Please let us know if you like to go further into that.

Question to Rachel:

Name: Elena Hengstmann

Question: First of all, very interesting and nice talk! Do you have an explanation for the decrease of microplastic abundance in the urban river within your sampling period?

Ok, thank you for this explanation. I'm curios whether you actually see this maximum during snow melt which you expect. Sounds reasonable!

Tim van Emmerik

Interesting and worrying results! You mentioned the change in taxonomic richness as potential early warning for ecosystem change. Is this warning early enough to reverse the change? (i'm a hydrologists, so no clue about this :) ) And what interventions would you suggest?

## E. Rødland:

very nice talk. You did not confirm the tire rubbers with any analytical method, is that correct? Are you planning on doing this?

Christine Knauss

Thank you for a great talk! Did you sample in the same stream just up stream and down stream or different streams?

Sarah: Was the morphology of the streams the same between the sampling sites? Thanks!

Winnie: You already answered- Thanks!

Question for Decio(?)

Tim: Muito obrigado! Did you also find and quantify plastics larger than 5 mm? Do you think Van Veen grabbers are the right tool for studying abundance of larger items in river sediments? If no, what would suggest as an alternative?

Bruno : very cool presentation ! thank you . As the Suspended solids concentration is very different in Rio Negro and Solimoes, do you have an idea of the difference of concentration of microplastics also in the water columm in both parts of the river ?

# Questions and comments for the session 25.6\_Me

Name:...Gholamreza Shiravani

Question 1:...Great work. How the saliniy of water body could increse/decrease the zeta-potential value? As an example the salinity of 12 PSU in comparison to 33 PSU? Could the salinity increase the aggregation rate?

Name:...Yuanhu Zhang Question 2:...Nice talk! did you observe PS aggregation which is not in the repellion region. How about the NPs stabillity when Zeta potential is bigger than -20 mV?

Name... Gerardo Pulido-Reyes Question3... Really nice talk! Thanks. I am quite interested in the size of this complex that can be formed between the nanoplastics and soil component. Would you have any estimation? Thanks again!

To Rachel:

Name: Cleo Question 1: Really nice study and talk! And nice study area I imagine! What was your reason to choose 1cm top layer? thank you

Name: Romain Tramoy Nice talk, thank you. Why choosing report result in Mps/100Ml and not in Mps/g of sediment?

Name: Hamsun Chan Question: how do you account for sample lost that is not released from the density separation? Name: Stephan Rohrbach: Nice talk. As far as I know, the california bay is prone to seismic activities. Any ideas how lets say earthquakes might affect your studies?

Name: Julia Möller Great Talk, thank you! Which density of Zinc chloride did you use? And sorry if I missed it, but how did you determine the plastic types that you saw? Hi Julia. 1.5g/cm^3 and we determined the plastic morphotypes by microscopic point count. -Rachel

Hi Julia. 1.5g/cm<sup>3</sup> and we determined the plastic morphotypes by microscopic point count. -Rachel

Name: Hamsun Chan Question: How does the sinking rate of MP aggregate compares to typical marine snow? Hi there, Hamsun. This was the paper I was referring to: Role of Marine Snows in Microplastic Fate and Bioavailability Adam Porter,† Brett P. Lyons,‡ Tamara S. Galloway,† and Ceri Lewis\*,† Which explains the change of MP sinking rates. -Rachel

Question for Gunhild Name: Julia Möller How do you explain the higher abundance of microbes in soils contaminated with plastics compared to the surface of the plastics themselves?

Astrid Delorme: Thank you for your talk! When you collected samples at a depth of 20 cm, did you find plastic debris at this depth too? if you did, did you measure the concentration of plastic debris in some way?

Hi Astrid, Thanks! Yes, we found plastic at all depths, but is was difficult to pinpoint the exact depth because the soil was so porous and loose. We will measure microplastic and estimate volume of larger plastics.

name : Jafar , Hungary thanks for nice ppt, did you check the plastic particles for any signs of degradation? Hi Jafar, thanks! No, we did not focus on the degradation in this study, but would like to include that also at a later stage. thank you, good luck

Johnny Gasperi. Very nice talk. Do you also investigate the organic pollutant pattern in soil with plastic ? Maybe, you plan to analyse those patterns.

Name: Vincenzo Donnarumma Question for Gunhild: Which DNA extraction kit have you used for both plastics and soil?

Name Stephan Rohrbach: Splendid talk. Have you checked for biofilm formation on the plastic debris which are abundant in the contaminated soils?

Idea: You can try to get plastic waste which is just about to enter the environment by throwing away and check if you bring some alien species into the habitat.

# Questions and comments for the session 25.6\_Ma

Orior Rodriguez-Romeu e-mail adress: oriol.rodriguez@uab.cat Name:...Judith Weis Question 1:...Comment for Dr.DeHaan - by collecting your samples with a Manta net you are losing many or most of the microfibers.

Name: Camille Richon (Question for Oriol) Question 2: thank you for your talk! Do you know at what depth range do anchovy feed? Could this explain the type of plastics they ingest? Thanks! That was super interesting :) Thanks Camille!

#### Judith Weis - for Oriol

It is interesting that most field studies are like yours on anchovies (f- no correlation with indices of health. In lab studies people us much higher concentrations of MPs than are found naturally, and find all sorts of effects - but this is not realistic. Thanks Judith for your coments!

#### Name: Alejandro Usategui (for Patricia)

Question: It has been a very interesting presentation! in relation with Thr plastics, appart from contamination, what do you think is the origin of this type of plastics?

Thanks Alejandro!

### Patricia Ostiategui mail adress: patriciaostiategui@gmail.com

Name: Liam (for Patricia)

Question: Nike talk! Apart from fishing lines/filaments, have you found any artificial turfs/artificial grass, or a preference of ingestion for green coloured plastics? Thanks a loT As I said, we didnt analyse by colour yet, anyway loggerhead turtles are oportunistic and onminovorus animals. It would be great to comare with green turtles, but those strandings are much less normal in Canary Islands.

### Name: Joao Frias (For Patricia)

Question: Brilliant talk! Are the differences in types and quantities between live and dead animals over time? Meaning, to alive animals today have more lines (Thr) than historical data for the region? Thanks for this! Thanks! We only have been sampling alive animals for the lasts years, so sadly, we can not compare

name: alicia mateos cardena

Q: nice and very clear talk! Thanks - you see 100% frequency on the turtles, do you have any idea of what impacts the plastics could have on them if any?

# name: Clara

Nice talk Patricia, thank you! How did you exactly take the fecal samples from the turtles in the tank? Also, did you take blank samples of the water and possibly also of sedimented particles in the tank? Tnak you Clara. We are still taking more blank samples, trying to do at the same time than the sampling, in order to have a better aproach of the contamination.

### Cynthia Munoz

Thanks for the presentation. Do you have an idea of how much is egested via faeces relative to the total amount still in the gut. Did you consider doing a stomach flush. Stomach flush might be very

difficult due to the turtle esophagus (and the last thing you need is to cause more pain to alive animals). Thanks (feel free to discuss further c.munoz@science.ru.nl)

Name: Joao Frias (For Liam)

Question: Excellent talk. You mentioned that concentrations increased over time. Is there any reason for this? What is your theory? Also, do you also have results of no. MPs per volume (m3) to be able to compare with other coastal sites/bays? Thank you!

Name: Matthieu Mercier (for Liam)

Question: The characterization is very complete, can you tell us how do you account for the sampling context (sea-state and seasonnality, corrected concentrations)?

# **Questions and comments for the session 25.7\_O**

Questions for Kirstie Jones-Williams:

Name:...Darshika Manral, Utrecht University

Question 1:...Wonderful presentation Kirstie and animations! I might have missed but could you mention if you were able to identify the sources and types (fragment / pellet etc.) of these microplastics? Partly covered in your answer to Andy. Other sources?

Kirstie: I hope I was able to answer your question but happy to discuss further.: kirnes79@bas.ac.uk

Name:...Gissell Lacerot

Question 2:...Not so much a question, but a comment. We also found a lot a paint remains in water samples from Fildes Peninsula, Antártica, both in coastal samples (sampled from a zodiac boat) and freshwater samples from ice streams discharging on the coast . Not only ships but also scientific bases we think, in our case, also wind effect. Great presentation!. Thank you!

Kirstie: THis is really interesting Gissell - I'd love to discuss more with you about your methods for analysing these? Myemail is Kirnes79@bas.ac.uk

Sure!. We are mainly analyzing with polarized microscopy, but we have recently also started to use microftir, although not for everything. We do an integrated sampling of macro and microplastics in Fildes peninsula, coast, streams, ice, marine water, organisms. We have a recent short communication on wind effect, I'll email it to you. Would be great to be in touch.

Kirstie: Please do :)!

Dan Wilson: Really interesting presentation! Where on the ship did you find/do you think the contamination was coming from? Ship hull or deck for example? Also the ship footprint sounds like a really interesting research question!

Kirstie: Thank you. It appeared that most was coming from the ship hull. I took samples from the hull and also random samples from across the deck, where you can see that coats of old paint flakes have collected. I was on a recent expedition where they were repainting parts of the ship whilst we were anchored off the coast so I think there is a lot of potential for pollution there too.

Thanks for the extra info! Ahh that's interesting, hmm yes it does seem like significant potential for paint flakes to be deposited into the Southern Ocean whilst repainting is going on! What size were the paint flakes roughly, or were there quite a variety of sizes?

Questions for Louise Schreyers :

Name:...Rachel Giles Question 1:...Do you have an idea of why the entanglement varies? Is it because there is less water hyacinth, or because of other factors like a rainfall? Ok cool! so probably the plants :)

Name:...Rachel Giles (if someone else wants to ask, they can :)) Question 2:...Foam is a very common - you mention that foam was very common in the water

hyacinth..did you see that there was more foam captured by the plants relative to other free floating debris, or that it was the most common item found in the water hyacinth. great. thanks for clarifying. :)

Questions for Katie Reilly:

Name:...Kristina Klein

Question 1:...Thanks for the talk! What do mean exactly by commercial formulation? Did you wash the particles prior to dispersing them in medium/river water? me too, thanks ;-)

Christine Knauss

Really interesting talk thanks! What concentrations of tween were you using when you saw toxicity? I will email you thanks!

Name:...Anja

Question 2:...Which staining technique (which heavy metals for instance) did you use to stain the corona on the particles for TEM imaging?

Questions for Anna Diem: Name:...Sonja Ehlers Question 1:...Thank you very much for your really interesting talk! Have you considered to also expose the predatory crabs to microplastics? Thank you, Anna.

Name:...Miriam/HYDRA Question 2:...Great study, thanks for that! Would the crabs also feed on meiofauna and so grab MP from between sand grains too? I wonder if the crabs could have taken up MP elsewehre too.

# Questions and comments for the session 25.7\_Me

Name:.Lisa Zimmermann No question but a wish to Susanne Kühn: Unfortunately, I won't make it to your talk today. Any chance that you can share it with me? (l.zimmermann@bio.uni-frankfurt.de) Thanks a lot! Susanne: Yes sure. I'll send it to you!

Nombre : Ana Liria Loza (To Marga) Pregunta 2: The review is related to macro and microlitter, or only to microlitter? There are different methodologies? The studies are on stranded animals? Thank you very much. Nice work!!!!!

Name: Eliana McCann Smith

\_MICRO 2020. Fate and Impact of Microplastics: Knowledge and Responsibilities. 23-27 November 2020\_\_\_\_

Question: Did you also look at the relationship between MP and fatty acids and find no significance? Thank you!

Christine Knauss

How do you think your method for milling your plastics and increasing the surface area influence the leaching of additives? Were your microplastics similar to the sizes found in sea birds? Great thanks!

Francesco Saliu

question for elvis.. your sample are pretty plastic contaminated, high concentration. But what about low plastic concentration. How do you calibrate pyrolisis to reach low detection limit.. thank you :D

# Questions and comments for the session 25.7\_Ma

## SPEAKER 1: JOSEF BRANDT

Name: Garth Covernton Question 1: Thanks for this! What background media are you placing the particles on? Do you need a reference point? How do you make sure that particles aren't moving around between different machines when moving the filter?

SPEAKER 2: BRUNA RAMOS

No questions from the audience for you today Bruna.

SPEAKER 3: MARIA GARCIA PIMENTEL

Name:...Rachel Sarner Question 1:...Thank you María for your presentation! This was very fascinating. Was there a reason you chose to sample in the winter?

## SPEAKER 4: ERIKA CEDILLO-GONZALEZ

Name: Coco Cheung Question 1: Interesting presentation! How do you evaluate if complete oxidation of plastic into CO2 and H2O occurs, or if it stops at the stage of fragmentation to nanoplastic? Thanks a lot!

Thanks and one more question, would you also consider testing the reaction of biodegradable plastics? Wondering if the oxidation could assist further biodegradation..

Thanks erika for your talk! I'm working on bioplastics and would be happy to keep in touch with you regarding the facilitation of biodegradation of bioplastics by your technology. Here's my email address: kahei62000@gmail.com. Thanks again!

Name:...Nicolas Keller Question 2:....Nice talk, did you evaluate possible retention of microbes at the surface of the film or within the film porosity ?

# **Questions for The Marine Strategy Framework Directive perspective panel**

### Name: Kathleen Michels

Question: granulated tire waste infill and infilled plastic synthetic-turf carpeting have been of concern as a source of macro and microplastic and chemical and heavy metal Pollution. Is there any discussion on regulations and further policy and research?

Name:Niels Mast Question: Apparently MSFD micro at the moment restricts to a size range 100 micro – 5mm. Any outlook that nanoscale will be included in later stage? Great reply! Thanx!

### Name: Frédérique Mongodin

Question: how is the European Commission working towards a coherent legislative framework, including at the international level, to address microplastic pollution? And will microplastics be part of the negotiations towards a global treaty on plastics?

### Name: Catharina Pieper

Question: When considering definitive protocols to work on MPs, will you consider different approaches for labs that don't have state-of-the art equipment available? This means, offer alternatives in terms of protocols that are still able to match some of the MSFD pre-requisites (e.g. comparability, standardization). Also, the creation of a network of collaborations between labs would be interesting. Thank you!

### Name: Marie Russell

Question:How do you see MSFD/ICES/OSPAR and also regional seas conventions working together to produce internationally recognised methods and protocols, including the very necessary QA/QC

### Name:Miriam/HYDRA

Question:Thank you for the nice overview and time for discussion. We do a lot of fieldwork seeing the constant input and fearing the tremendous increase in the future. That makes me nervous that we have to wait, seems to me, quite some years to get harmonized actions and implementations. I want to provoke "Can we live with 'good' and not aim for 'perfect'?" and understand what are the possibilites from the policy side that we scientists can accept 'good' asap? Can you explain tools you apply if science is not as fast as policy?

#### Name: Peter Vermeiren

Question: How much attention is being given to modelling approaches as tools to 1) convert among data collected using different methods, and 2) extrapolate beyond current monitored datapoints. Is modelling considered as a basis for informing policy in addition to monitoring data (e.g. as in pesticide regulation).

### Name: Julia Taylor

Q: How is your work harmonized with the work going on in CEN and ISO? There is for example a vocabulary standard being developed (published by the end of the year) for microplastics within CEN, which will be harmonized with ISO. Will this change anything for you and will you start using the same definition as CEN?

## Name: Careen Krüger

Question to Georg Hanke: Can you justify why policy is faster than science in this matter? What is the evidence of this? what do you expect from the researchers to move faster in future? Thank you!

## Name: Cleo

Question: 1) How can young researchers help to contribute good data to the MSFD in their studies? 2) How, if you might, would you connect or link the marine litter abundance with input from freshwater ecosystems, so how does come freshwater microlitter research possibly into play?

## Name: Juliana Ivar do Sul

1) I hear often researchers interest in going deep into complicated methods to estimate microplastic particles in the environment because they think its not so relevant to have data on meso and macroplastics. I try to focus more on the research question than on the relative importance of micro (in relation to other size categories). any thoughts on that issue? I believe all have their importance, once the larger ones are significant sources of the smaller ones. and once results can be more reliable in relatively larger sizes (>500 Micrometers) when methods are missing e.g. confirmation of plastic/polymer identity. Thank you.

2) maybe relevant in this Forum, how do you think we have evolved in our plastic research from 10 years ago to now? are we in the way to have a permanent research field?

## Name: Ana Markic

Question: Would funding by the EC and JRC be available for private consultancies with expertise to execute data collection, or solely for research institutions and academia? Also, would this kind of data be accepted if collected independently/privately on one's own expense? I hold a PhD in marine science with plastic pollution as my topic, but I am not associated with any institution, but my nonprofit and consultancy. Thank you.

Name: Niels Mast Question: Re. harmonizing efforts: what are the thoughts on units (#count, weight, concentration, volume) ?

Ryan Shum: Is there scope for citizen science to contribute towards the collection of data?

Frederique Mongodin: totally agree with Gunnar on the added value and potential increase in contribution from citizen science, this is valuable and has to be further encouraged and be used as a complementary dataset. Thanks for the very interesting exchange this evening!

Amy Lusher: Thank you all for a fantastic discussion. Really looking forward to the future of plastics research.

Ignacio de Sobrino: Looking at the presentations, it seems that almost all of the studies are focusing on the distribution of MPs and the effects that those have on the organisms. There shouldn't be much more investigation and funding to find out ways to eliminate the plastic? For example, through microbial digestion.

# Questions and comments for the session 25.8\_O

Sascha Müller: Cool Poster! Your figure showing the size, pd and zetapotentila. you show a wide range of potetnials. You adress it to different surfactants. What type of surfactants you have so far? The zeta potential depends on the pH, so if for example alkali is added to a sample, the particles tend to acquire more negative charge and the zeta potential becomes more negative.

# Questions and comments for the session 25.8\_Me

Name:...Rachel Sarner Question 1:...Thank you for the really interesting presentation! Any theories on what the source of plastic contamination might be? Wow, thanks for the great answer!

### Name:...Giuseppe Suaria

Question 2:...Hi Liliya! great talk! What about settling rates of fibers? Can you give us any hints? Hello Giuseppe! )) The hint is ~1mm/s, which is actually quite big comparing to the rates of suspended matter and some other natural particles. Hopefully I will end the paper about this soon ;) Thanks Liliya! looking really forward to read it! the big mistery now is why the fibers are not sinking down despite being mostly denser than seawater!

Hey Sebastian (Stephan Rohrbach C04) have you read the recent publication from tournier et al who also tried to bioengineer a PETase. Are they using similar algorhithms? Hey Stephan, yes indeed I'm familiar with their work. They were testing individual mutations or pairs of mutations which is somehow similar to the GRAPE algorithm. This algorithm would then suggest which of your tested mutations you could try to combine in strategically useful way. Ok, well I was pretty sure you already know the paper. Just wasn't sure if you can use it somehow.

# Questions and comments for the session 25.8\_Ma

### Name: Tenzin (question to Lorenz)

Question 1:...How representative the sub sample of the lower size analyses would be? Is the sub sample concentration later converted to represent the whole sample of lower size? @Tenzin: The subsample volume represented 3-15% of the total sample and was then extrapolated to the 1m3 sampled. The sample preparation and results are presented more in detail in the report which can be found here: https://www.miljodirektoratet.no/globalassets/publikasjoner/m1572/m1572.pdf. We also have more samples from this study where we will analyze 20-50% of the sample to increase representativeness which we can hopefully publish next year :) Thank you also for your interesting presentation.

## Questions and comments for the session 25.9\_O

Question to Larissa: Name: Clara Marnie Q: You mentioned an increased microplastic abundance at one your sampled spots. Do you have an explanation for this increased concentration? Name:...Francesco Saliu

Question 1:...Yoshio thank you!! provide also data with solar box conditions, I think that it will works when there will be the full picture of all the possible aging experiment. I undestood it was only accelereted in oven, ok thank you

Name:..Hannah De Frond.

Question 2:...Thanks Yoshio for your presentation. Did you compare the effectiveness of your polymer library to any pre-installed polymer libraries? Did degrading the polymers lead to improved HQI scores using these spectra?

No questions for the Session 25.9\_Me

# No questions for the Session 25.9\_Ma

# Questions and comments for the session 25.10\_O

Question for Victoria Name:...Rachel Giles Question 1:...Have you thought about modelling the sediment fate, instead of or as well as surface water fate? How do you expect that the estimates from a sediment model will differ from the estimates in the surface water fate model? Great. Thanks! Can't wait to see the final model. :)

Name:...Simona Mondellini Question to Victoria:...did you expect to be any seasonal change maybe related to the use of winter tires?

To Victoria do you know wich type of shape tyre wear particles could be classified wich type of class do you use?

Question for Irina From Rachel Sarner: Can you clarify if the sea ice is always frozen? Or does it melt and then refreeze? Never mind! She answered my question :) Thank you! :)

Question for Marco

Rachel Sarner: This is a very interesting presentation, thank you! I am not sure if I missed it, but are there any theories on why Miserin had a much greater amount of MPs? Thank you!

For Marco: have you found any cellulosic fibers in your samples?

from francesco saliu for irina : just curiosity did someone collect ice sea samples from okhotsk sea area? Thank you for presentation super interesting :) ok thank you

Thank you! -Rachel

# Questions and comments for the session 25.10\_Me

### Name:... Bethany Jorgensen

Comment 1: For Anna -- Just to say, congratulations for this work; really helpful to illuminate gaps in studies of mps in freshwater ecosystems, and suggestions for moving forward ! Thank you! There is definitely need for unifying our approach. Also we need more data around the world!

### Name:...Gissell Lacerot (for Anna)

Question 2:...In the recommendations for lab studies you mention food provision. What do you mean by that? To combine with natural food? Thank you for your presentation, it's great to have studies that summarize current findings.

Hi Gissell, thank you and sorry I did not have time to go over the laboratory recomendations. So for food provision: sometimes we see acute toxicity testing starving the organisms before testing to ensure that ingestion occurs. However, especially when talking about extended toxicity acute testing (i.e. 72h+ for Daphnia) some stressing could occur due to starving and this could cofound the results. Also for chronic testing care should be take to make sure that enough food is provided so that any effects cannot be due to starvation etc.

Great!, now it's more clear. Thank you very much!

Name: Eliana McCann Smith (for Tae Young Kim)

Question 3: How do you know that the mice are ingesting the same amount of plastic. It would be normal for mice to drink different amounts of water and therefore ingest different amounts of plastic, would it not?

I am also very interested in your results-- you said that the data analysis is still ongoing? Answer: Thank you for your question. Based on the calculation that I have shown we provide mice with water bottle containing a given amount of microplastic. As you pointed out, mouse may not drink the same amount of water daily, but mouse will take microplastic that we provide eventually because we feed water containing microplastic for a month or up to 6 months.

As for the data analysis, we collected brain, heart, liver, stomach, spleen, kidney as well as blood for this experiment. If we finish the data analysis of all of these organs, we could have a holistic picture of how a long-term exposure to microplastic affect the physiology of mammals.

Thank you very much for the reply! And nice presentation too. I am interested in looking at your poster more closely but can't seem to find it on the website...if you are okay with sending it to me that would be great! My email is mccannsmithe@gmail.com

Yes, for some reasons I cannot see my poster on the website. I will send you my poster at your email. Thanks a lot!

# Questions and comments for the session 25.10\_Ma

Simon Wieland: If any questions are left, or not answered to your full satisfaction, you are very welcome to contact me, of course! My email address: simon.wieland@uni-bayreuth.de Thank you again everyone for all of your questions :)

### Question for Simon:

Barbro Melgert: what kind of cells did you choose and what was your rationale for chosing these?

Nooooo, where did you go? What happened to the zoom? Some random other people now without sound. Sorry missed your answer now:-(

SImon Wieland: @Barbro: Thank you for that question! Up until now, we looked at immune cells, Macrophages (J774) and B-Lymphocytes. When MPPs pass barriers, for example the gut epithelium, these usually are the first cells that might come into contact with the particles. They also might be "distributors" of particles in the organism, so we thought they would be interesting.

Currently, I'm working with gut epithelial cells (STC-1), because these might be the cells, that might come into contact with the particles first, when MPPs are taken up via food. I hope that answers your questions.

So what about epithelial cells then (they are the very first to see them)? Any plans of checking those too? They are less phagocytic, so I wonder if the particles will stick as much

This chat is a bit awkward, but thanks for taking the time to answer, very interesting study! Yeah, it seems like we have a bit of a "jet lag" while typing...;) Thank you so much for your interesting questions!

## @Simon:

Alf Red: Do you think that fragments or fibers behave different?

Alf, thank you so much for your question. Well, I think that the primary reason for particle stickiness is probably due to their surface properties or properties of the ecocorona. This should be independent of the particle shape, so probably they would behave similar, at least qualitatively. But since our method requires Stoke's law of friction to calculate the force on the particles, we need to stick to spheres. So our method cannot really answer your question, unfortunately.

I could imagine that the contact area is larger for fragments than for spherical particles of the same size. Wouldn't the stickiness be higher? Thanks for your answer ;)

Yes, of course, I'd assume that the stickiness of a particle is proportional to its contact area. So larger particles, or especially particles with larger surface/volume ratio probably will stick stronger. Good point of yours! Thank you :)

Simon is the functionalisation specifically COOH-groups? So you may could just reduce the pH to test to which extent the electrostatic interaction contribute to your interaction. (Stephan Rohrbach C04)

Hi Stefan, thank you for that interesting question. The functionalization was COOH, yes. But we probably can't just reduce the pH to test for electrostatic interactions, because then our cells will die. What we are currently doing, is measuring the attachment of particles with different Zeta-potential to the cells, and trying to find a model for the stickiness as a function of the zeta potential. That will probably help us to understand the extent of electrostatic interactions in the "stickiness" of the particles.

# Christina Bogner

@Simon: Can you exclude that the effect is due to surface roughnesses: pristine paritcles beeing "smoother" than those with ecocorona?

That is a good question, Christina. We did SEM images of the particles; the functionalized and pristine particles do not show any surface roughness. The IgG particles and ecocorona particles have a slightly increased roughness, but I'd be surprised if that is enough to explain the differences in stickiness. But yes, of course you are right, this probably has an influence at least to some extent. Is your flow turbulent and would remove the particles if they are not properly attached to the cells? No, the flow is completely laminar, the Reynolds number of that system is far below 1. This is a prerequisite for Stokes law of friction, that we use to quantify the forces acting on the particles. I hope that I answered your questions with that :) Thank you!

Questions for Alison Foley

Name:...Christina Bogner Question 1:...How old are your participants on the average? More kids or more adults?

Name:...Markus Rolf Question 2:...How many people are involved in ten little pieces?

Hannes Laermanns: Can you say something about the size distribution of the particles? So more small or more "large" microplastic particles?

Christina Bogner:

@Thomas: Where are your plants situatied? Bavaria only?

The plants we investigated were located all over Germany, we inquired and we examined the plants that gave their consent. The composting plants and the combined plants (biogas - composting) were all municipal plants, they used as substrate the organic waste from households. Otherwise, commercial plants who produce potting soil, didn't want us to examine their fertilizers. Markus Rolf:

Do you know why you found less plastic particles in biogas plants than in other composting plants? The reasons here are the used substrates. As described before the combined plants (biogas - composting) and composting plants used organic waste from households as input. This is often contaminated with plastic bags and other impurities. The biogas plants we investigated, were agriculture biogas plants. There the substrate was liquid manure, silage, renewable raw materials and so on. This substrate is rarer contaminated with plastics.
Day 4/5 Pads

# **Questions and comments for the session 26.1\_O**

Name:...Amy Lusher

Question 1: Regarding your method, would you consider an adaption that allows you to collect the smaller particles and perhaps run this through a different analytical technique (Pyrolysis or  $\mu$ FTIR).

Name:...Maria Kazour

Question 2:... Thank you for your presentation. Just a methodological question on using KOH 10%. Why was it left for 3 weeks at room temperature instead of heating the solution (at 50 or 60°). Wouldn't that decrease the digestion time of your solutions and gain more time?

Name: Eike Esders

Question: Thanks for this very clear presentation. Are the sea salts used by Norway coming from the two mentioned regions? Do you think other regions could have higher loads of micro plastics? Thanks!

Name: Aline Carvalho

Question: Thank you for this nice presentation. Could you please explain again how you get your samples? And did you perform blank contamination control?

Kristina Klein: Sound is fine, when you turn up your speakers :)

Question: Probably missed it, but what kindd of PE-MPs are we talking about? Do you know whether these MPs are high/low-density PEs and whether they contain any chemicals? Thank you for your talk!

Alice Horton

Very nice study! How did you measure ingestion? E.g. Were your particles labelled/did you digest your organisms?

Hello Gema, I am very sorry I lost your presentation. any chance to share it with me? I am interested in the treatment you have used for digest the samples. Thank you. Juliana Ivar do Sul (julianasul@gmail.com)

(Gema): not sure if I can answer here. I will try to upload the video somewhere (freely available) after the weekend if my co-authors autorize me. I let you know. Thanks for the interest.

# COULD THE CHAIR PLEASE PLAY THE SOUND ON VIDEOS DIRECTLY VIA THE PC AND NOT VIA THE COMPUTER MICROPHONE?

Is it better now? There is no sound There is no sound. When sharing, check the computer sound not the microphone No... no sound!

It is not your talking that is the problem, it is when you play the presentations they are way too quiet. you need to share your sounds via the computer NOT your microphone

Yes it is working now Perfect!

ok! thanks!!! yesterday it worked perfectly....I am so sorry

WHEN OPENING ZOOM, SELECT PLAY PC AUDIO, THEN YOU CAN USE YOUR MIC AS YOU WISH, AND THE SOUND FROM PRESENTATIONS GO OUT DIRECTLY, AND NOT VIA THE SOUND PLAYED INTO THE ROOM YOU ARE IN. :)

Stephen Kneel How long and and what temperature were the mussels digested for? Great project by the way.

Catharina Pieper

Q: what was the most challenging matrice to work with? super nice work!! :) its is so important to have the big picture and include water, soil and organisms

#### Name: Aline Carvalho

Question: Thank you for this really interesting presentation. Do you think the difference in digestion protocols would interfere in the comparison between MP abundance in different compartments? And how many replicates for each sampling event and compartment? Thank you :)

Marie Babinot : Great presentation, thank you! How can you explain the fact that there is no seasonal variations change in sediment concentrations when the geomorphology of the beaches changes a lot according to the seasons?

Name: Conrad Sparks: Great presentation and work. How was potential contamination accounted for by using citizen science method used? Thanks!

Great talk Lola and really interesting. Were the polymers in each matrix of a similar proportion or were there noticeable differences? Thanks for this Lola.

Jessica Stead: Do you think there could be any effect of where you sampled on the beaches? E.g. low tide vs high tide, one is likely exposed more to seawater. Lola: answer: Yes I think there is but it must depends of the hydrodynamic of the sites/locations, may

be the sediment composition as well. (thin or coarse grains)

Here is my email if anyone has other questions: lola.paradinas@sams.ac.uk Thank you for all your questions :-)

# Questions and comments for the session 26.1\_Me

Name:...Bethanie Carney Almroth Question 1:...Great study! How are you measuring the surface area of the tissue? Are you extrapolating from standard data? is peristalsis active in the tissue ex vivo? Surface area including villi? ok thanks

Name:...Thibault Masset

Question 2:...Thanks ! Did you analysed Pd via Single particles ICP MS ? If yes, How did you separate MPs from the the tissues to have a clear count of the MPs ? Name: Ika Paul-PonQuestion: Great work and presentation thanks! Did you measure Pd in the external media (ie did the NP translocate from the gut sac to the external media)? Thank you!

Name: Camille Richon (Question for Melisa)

Q: Thank you for the nice presentation! Do you have an idea why the fibers found in GI are smaller? Are they fragmented or just from different origin?

Cheers! :) Thanks Camille if you have further questions contact me (melisafs@criba.edu.ar)

## Name: Alex McGoran

Q: Really interesting talk, Melisa. I was wondering whether you would be able to share a copy of the video of your presentation. I am also looking at microplastic ingestion in shrimp and I would love to be able to watch it again. Also, are your results published?

#### Juliana Ivar do Sul here

Hello Carmen, very nice presentation! happy to see you doing a interesting work! where do you go from here? if ingestion seems to be not an important effect, what would be an important effect from aquaculture?

Ok, so there is an effect even if microplastics are finally egested (thats a comment only) :) they are nor very heathy

Bettie:

Why did you choose to use 10% of MPs in diet? Is it an environmental concentration of plastic? Sorry Carmen I've got disconnected so I did not listen to your answer, is it possible to write it instead...

## Name: Tang Tony (Question for Yang liu)

Question : Nice presentation! what is the major difference between the experimental environment and actual aquatic environment such as lake and river. Could your adsorption curve be applied to lake or river environment.

Bettie:

Thanks for your presentation, you used artificial fish fluids, but from which species did it come from? (zebrafish?) Can you send me the literature you used by mail please, bettie.cormier@u-bordeaux.fr

Thanks :)

# Questions and comments for the session 26.1\_Ma

#### Name: Katrin Wendt-Potthoff

Q: did you think of using benthic animals (shredders) in future experiments, if they increase degradation? Thanks a lot for the nice talk! This is definitely something we are looking into in the future, especially now that we have this initial baseline understanding of flume dynamics. Q to Barbara: great presentation, but the time course was too quick for me: do you expect change with time regarding the ship-based polymers, and will the lockdown maybe have an effect?Hi, Katrin, nice to "read" you! I wonder if the lockdown will have a really big effect on shipping trade

but it might will. Most plausible we have more a kind of actuell blueprint and high dynamic conditions. Accordingly the signal might not change in general and poit still to the maybe lesser impact.

## Name: Pruvost Jean

Question 1: Thanks a lot for this awesome presentation ! Will it be possible to obtain the video of your presentation in order to show it to colleagues currently in a meeting ? Thank you very much. Great presentation and subject !

How did you mange to prevent MPs pollution from the used sediments ? Thank you, because we knew what we spiked the flumes with we only counted those particles. Ok thanks

#### Name: Alice Horton

Question 2:...DId you pre-filter the water you used in the flumes, and might there have been small particles in that water which is why you were detecting very small ones? Or particles derived from the flumes themselves?

Thank you for the question: We did not prefilter the water. But we did have three control flumes which did not have these particles. We also only counted particles that had the pink colouring.

## Name: Cleo Stratmann

Question 3: Did you measure MP in sediment and water column? what means "settling out" ? does it mean settling onto sediment? sorry if I did not understand all, the sound was difficult. Thank you: Hope I answered your question. We just looked at the water column for now, but looking at the sediment in the future would be important. Trying to find a way to collect a sediment sample without interfering with the flume needs to be investigated though.

## Name: Stefan Dittmar

Question: Thanks for this really interesting presentation! Did you already publish the results? I would like to read it in more detail :) And one question: What was the diameter of the fibers? We are only analysing the results now, but hope to publish next year. The fibres were 500 micron in length, diameter was 30 micron (but let me check :-)). Ah, thank you for this information. Really interesting as I am investigating settling velocities of small microplastics (< 300 µm) at the moment... Correction - 13 - 15 microns Perfect, thank you! My e-mail is h.a.nel@bham.ac.uk if you want more information and also what did not work lessons learnt :-)

Hi, the audio is not so good for Holly Nels presentation. is it possible to turn up the volume? theres also interference of mouse clicking and keyboards

sharing some advice from twitter to get better sound, when you share screen, there is option for 'share computer sound' then sound will be better

Ok, thank you :)

Happy to share the video with anyone who needs. I'ld be glad to have it, thanks a lot (jean.pruvost@entpe.fr)

## Name: Cleo Jongedijk

hi, very nice work, this kind of experiments are really needed in the modelling field! I was wondering if you also did reference testing with quiescent water to check the different settling of smaller and larger particles/fragments?

## Barbara Scholz-Böttcher

Q: Do you plan experiments with salt water /marine conditions as well? We are not planning these, as the flumes are more appropriate to understand stream dynamics. But I think the lessons learnt will be useful for all mesocosm experiments.

Question to Andreas: Have you checked the TOC-contents as well?

Miguel Tamayo: Thank you for such nice presentation, did you check if the smaller grains generate hihger abrassion what would contribute to increase the abiotic degradation rate? (in parallel to the biotic degradation) Thanks. We tried to exclude potential abraison, the sample films were anchored on the sediment surface and did not move, therefore I think there was no physical impact on the samples Thanks!

Name: Stephan Rohrbach: Great results: Have you tried to check for bacterial activity (qPCR etc.) in the different sediments? No, these were pilot experiments to get a general idea if there are differences at all. But of course we'd now like to know more about the reasons for the differences.

Matthias Egger: Exciting work! Curious to hear your thoughts on the relative importance of different grain sizes vs. different environmental conditions (UV, temp, precipitation)? What is more "important" in terms of biodegr. rates? Hi thanks for your question. This is tricky, since all factors influence each other and also the microbes. With this experiment I cant answer the question, personally I think that temperature is very important, but it doesn't explain all the impacts on biodegradation. We need more and specific experiments for this kind of questions.

RACHID DRIS : Hi Barbara, Thank you for the nice Talk. You said samples go through a size fractioning cascade. Do you pryrolise all size fractions together or one by one ? Is PyrGCMS suitable for all size fractions. Maybe some are more challenging ? Hi, Rachid, greatings to Paris! In addition to the already given answer the higher size fractions were rarely abundant. Accordingly, there was no problem measuring the samples.

Sonja Oberbeckmann. To Barbara, greetings and thank you for the very interesting talk! Do you think the different distribution of the polymer types can also have to do with the polymer characteristics/density and different floating characteristics on top of being from different sources? Hi, Soja! We assume that the paint particles are very small according to abrasion conditions. In the 2.5 m depth we sampled and the point that we hardly observed any "big" particles in the data of the sea surface samples are supposed to be diffrent and complementary but should reflect similar observations. I don't think that another "source signal" occurs in different layers due to severe mixing conditions.

Winnie Courtene-Jones: Thanks for this. is it possible to work with any of the shipping companies to understand (or sample) some of the paints and coatings they use on their ships? perhaps there's too many - yes there's a lot of variability in the paints. Thank you

## Cleo Jongedijk:

Hi Barbara, really interesting thanks! I was wondering if you think there might be a correlation between the larger stirring/mixing of water column in the shipping lanes and the sampling results of the paint fragments. I remembered most paint fragments to be slightly heavier than water so do you think in other regions they have simply sunk away from the observable area (surface)? Dear Cleo,

thank you for your interesting question. The point you mentioned is propably the most plausible one. Accordingly I would expect a great share of paint fragments in the sediments as well. Dynamic conditions might lead to higher mixture processes and therefore the shipline-concentration hint might get lost. Thanks Barbara, So I guess if we can show that the sum of the plastic abundance in the total water column + sediment is high around shipping lanes, we might have found a clear source. That would be a really cool (although not so cool voor the ecology/environments of shipping lanes) result! Are you planning to do any sediment measurements in the future?

Bianca Unger: Thanks, Barabra, for presenting this interesting results! Do you know if areas with a high amount of MPs in sediments in the German bight overlaps with the areas you identified? Or are they completely different? Hi, Bianca, nice to "read" from you! So far we are concentrated to analyze only sediments that come from closer coastline sampling areas. Here we see at least similar patterns. We have some open shore sediments as well but they are still on our "to do" list. Thank you Barbara, for your answer! Good luck with your further research!

# For MELISSA

Name: Matthieu Mercier (IMFT, Toulouse) Q: can you describe the "turbulent" conditions you mention ? It can play a very important in fiber clustering.

# For MELISSA

Name: Rachel Hurley (NIVA, Norway) Did you chemically characterise the fibres that you found? Were they all PP or could some have come from background contamination?

# Questions and comments for the session 26.2\_O

## Name: Bettie Cormier

Question 1: thanks for your presentation. Do you have an explanation why you were not able to see effects on the valve opening when oysters were exposed to the cocktail while you saw effects when oysters were only eposed to MPs alone? Another hypothesis would be the speed of MPs effects. If it's a mechanical or physical interaction with mantle or gills of oysters, maybe a chamoreceptors could activate and show by behavior response like increasing micro-closures... Thanks Arno, was just surprised since I am not working with oysters and valve activation is new for me! Nice talk! Just another question, I missed the beginning of your presentation, which size of MPs did you use? Thank's Bettie :) We used 20-25  $\mu$ m PE MPs microbeads for this experiment but we're going to use environmental MPs with focus on shellfish plastics materials in future experiments. Great! I will follow it then ;) Thanks

Did you check particles distribution in tanks in MP and MP+contaminant conditions? Similar exposure/bioavailability of MP?Thank for your nice presentation! Yes we checked and validated with Flow Cytometry to find MPs class size of 20-25 µm.

Questions for Guilia Brocardo :

Name: Christoph RUmmel (@ Francesca): Q: thanks for the talk. did you have the chance to sample natural substrates (leaves, wood?) Name: Vincenzo Donnarumma

Question 2: Very interesting talk, Francesca! You said you have found different classes of degradation, how did you evaluate it?

Name: Rico Leiser (Magdeburg, Germany)

Question:@Franceca Congrats to the nice talk and work! What families of cyanobacteria did you find on the plastics?

Stephan Rohrbach: Do you think the differential colonization is more dependant on the degradation state of plastic or rather on the time the plastic was in the lakes? Very great work! Jafar , Hungary great work, my question is: on which bases you classified plastic particles into 40 groups? how did you determine the initial colonizers on microplastics?

#### Name: Vincenzo Donnarumma

Question (Brittan): Interesting talk! Have you got any information of time residence of the microplastics you analysed? As you found a lot of Alphaproteobacteria and being them usually initial colonizers, can you tell the link between biofilm and time residency?

Name: Stephan Rohrbach: Excellent talk! Do you think there is a correlation between the deep water horizon catastrophy and your findings in the sargossa sea or is there another reason why you took samples from this specific location?

Name: Robby Rynek

Question: Thanks for the nice talk! If I got that right in the plastic fraction you had some samples that were classified as "unknown plastic". How are you sure that they are actually plastic and not also something natural?

great work, did you publish this work?

# Questions and comments for the session 26.2\_Me

Name: Joao Frias Question 1: Great talk. How did you distinguish sources among the several sites in the Mjosa lake? In other words how did you distinguish sources within the core/core slices? (I refer to the slide with different colours circles) Thank you for your answer.

Name:...Florane Le Bihanic

Question 2: thanks for the very nice presentation. Could you please detail how did you calculated the initial 200t of microplastic budget

Second presentation

Name: Juliana Ivar do Sul Question: Hi Gerson, thanks for this cool presentation. Thats a huge effort! did you consider plastic fragments in the same sizes as pellets or within which size categories? Thank you excellent! this gives the data a good power. I wish this index starts to be used more often

#### Name: Ghezali Yousra

Question: thank you for your great talk! can you tell us more about the difference in distribution of MPs between winter and summer!!

Name: Fernanda Santos Question: thanks for the very wonderful presentation!

3rd presentation

Name: Winnie Courtene-Jones

Question: Thank you for the presentation. the yacht make a great opportunity to collect these measurements. can you explain the methods for microplastics more. how are you sampling for microplastics? are you sampling continuously? can you elaborate more please. - great Thank you

#### Name:Carla Elliff

Question: This is a great example of ocean stewardship and ocean literacy, thank you for the presentation! I was wondering how the partnership began. Was it the Ocean Race that approached the scientific community or was it the other way around?

As we are now entering a new era for the race i am also getting in contact with other institutes. So now it's both ways. Feel free to contact us: soren.gutekunst@ext.theoceanrace.com

#### Name: Victor Onink

Question: Thank you for your presentation. The sailing ships do indeed provide a great opportunity to sample remote parts of the ocean, but the sampling methodology is quite different from most other studies. Given these methodological differences, how easily can you compare the amounts of plastic measured by your ships with those from other studies? Thank you!

#### Name: Cleo Jongedijk

Question: The machines that have been developed for the volvo ocean race boats used the availability of the huge budgets of the development teams. Is there any plans to install/convert the automated sampling machines as installed in the keels of the ocean racers also in normal sailing boats (cruisers/other racing activities like Vendee Globe or (mini)Transat type races?)

## Name: Delphine Dobler

Question: Thank you for your great presentation. These measurements offshore are indeed very precious. Have you made the measurement of microplastics concentration publicly available ? Thank you very much for your answer.

Dear Delphine: You will find the paper with the following DOI within the next one to two weeks here:

https://journals.plos.org/plosone/article?id=10.1371/journal.pone.0243203

Title: A near-synoptic survey of ocean microplastic concentration along an around-the-world sailing race.

And the data of 2017-2018 either in SOCAT for CO2 or here: https://www.ncei.noaa.gov/access/metadata/landing-page/bin/iso?id=gov.noaa.nodc:0170967 \_MICRO 2020. Fate and Impact of Microplastics: Knowledge and Responsibilities. 23-27 November 2020\_\_\_\_

or here:

https://www.emodnet-ingestion.eu/submissions/submissions\_details.php? menu=39&tpd=232&step\_more=9&step=01223

Thank you for all the details. I really appreciate this.

Fourth presentation

Name: Carla Elliff Question: Were you able to correlate or estimate the age of this deposit?

# Questions and comments for the session 26.2\_Ma

QUESTIONS FOR PRESENTATION 1 (Natalia Sanchez; Concentration of marine litter on the beaches of the Galapagos Islands and main factors that influence its distribution)

## Name: Christina Bogner

Remark 1: Thank you! In your plots showing correlation, you have one or few points far right (large data). They influence your correlation. Other points are clustered and don't show a linear relationship. Perhaps you would like to check these large data points and try to find more robust statistics.

## Name:... Alicia Mateos Cárdenas

Question 2:... Hi, nice work Natalia! I have two questions. When were the samples taken (year, season?). Also, the Galapagos have several beaches closed to tourists. Sorry if I missed this, did you sample both touristic and non-public beaches? If so, any difference on the type or abundance of plastics? - Gracias :)

Winnie Courtene-Jones: Thank you for your talk, already a lot of work collecting these samples. do you plan to sample in different seasons? i think the dominant wind changes, so you might get different distributions?

- very good, thank you. interesting work and a interesting future investigation!

Stefanie Ypma: Thanks for your presentation, observation at the Galapagos Islands are very useful for our project, where we try to model the pathways of debris towards the Galapagos! To comment on the question of Winnie, we model a strong seasonal dependence of plastic beaching, so would be very interesting to compare this with observations from different periods! Also, are you planning to look at an estimate of the age of the samples you find? So we could figure out what the possible sources could be?

Hi Stefanie, currently we do not plan to do so due to the means we have because we are working in Spain so it is very difficult to carry out a temporary monitoring of these characteristics. However, what you comment would be of great interest for the project. Thank you very much for the recommendation and we would like to take into account in the future.

Natalia Sánchez-García: Thank you very much for your interest, I would like to share the following links about sampling in case it is of your interest.

Microplastic extraction protocol in marine sediments - Laboratory

https://drive.google.com/file/d/1ite6o2zUQNe-4gmV8vzzIxS1fgSvNy0h/view?usp=sharing

Protocol for extraction of micro and mesoplastics in coastal marine sediments - Field work https://drive.google.com/file/d/1Sr-6rp8avlCIZI1UtXAPpGV4C9z5iQZm/view?usp=sharing

QUESTIONS FOR PRESENTATION 2 (Christian Laforsch; Ecotoxicology of microplastics - knowledge gaps. and possible steps forward)

Name:...vThibault Masset Question 1:... Thanks for the talk ! Do you recommand to apply the same guidelines for tire particles ecotox testing as for MPs ? Some of their characteristics are very similar to MPs (they are actually considered as MPs nowadays)

Name: Christina Bogner

Q: Thanks for the talk! You mentioned that the effects could be different for different animals. Do you recomment particular animals for particular ecosystems? Thanks :-)

Name: Alicia Mateos Cárdenas

Q: thanks for your presentation, what type of natural particles do you suggest that we test (as controls)?

A: silica beads have been used successfully, or kaolin in other studies. I think creative choice and testing of other alternatives is a research topic for the future

QUESTIONS FOR PRESENTATION 3 (Sabine Pahl; A snapshot of global actions to reduce the flow of microplastic to the ocean from the UNEP stock-taking exercise 2020)

Stefanie Ypma: Very interesting talk! Can you provide a link for the interactive dashboard and the repository here? thanks!

We're just checking if we're allowed to share these links already, as not the final version as far as I know. My e-mail is sabine.pahl@univie.ac.at if I don't have time to share on here soon. Thanks!

not sure you're still here Stefanie but here are the links:

https://environmentassembly.unenvironment.org/stocktaking-dashboard

https://environmentassembly.unenvironment.org/stocktaking-online-repository

QUESTIONS FOR PRESENTATION 4 (Cristina Panti; Assessment of multiple impacts of marine litter in the Adriatic Sea: from fishing for litter to fish species)

Name:...Stjepan Budimir

Question 1:...Great work, greeting from other side of Adriatic. In the macro litter found on the bottom, was it possible to identify the items, and labels indicating the country of origin? DO you think in the regional shared seas like Adriatic, Baltic is there a point of looking for country of origin or is just an "excuse" to point fingers on the other side of the sea. Such behavior i observed in Adriatic and the Baltic sea.

Thank you, its almost a joke how in every country its always "the other side" when asked the fisherman :)

Nur Hazimah: In your findings for the comparison of chemicals in fish with or without MPs, what do you define as no MPs? Could it be that these fish ingested MPs but had egested them? Thanks! for this reason we believe that measuring MPs ingestion, contaminants and, biological effect together could be the right approach not only one variable at the time

# Questions and comments for the session 26.3\_O

Speaker: Latchere Oihana

Name: Anna Kukkola

Q1: Great presentation! Wery interesting and informative. How did you obtain the nano-fractioned environmental plastics? Did you produce these in laboratory and what size distribution were these? Thank you

Name:Anja

Q1: How did you isolate the hemocytes?

Name: Bénédicte Morin

Have you checked the presence of NP or MP in the organism? What is the concentration into the organism?

Thanks !!!

Name: Nina Paul

Q: Thanks Oihana for the interesting talk! Do you think that there may be less ingestion of NP/MP because of lack of food in the water? I came across some studies (and even in the lab) that the intake of MP was much less when there was no "trigger" from food in the water?

Name: Julia Pawlak

Q: Thanks for this nice presentation! Do you also want to analyze the environmental plastic for any absorbed chemicals/pollutants or plasticizer? I'm looking forward to results of the trophic transfer experiments! really interesting!!

Speaker: Gonçalves Ana M. M.

Name: Ana Luísa

Q1: Hi Ana, thank you for the nice presentation. But did I see well? I believe you had a decrease on LPO (in lipid damage). How can you explain that? Do you have any hypothesis for this? Thank you Nina. My internet is not nice and I can not go back in the presentation :)

-> Nina @Ana Luisa: I think it was an increase of LPO in bigger size mussels - you're welcome :)

Speaker: Staal Yvonne

Name: Anna Winkler

Q1:Congratulations, substantial and comprehensive work! Did you choose these size ranges because you generally assume particle transfer into the blood for these sizes? --> Did you also observe a cell internalisation?

Q2: Given that you analysed the physico-chemical properties of used polymers, would you refer the observed results only to your plastic particles or also to any non-plastic particles? Thank you for answering. PS: if you are a molecular biologist, maybe you are interested in the findings of my poster 24.8\_Ma ;) (microplastic fibres tested on human lung organoids) Yvonne: Thanks! I will have a look at your poster! To answer your second question, we have tested plastic particles, but cannot tell whether the effects are due to the plastic or to the particles. A challenge is to find good particle control samples for this. Though I would say these are a combination of both.

## Name: Anja

Q2: Thanks for the nice talk :). How do you explain the observed effects? The particles you used are rather large to interact with cells. May the effect you observed be due to the oxidation of the particles by using H2O2 rather than the particles themselves? How did you ensure the removal of the H2O2?

--> (A.Dehaut) Removal of H2O2 --> rincing samples with 4 bath of pure water.

#### Name:Marte Haave

Q1: Very nice work. Thanks. interesting techniques on i.e exposure in wells.:) could the higher response in smaller particles be due to a higher number of particles? Any view on whether cryomilling caused different shapes than natural MP? Yvonne: microscopically the particles largely resemble natural MPs Thanks :)

Name: Kristina Klein

Q: Thank you for the talk! Have you considered testing the MPs without prior H2O2 treatment as such would represent truly "aged" particles?

Speaker: Gravato Carlos

Name: Thibault Masset

Q1: Thanks for the talk ! Do you think that Larger particles (100 - 200µm) could be ingested by these organisms ? If yes, would they promote clogging ? Also, Can't the effects observed be a results of chemicals uptake of toxic compounds ?

# Questions and comments for the session 26.3\_Me

Add your email address if you want to have an exchange with the speakers later.

Questions for Alessandra Cera

Questions for Laura Markley

Name:...Susanne Belz Question 1:...How did you validate you identification of microplastics? what is the uncertainty/trueness of MP identification? How was the decision tree developed?

Name:...Rachid Dris

Question 2:...Thank you for the nice presentation. We can suppose that the small amount of plastics found in the net samples is due to the cuttoff.

Why do you think we have always more plastics in the bulk in comparison to the bucket ? Which method do you concider more biaised at the end ? As the volume with the bulk is lower, we should expect higher variability (due to lower representativity)

#### Name: Garth Covernton

Question 2: Thanks Laura!!! Why do you think your bucket sample concentrations were 2 orders of magnitude less than the jar samples? Can you remind us of the lowest mesh size each sample type got filtered to?

## Sam Athey

Q: Hi Laura – great talk!! I know you have not conducted chemical composition analysis of particles, but do you expect to see differences in concentrations of the three methods dependent on other particle characteristics, such as polymer type? Has this been concluded in other studies comparing sampling methods?

#### Trang Nguyen

Thanks for your presentation. In your opinion, which method should be used to collect representative samples for moving water bodies like channel, streams, etc with effects from peak events like runoff/stormwater and overflows? Thank you!

Claudia Lorenz (AAU): Thank you Laura for this great presentation! My question relates to Rachid's...If you compare only the MP larger than your net mesh (300 µm if I got it right?) do you still see this huge difference between net and bulk sampling? Also, how did the size distribution look like?

Questions for Mariana Oliveira Rodrigues

## Name: Trang

Question 1: Thanks for the talk! I want to ask if you checked the density of the reused ZnCl2 solution? In my case, the density of ZnCl2 is prone to decrease after using Name: Mariana Rodrigues

Answer: If you do not add any water during the recover to the initial solution the density does not decrease. The solution should be stirred during 24h approximately to guarantee that the solution is homogeneous. Thus, when we filtered the solution the loss is neglible (density did not decrease).

Another question: One of my colleague observed the difficulty in sinking of heavy particles due to the high viscosity of ZnCl2, is this your case as well?

Name: Mariana Rodrigues

Answer: I don't know if I explained myself correctly. Using the correct concentration of zinc chloride we do not expect that heavy particles sink, unless they have a specific density above the 1.6-1.8 g/cm3. I've tried different concentrations of zinc chloride solution until I found the correct. When I tried concentrations below 700 g/L, particles tend to sink and I do not see viscosity in the solution.

## Name:..Rachid Dris.

Question 2:...Hi, thank you for the good presentation. Can you explain more in detail the "deagglomeration" of fibers with the ZnCl2 density separation. Do you mean that we should not concider the fibers results good when we use ZnCl2 separation

Robby Rynek

Q: Thanks for the great talk! Except the price, is there any advantage of ZnCl2 compared to NaI or sodium polytungstate? From my experience both are also reusable. Thanks for the answer.

Marie Babinot : Great talk! Have you tested this method with other high density salts (calcium chloride of density 1.4 for example - It does not have the disadvantages of ZnCl2 but a higher density than NaCl and is also reusable)?

# Questions and comments for the session 26.3\_Ma

Please add your questions early as there is a 30 second lag between recording and the video displaying on the website! :) Thanks for beeing so well-prepared<sup> $\wedge \wedge$ </sup>

SPEAKER 1: MEREDITH SEELEY contact info: meseeley@vims.edu

Name: Camille Richon Question: Thank you for this talk, that's a very interesting study! How did you characterize the bacterial communities? did you visually identify or did you use genomics...? Cool thanks! That's a great study :)

Name: Stephan Rohrbach

Question: Splendig presentation! Have you added virgin plastic to the incubation or was there already attached biofilms of some form attached to the plastics? And if you used virgin plastic: Have you analysed biofilm formation on the added plastic after the incubation?

Name: Alice Horton Q: Thanks for a very clear talk! Do you think the same effect would be seen with macroplastics? Or is surface area important?

Name Vincenzo Donnarumma Q: THank you for your cool and clear talk. Just a curiosity, why 16 days at longest time scale?

Name: Linda Hink

Q:Great talk! Did you consider looking at the archaeal community? Marine nitrification is dominated by archaea - they are not that abundant and could be missed when looking at 16S rRNA only

Winnie Courtene-Jones:

Excellent presentation, really clear and informative. Thank you. just curious, the effects you saw with N cycling, how do you think this would translate to wider ecosystem effects?

- Thanks, all the best with the rest of your study - such fascinating work!

also, please paste a link for your paper :)

Meredith Seeley:

Thanks for all the great questions! Here's a link to the paper: https://rdcu.be/b3697

- Thank you :)

## SPEAKER 2: ESTER CARRERAS-COLOM Contact: ester.carreras.colom@uab.cat

#### Name: Anja Rebelein

Question: Great study! Did you determine the number of fibers that equals 0.15% in the feed? So, what was the "particle number" you used? Fibers are usually really light-weighted and 0.15% sounds a lot.

#### Name: Meredith Seeley

Question: Interesting study, and cute crab illustrations!! Very vague question...but can you guess at how your results would vary between other species (e.g. fish)?

#### Name: Mandy So

Question: Great study! Does this mean the ingestion by crabs would promote biofragmentation of plastics? And if there is any accumulation of plastics in the gut? Thanks for your question! Based on the observations of the fibres that came out in the faecal pellets we could think that, yes. Not sure the size changed, but they appeared bended and with cracks sometimes but more studies would be needed to be sure of fragmentation actually happening. We didn't target that specifically. Alici Mateos-Cárdenas (et al) recently published a very nice work on fragmentation in amphipods :)Thank you very much for the answers. I am actually working on something similar but in HK :) Looking forward to your future studies!

#### Name: Alex McGoran

Question: Really interesting talk. Unfortunately my internet cut out and I only heard the second half of the talk. Would you be able to share your presentation video with me as I work on a vary similar topic and would love to hear about your research. My email address is alexandra.mcgoran.2012@live.rhul.ac.uk. Sure thing I'll be happy to talk with you! :)

## SPEAKER 3: ADAM PORTER

A.porter@exeter.ac.uk / @ap3489

## Name: Garth Covernton

Question: Thanks Adam, this is a great study. Have you considered controlling for methodology to see how it affects reported concentrations? I've done this for fish globally using GLMMs and it plays a large role in reported concentrations, especially the lowest mesh/pore size a study filters their samples too. Happy to chat more about this if you like (gcov@uvic.ca).

## Name: Amy Lusher

Question: Awesome talk as always Adam, did you take into consideration any of the methodological differences in reporting (ok you might be answering it now!) or make any corrections for studies that may or may not have made any data correction?

#### Name: Camila Vidal

Question: Thanks Adam for an amazing study! Did you find or do you think that there may exist differences between the types of plastics and predator preferences? shape, color or even type of plastic

Winnie Courtene-Jones: HI Adam, Thanks for your presentation. It's great to pull all of this data together. How did you classify organisms which fed in multiple ways? did you notice any polymer specific differences between feeding modes?

Thanks! great for the future work. good luck! metanalysis is challenging for all the reasons you mention with differently reported data. great work :) i'll be interested to read the pub.

SPEAKER 4: MATTEO GALLI E-mail contact: galli13@student.unisi.it

No further questions for you today.

# **Questions for The Editor's Perspective**

Your Name: Katrin Wendt-Potthoff

Question For:all

Question: there seems to be a bias towards microplastics in the publications. Are more papers about meso- or macroplastic being rejected, or are there fewer studies?

Martin: @Katrin: A recent meta-analysis demonstrates that there is no publication bias in the microplastics literature. Good news!

Thanks Martin! Do you have a link for me?

Will try to find it, also that is on microplastics/negative results, so, I think there is a bias towards microplastics but rather not towards reporting positive results (i.e. toxicity).

Rachid : @Martin What does no bias mean exactly ? Martin: @Rachid: Work from Chelsea's group. Will dig it up. They did not find a bias towards reporting toxicity compared to null effects.

from Winnie: - the Bucci et al paper 2020 i think ? Hey thanks Winnie! I was late to the pad :) Here's the link to Bucci, et al. 2020: doi.org/10.1002/eap.2044

Martin: Thanks! They say "Overall, 59% of the tested effects were detected." That means that 41% were null results.

Kennedy Bucci: I just wanted to clarify, we didn't specifically investigate whether or not there was bias in mp publishing, but we did find many published tox tests that report "no effect" (i.e. no statistically significant difference between treatment and control), which does tell us that these papers were not rejected (obviously). however, I will also say, anecdotally (meaning we did not specifically test this), that most studies reported ~at least~ one "detected" effect, even if they reported a bunch of non-detected effects. => Rachid : Thank you, i understand better. It is a positive information !

TvE: Blettler et al. did identify a "bias" towards microplastics, marine ecosystems, and M-LIC, although this was not with focus on toxicity or effects, but merely abundance/transport/etc.: https://www.sciencedirect.com/science/article/pii/S0043135418304597? casa\_token=YyQ293au4JsAAAAA:2dQFIzGzeOrYUGYfWoeNmDFiR7NK-o98Ptcu-YMmo47YkVxVtdAcSwCrOuXM8o402DN2x9A7SA

Your Name: Martin Wagner

Question For: all

Question: Your take on publishing negative results? Any problems with publication bias? same as Martin's Q :-)

Rachid: Great question, in the MP field, with a lot of "early" investigations, negative results can be helpfull !

Your Name: Christina Bogner

Question For: all

Question: What is the policy in the journals to publish negative results?

Your Name: Stjepan Budimir

Question For:all

Question: Can you give a comment about your view to the future of publishing concerning the publishing fees in open access journals, and even more important the high cost of subscriptions to the journals.

Do you think the prices for journal access are reasonable, and justifiable?

Widespread popularity of Sci-Hub gives us the indication that there are a lot of people in search for knowledge, but no institutional access.

<thank you for asking this complex question

Your name: Thomas Maes

Question for all: there have been a few Frankenstein studies recently e.g. anti covid jade chrystals, female phd supervisors, ... in relatively high IF journals. How does this happen? How can we avoid it?

HI Thomas, Ana Catarino here, what do you mean by female phd supervisors? Matt Cole: There was a paper in a Nature journal last week suggesting female supervisors was disadvantageous to students... oh I see... will take a look! Thanks!

https://www.nature.com/articles/s41467-020-19723-8

https://www.sciencedirect.com/science/article/pii/S0048969720363592?via%3Dihub

[Carla Elliff here] Have a look also at this link showing different groups speaking up against the results of the paper: https://www.sciencemag.org/news/2020/11/after-scalding-critiques-study-gender-and-mentorship-journal-says-it-reviewing-work Thanks!

Thanks for the reply, female supervisors rock! I had Heather Leslie!

Your Name: Matt Cole

Question For: All

Question: Bucket-loads of peer review requests come in every week... can you provide insight into your screening processes before papers gets sent our for peer review?

- i am interested in this too (Winnie)

Important Q (Martin)!!! Nobody can review all those manuscripts anymore...

Good questions (Joao)

Your name: Winnie Courtene-Jones

Question for: the panel in general

Question: with an ever increasing number of microplastics/plastics articles, are you finding (or anticipating in the future) a higher rejection rate, or increasingly stringent requirements, such as some of those discussed during this conference, that authors need to be implementing/striving for as we move forwards in this discipline I was going to ask this question as well, will you (in the new journal) state the basic requirements for publishing monitoring and ecotox studies? (Alicia Mateos) MattC: Great question.

Marte H: Echo this!

(Winnie Courtene-Jones): Question 2: What do the panel think about double blind reviewing process? while we all are 'unbased', i sometimes feel there are variations in reviewing process, and a double-blind could benefit new teams, or new researchers e.g. PhD students who don't have a 'name' in field. YEAH! Or fully open review, much more civilized (Martin)

- true!! this is probably a better option, as you say Martin.

Your Name: Anja Rebelein

Question For: All

Question: What do you think about publishing negative effects of microplastics on organisms. Do you want to promote this kind of articles to be published?

Ana Catarino, question for all. What's your view on supporting publication from researchers from emerging economies? (fees) Great question.

#### Name: Juliana Ivar do Sul

To Bart Koelmans: I have seem that your new journal is accepting suggestions for special issues. Any tips for how someone can be successful when submitting it?

Related question - Rachid Dris : how do you ensure that spetial issues are not redudant, there are so many right now

Your name: Juliana Leonel

Question for: to organization

Question: why there are only male in this panel? It would be great to hear a woman to answer the question regarding "Fransketein publications".

Thank you for bringing this up Juliana (it would be great for Micro2020 to ensure panels have a gender balance going forward (Nia Jones) (edit - thank you to the organising team for the great work already done on this!))! +7 Thanks! Thanks to the panel for addressing this question! (Noreen Kelly) so the problem is that the female editors need to be given a voice ? And soon, we have been talking about this for years. Excellent question!! I think the conference has made good steps towards more balance already though, both in terms of gender and in terms of geographical spread (Sabine Pahl)

I agree that we are doing better here (MICRO) Ju, I have the feeling we have more females presenting works than males, maybe good to check that! Yes and it is great, but if we do not have equitity on editor board we still have a problem (Ju Leonel). I agree, one step after the other? we have now maturity I believe to start taking the right decisions - in so many ways in the plastic research and including equality of genders

I think the MICRO2020 has great names to join the team of a new journal.

Next time aim for 50-50% panel!

Regarding the whole panel of Chairmen/Chairwomen/Intervenants at this Micro conference, I feel that women are really well represented and it really great ! (Charlotte Lefebvre) I think as well in a panel of just 4 people its very hard to include equality in gender/ geographical location and state of carrier for instance. Yes of course!

## Your name: Garth Covernton:

#### Question for: All

Question: In expansion to Matt Cole's questions above, do you have problems finding peer reviewers with so many submissions? I have been a reviewer for papers and had papers reviewed where there is only 1 or 2 reviewers, when 3-4 seems to be the standard in most other fields. Also, what steps do you take to ensure that peer reviewers are properly qualified? I see a number of papers

still getting through peer review that don't necessarily meet the standards of the field, especially things like transparent reporting of methodology, which peer reviewers should be calling out. Important Q, Matt has a similar one above. Thanks, will modify a bit!

Name: Christina Bogner

@ all: What is the journals' policy to data availability? I think of reporducibility of research. As data is often very difficult to generate (long lab work etc), it is a challenge to reproduce/check results.

Katrin Wendt-Potthoff, Question to all: what do you think of preprint servers for unreviewed stuff? These things stay around, not very motivating for reviewers...

Victor Onink: When submitting, journals tend to ask for recommended expert reviewers. For all the editors, do you generally take these recommended names for review or do you make your own considerations as to which people might be suitable?

Question: François Galgani, the paper from José Derraik published in 2002 at MPB

has thousands of citations now. what would be the next sub-topic on plastic or microplastic research to reach this level of citations that he now has in your journal?

Question from Giuseppe Suaria: Can you expand a little bit of single-blind or double-blind review? Don't you think that double-blind review would be more impartial? (especially from a reviewer's point of view)

Martin and I (Winnie) were also discussing this above..... regarding double blind, but actually as Martin pointed out, better for open process- probably have a better quality of reviewing

## Name: Eleni Christoforou

Question: How can a young scientist (finishing up a PhD) get involved in the journals? What career level should one be to become an editor?

yes, i was wondering this too (Winnie C-J) - not as a reviewer though (already do this), but in special issues, editing board etc.

I (Tony Walker) personally recommend alternate reviewers such as junior colleagues (PhDs, post-docs etc) to review a paper that I can not review myself when invited.

(Carla Elliff) I'm having my first experience as a guest editor for a special issue (I'm a postdoc, finished my phd in 2019). I feel it was a matter of showing up and a bit of luck in being at the right place with the right people. It is so important to actively promote your work - there is so much going on the in world, it is difficult for editors to keep up with new early-career talents. I hope I do a good job :) Carla, how did you get your name out there? Was it through conferences or did email some editors expressing your interest in joining their teams? I'm part of several networks and, in this case, it was through the Proplayas IberoAmerican beach certification network. There was a call for contributions for a document about covid-19 and beach tourism. I contributed an opinion paper about marine litter and covid-19. Being active like this caught the attention of the organizer of the document, who went on to propose the special issue (Ocean & Coastal Management) and he invited me to be a part of the team. I also liked the suggestion given by the editors in the panel of contacting them directly with your CV and explaining that you are interested in helping out as a reviewer or eventually a guest editor etc. I'm sure that will keep you in their radar when they have to think of names! Great thank you for sharing and the suggestions!

Thank you all for your answers! Tony that is actually a great idea I can recommend it to mu supervisors, in case they would like to pass me over some manuscripts.

Loved the lively chatter. Well done everyone and thanks Andy

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Winnie: Thanks for your comment Carla, i am at a similar career stage to you, so nice to know these options exist.

Your Name: Joao Frias

Question For: all

Question: Open access science is becoming more relevant, and in the Europe context, mandatory for research publishing. We spoke already about the large amount of review papers. Should focus shift, and special issues promote more empirical data papers instead of reviews? What is your view on the promotion of empirical data papers?

Great moderation Andy!

THANKS EVERYONE

Do chief-editors get paid?

AC: thanks! great debate

Thanks a lot to all!!

Ju Leonel: thanks for the panel, it was great! Congrats to the organization!

that was great! can we do it again? Thanks

Carla Elliff: We could talk about this subject for over an hour! :) really important and interesting topic for the conference

# Questions and comments for the session 26.4\_O

Would be useful to annpunce this on the website to be known by everybody! Thanks. Jessica Song: jessica.song@awi.de Nur Hazimah: hazimah.mohamednor@wur.nl Inga Vanessa Kirstein: ivk@build.aau.dk

Name:...C. Maes Question 1: Did you have one idea about the mass of each contribution of tyre particles entering the oceans?

Name: Christina Bogner Question 2: Can you comment on the difficulties to detect those particles e.g. via FTIR spectroscopy? What is the best method to find them in an environmental sample (complex matrix like soil, for example). Than

Name :Joao Frias Question 3: Great talk Richard. What is your view on roads made with recycled plastics? thanks for your reply. Name: Christine Knauss

Question: Thanks Richard, great talk. Was there a difference in size and density between the tire particles in storm water run off vs. atmospheric deposition? Also can you explain your method for collecting atmospheric particles a bit (ie a gradient away from the road)? Thanks!

Name: Nur Hazimah Question: Do you think there would be seasonal differences in the release of tire particles?

name; Steve Allen Sir Richard; What equipment are you using to characterize and quantify the tire particles?

Name: Kryss Waldschläger

Thank you for the interesting talk. Have you thought about temporal changes in tyre particles properties in the environment and the influence on the transport?

Name: Verónica Godoy

Which variables need, to be considered when accounting for tyre microplastics?? Has an influence the type of tyre, the pressure, the type of vehicle (light duty, trucks, etc.)the speed and acceleration...??

It might be my computer set up but the slides appear blurry, Richard would you be able to fix this issue?

i have the same, i can't read the text, professor thompson though is clear in the top corner, so it's not my internet connection i think -> it seems to be a bit better now

Name: Claudio Marchesi Why did you not perfor PCA instead of Cluster Analysis?

Name: Stephan Rohrbach

Have you also adressed the question if antibiotics is adsorbed by plastic surface? Very nice talk!

Name: Francisca Ribeiro

Question: You've mentioned that the concentration in number of particles (assessed by FTIR) and in mass (by Py-GC/MS) were in the same order of magnitude...How did you compare? Did you perform a conversion between the two measuring units? Great! Thank you

NAme Barbara Scholz-Böttcher

Q: Very nice talk Inga! How do you explain the striking difference between the qualitative composition between your estimated mass quantitative data and the py-GCMS data? Is the mass equivalence just by chance?

i think it's the polymer type composition Yes, thank you...

Have you compaired particle sizes? Types representing the bigger fraction FT-IR and masses/types via Py-GCMS...

Thank you ; ) ! Sounds very familiar : ) Inga, I would locking forward to a PDF : ) Thanks Barbara. It is available open access https://www.sciencedirect.com/science/article/pii/S004313542031054X

# ~Name: Thomas Maes

Q: I might have missed it, but what were the potential sources of these microplastics (I would think that reverse osmosis takes them out at the water plant)? Pipework, joints, plastic parts of taps, external inputs, ....? Nice talk, good to know what I`m drinking daily! Thanks!

# Name: Sam Cherniak

Q: Is it not the source water concentrations that would influence the final treated contamination? Is that what might influence the variance between the previous studies?

## Name: Verónica Godoy

How did you extract the microplastics from the samples? Thank you!

Very simple. The stainless steel filters were transferred into muffled glass beakers and incubated in 5 % SDS for 24 hours at 50 °C. Subsequently, filters were ultra-sonicated for five minutes. The filters were thoroughly flushed before being placed on a glass filtration unit. SDS was removed from the sample by flushing the beaker and the filtration unit intensively with particle-free Milli Q water and 50% ethanol.

# Name Joao Frias

Question: Excellent talk Inga, and excellent results. About the impact on human health (considered low), what was the reference of the report you mentioned? Thanks for it :)

EFSA, 2016. Statement on the presence of microplastics and nanoplastics in food, with particular focus on seafood, EFSA Journal. (EFSA Panel on Contaminants in the Food Chain), p. 30 pp. thanks :)

Name: Kristian Madaschi

Great talk. Could you please post the reference to the study about people ingesting 5g of plastic per year? Thanks.

https://www.sciencedirect.com/science/article/pii/S0304389420319944

Thanks :)

Name: Francisca Ribeiro

Q. Thanks for this lovely talk. Regarding fish, did you only consider edible portions (as the muscle) or other parts of the fish as the digestive tract for example? I'm guessing the risk of exposure only exists if there is plastic in edible portions...? ok thanks!

## Name: Kryss Waldschläger

Thank you for the interesting talk! Why do you only focus on the gut and so on and not the lungs? Shouldnt there be lots of microplastics?

Name : Margot Thibault

Thank you for your presentation, quite interesting !! For the future, will you study the quantity of particule oranic persistants fixed/added on this MicroPlastic ? To understand, the bad effect of ingestion.

# Questions and comments for the session 26.4\_Me

Speakers, please add your email address in front of your name for eventual further questions.

QUESTIONS TO Sarah Nelms s.nelms@exeter.ac.uk

Name:...Stjepan Budimir

Question 1:... Do you think for your research would active collection of floating macro litter be benefital, in a form of litter booms or litter collection vessels? Great work, non the less!

Name:... Adam Porter

Question 2:... Brilliant work and great to see such a blend of biology and socio economic investigations...really well done! I assume the threat scoring may be biased towards those species that have actual reports for them ingesting plastic or being known by IUCN...do you have any thoughts how to develop this really useful tool get round this

Name: Holly Nel

Thank you for this. How do your litter numbers compare to international values? Do you assume a lot gets removed every year through monsoon transport? Thank you :-)

For anyone who is interested, our study on waste fishing gear in the Ganges has just been published - https://www.sciencedirect.com/science/article/pii/S0048969720368364?via%3Dihub

QUESTIONS TO Jasmine Yu (jasminethea.yu@mail.utoronto.ca)

Name:...Coco Cheung

Question 1:...Thanks for your talk. Many studies use a combination of shape/ polymer categorisation (e.g. pellet, fibre, expanded polystyrene). What do you think about the mixed combination of categorisation?

Name: Holly Nel Thank you - How close are we to getting a "taxonomic guide" similar to what is used for invertebrate etc identification. Basically a key?

QUESTIONS TO Jennifer (Jen) Drummond (j.drummond@bham.ac.uk)

Name:...Christina Bogner

Question 1:...Do you consider density changes (due to ecocorona ect.) in your models? How does this change the exchange rates? Biofilm Thanks for your question. Biofilm growth at the sediment-water interface has been shown to increase particle retention. This was done through flume studies, but we have observed particle retention in cobble biofilms as well in field studies.

Name: Cynthia Munoz

Question 2:...Thanks. How do you recover the microparticles added to the stream. Do you measure the concentrations in the sediment, or how much of the tracer is left in the water at the end? Thanks for the clarification. Thanks for your question. To add to my previous response, we measured the particle counts in the water, sediments, and submerged vegetation over time.

QUESTIONS TO Annalisa Sambolino (annalisasambolino@mare-centre.pt)

Name:...Filipa

Question 1:...did you digested all specimens, the different species, together? Could you get information about the chemical composition of the MP you found? ok! thank you

Name:...Matthias Egger Question 2:...Great talk! Did you also look at diurnal variations/trends in terms of MPs/individual?

Name: Amedeo Boldrini, University of Siena

Question 3: Very interesting topic, thank you. Just a comment, maybe the increase of plastic amount in spring/summer could be related to the tourism rather than industries? Or as you said sea currents probably are the main responsible?

Hi Amedeo! There was no variation in the abundance of plastics in different seasons, but in the abundance of zooplankton.

Thanks for the answer, for the connection I misunderstood.

# Questions and comments for the session 26.4\_Ma

Comment for Barbro Melgert:

Name: Anna Winkler

Congratulations on your work, very substantial and relevant! We also work with lung organoids and fibres, focusing immune response (work in progress) and morphology effects. If you are interested, you can have a look at my poster 24.8\_Ma ;)

Hi Anna,

Saw your work presented indeed, very nice poster.Great to see your pictures, you have much more in depth study of the cellular interactions in those cultures, the works complement each other very well! Thanks, yes I agree ;)

We are currently chasing which leachate is responsible for the effect, not cyclic oligomers, we tested those.... and we find the same things with environmental fibers from fabrics bought in the store, similar to what you did, did you look at leachates of polyester? We did not... Not yet, but we should do it, always keeping in mind if natural polymers (cotton fibres) could have the same effect, so we want to test it as well. Gene expression should tell us more about leachate effects! We are planning to do this in the next step, also testing particles (not only fibres)

Yep, natural fibers are important to try as well. We did RNAseq of our epithelial cells and we found sooooooo many genes differentially regulated that I do not know where to start trying to tease out the mechanism :-) Maybe we will face the same issues ;) Have you published anything already? I would like to refer to your study/findings

No not yet, finalizing the manuscript as we speak, hope to deposit it on BioRxiv soon.... Name:...

Question 2:...

The fate of microplastics along salinity gradient and tidal cycles in a well-mixed estuary: a case study of the Seine estuary

Johnny : a nice video of sampling during the tide cycle here: https://wwz.ifremer.fr/lern/Projets-de-recherche/Pollution-plastique/Plastic-Seine-microplastiques

Name: Matthieu Mercier

Q: Do you plan to repeat observations over many tidal cycles, for reproducibility? Also how close is point 2 from the salinity front ?

Johnny: We conducted this on two cycle tide, and we observed a similar trend. A decrease of conc resulting from sediment sedimentation, previously remobilized during the flood tide

The salinity front is moving depending on the tide coeff and river flow, but station 2 is realy close to the front most of the time

Stjepan Budimir; do you think macro and meso plastic would behave similarly? Johnny : It's very hard to say. We observe accumulation of maro close to the max turb front too

Marie Babinot : Nice talk! Just a small question of methodology: what kind of filters do you use with  $\mu$ FTIR? Thank you :) We used stainless steel filters with the  $\mu$ FTIR Thanks !

Name: Tony Walker

Comment for Krista, Looking good at MICRO2020 Krista! I thought they had forgotten about you! I was afraid of the same thing! Nice to see eastern Canada being represented at MICRO2020. (Noreen Kelly)

Name: Noreen Kelly

Q for Krista: do you have any ideas about export of MPs from Bay of Fundy (to Gulf of Maine, say), given the tidal regime? Clarification: do you think Bay of Fundy is a source or sink for MPs? Thanks!

Name: Conrad Sparks Q1 for Krista, was this total weight or tissue weight of mussels. Very interesting research! Q2 Any Quality Assurance measures done? Kind thanks!

# **Questions and comments for the session 26.5\_O**

Claudia Lorenz: claudia.lorenz@awi.de or clo@build.aau.dk kevin.tallec@ifremer.fr

Questions to Adil Bakir:

Name:...Matt Cole

Question 1:... Hi Adil, great talk. Nile red is not specific to plastic. Are you able to account for the Nile Red staining of inorganic particles/organic particles that are undigested from particle rich/absent sites?

Hi Matt, sample prep is removing a fair bit (digestion and incubation step) of those materials and for some of the false positives the fluorescence is reduced and lower fluorescence is filtered out on the final excel produced by the automatic counting tool (Adil)

Name:..Marie Russell.

Question 2:...I am at early stage of using this method - do you have plans to automate the counting in any way - nice talk also and you have just answered this question

Hi Marie, we have created a simple tool using ArcMap but we are developing a new generation now. Happy to discuss iin more details! (Adil)

Name: Claudia Lorenz

Question: Thanks for the interesting talk, Adil. I was wondering what is your initial sample volume that you use per station in the centrifugation?

Hi Claudia, at the moment we are working with 5 g dried weight sediment in triplicates

Name: Filipa

Question (for Adil Bakir): Was it necessary to clean up particles (after nile red staining) before FTIR analysis? If so, how did you accomplished it? And concerning the digestion of the cellulose nitrate filter (i am not sure if this was the composition of the filter, sorry!), the digestion can eliminate completely the filter?

Thank you

Hi Filipa, the FTIR spectra were actually cleaner after the KOH:NaClO digestion process than before. Cellulose nitrate filter do dissolve in the digest during incubation (Adil)

Marie Babinot (Cedre, France): Thank you very much Adil for this very interesting presentation! Just a small methodological question about sampling: how much sediment do you sample?(We are trying to reduce because we are currently sampling 1kg)

Hi Marie, at the moment we are processing 5 g dried weight sediment in triplicates

Arnaud Huvet: Great talk, Thanks you Adil. Any trouble for some unwanted label from lipids or other constitants that may rest in the samples?

Hi Arnaud, yes some residues can be left after the digest process but more an issue for biota samples (we are using a similar method) than for sediments

Questions to Kevin Tallec:

Many thanks Jesus to give the opportunity to Kevin to talk (arnaud Huvet)

Name : Kyle K

Question : It looks like we have a Q&A session. The session is on which talk? I guess the session is Thursday 26th. 15h30-16h45 15h30-16h30, Ostrom's room. Session 26.5\_O Chaired by Jesús Gago, Vigo?

Yes

But I don't see the current presentation in the program

Speaker Claudia Lorenz, session 26.5:O

Thanks! I need to refresh/reload the webpage where the live rooms are located.

Name: Sara

Question: Thanks for a great talk and detailed study, Kevin. Can you tell us how did you measured the surface charge of PS beads, please? Hi Sarah, thank you! I measured the surface charge and aggregation state of nanobeabs using Dynamic Light Scattering.

Name : Arno Bringer

Question : Have you tested the effects of PS Nanoplastics on the fixation of pediveliger larvae ? And have you observed the formation of NP agregates on the mantle of D-larvae exposed at the embryonic stage ? Thank's Kevin for your great presentation.

Hi Arno! Thank you! Yes I used another assay to assess effects of nanobeads (and microbeads also) on the metamorphosis but I didn't observed an effects on the metamorphosis yield (proxy used in the assay for the settlement yield). Thank's Kevin. Maybe we could contact us to discuss on different results for potential effect of nano and microplastics on embryo-larval oyster stages.

Questions to Claudia Lorenz:

## Name: Joao Frias

Question: Excellent talk Claudia. What could be the sources of these paraffin waxes? Would you believe they result from recycling processes? Are they very common in other sampling campaigns? Also, do you believe that we need to improve our FTIR databases to reliably identify the paraffin waxes?

Thank you João! And thanks for the interesting questions :) So as potential sources we suspect discharge from vessels because paraffin wax is shipped in tanks and (un)loaded in liquid form. The residuals are removed by washing the tanks and should be disposed off in special facilities. Since the North Sea is heavily navigated entry from ships is most likely. For me it was the first time to encounter these particles in marine surface water samples (otherwise only washed up on beaches) but as I mentioned they could also be overlooked or identified as polyethylene (also their texture is more soft than other MP particles). Absolutely yes to your last question! Our databases would improve by adding paraffin waxes and blends into it! Sorry for the long answer (: If you are interested in details here is the link to our publication: https://authors.elsevier.com/a/1c1H5,ashxnC7

## Name Johanna,

thanks for the talk, Claudia. What are the reasons for the spatial distribution of the waxes as you found it?

Thank you, Johanna and good question. Since we expect cargo vessels to be the most likely source, the spatial distribution coincides quite well with the major shipping routes in the North Sea. Furthermore, at the station with the highest concentration of paraffin waxes (20) we also found the highest concentration of large microplastics that were in the same range as the paraffin waxes (500- $5000 \mu m$ ) which might hind to some local accumulation zone due to hydrodynamics. Thanks for the answer!

## NAME: Marte Haave

Question: Great work, as always, Claudia :) Do you expect any toxicological effect from animal ingestion of these waxes , and did you have them analysed for any other chemicals for that purpose? woull you say/suspect that these pose similar challenges and concerns as microplastics, or are they degradable in nature?

Thank you, Marte, for your kind words :) Unfortunately, we didn't analyze any other pollutants or added chemicals. However, paraffin waxes are known to contain benzene and toluene which are e.g. released when lighting a paraffin candle. As we saw partial melting and "leaching" of the paraffin particles on the filters (after drying them at 30 degree C) they seem to contain more volatile components which might also leach out in the environment. In the MARPOL context paraffin waxes are considered as "noxious liquid substances" and their discharge is now forbidden. I think they should be included in the regular monitoring for MP because they are as likely to be ingested as other MP in the same size range and a complete degradation of these will most likely take as long as for

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other MP. I think it would definitely be interesting to report findings of paraffin waxes along with MP to gain more knowledge there.

Questions to Gabriel C. Cardozo-Ferreira:

Name: Ana Lacerda, FURG- Brazil

Very nice presentation, Gabriel! How about fisheries being a big source of MPs at the region? Your data from reef fishes (e.g. dominance of transparent items, fibers, etc) is in accordance with what we recently found to several fish species (pelagic, demersal and demersal-pelagic) in South Brazil. https://doi.org/10.1016/j.envpol.2020.115508 Looking foward to read your paper!

# Questions and comments for the session 26.5\_Me

Name:...Peter Vermeiren

Question 1:...Thanks, interesting talk. Did the mortality event affect all age classes of birds. Are there differences in feeding preferences across lifestages

There is no audio

Name:...Anika

Question 2:.. Thank you for the great presentation. What do you think of solid state NMR as option for quantification ? Hello Anika, I've asked to more experienced colleague. He said that liquid (about solid he doesn't know) NMR looks promising but it is not so sensitive and you need huge amount of materials (in terms of grams). i hope this answer to your question

To Claudia Cella

Name:...Peter Vermeiren

Thanks for the presentation. Do you have any suggestions on how to avoid aggregation of materials on filters. Thanks for the valuable insight.

Name:...Peter Vermeiren

Are there incentives by the Malaysian to raise awareness on the emission of microplastics from washing machines, and to change consumer behaviour. Nice research. Great to hear the newspaper interest.

Hi Peter. There is no any incentive. This is a new information for the public as well as policy makers. Thank you for your view.

## Name: Camila Vidal

Thanks for the talk. Could you find any correspondence between what you saw from the washing machines and what is present in freshwater courses that recebe this discharge?

Hi Camila. Our research projects are still in progress to see the similarities between microplastics in laundry water & wastewater treatment. We have few difficulties to obtain wastewater treatment plants samples are the reasons for this delay.

Name: Claudia Heller Question to Sarva Mangala: Have you also found cotton fibres? What was the ratio to the man-made fibres? Hi Claudia. Yes we found cotton fibers in our laundry samples, about 29%. However, we didnt calculate the

man-made ratio. We reported as whole in percentange (29%).

Thank you! Did you also publish your findings as paper? Would be happy to read it! Yes Claudia. Thank you for your interest :). This is the link - https://doi.org/10.1007/s11356-020-10795-z

# Great, thanks!

Name: Julia Taylor

Not a question, but I wanted to inform about an international innovation competition - Zero Microplastics Challenge 2020. https://www.ri.se/en/what-we-do/projects/zero-microplastics-challenge-2020 The competition aims to reduce MP from textile washing. The competition is funded by the Swedish EPA.

HI Julia. Thank you so much for this competition information & link. My research group will read further about this. Thank you again :)

# Name: Victor Onink

Thank you for your presentation Peter, it is very interesting how you see such differences between different types of coastlines. What do you think causes such patterns? It is the difference in how likely plastic is to beach, or how likely plastic is to be washed off teh beach? Or is it local inputs that are the main driver, and morphology has a smaller influence?

Thanks, as I mentioned, it is likely the influence of a neirby channel that influences the plastic levels. Both sites were disspiative beaches with very gentle morphology and little tidal movement.

## NameL Trang Nguyen

Thanks for your talk. Could you please clarify the automatic analysis you used for identify MPs? Thanks, I hope I answered your question, you can also check the paper or email if you have more questions (p.vermeiren@science.ru.nl) https://doi.org/10.1016/j.envpol.2020.114298

## Name: Elena Hengstmann

Thanks for your interesting talk, Peter. I have a question concerning your staining method via Nile Red: Which solvent and concentration did you use for the Nile Red solution? Totally fine! If it is the Maes protocol, I'm familiar with it. Thanks for the question: 1 mg Nile Red dye in 1 mJ 99 5% acetone and diluted with 100 mJ

Thanks for the question: 1 mg Nile Red dye in 1 mL 99.5% acetone and diluted with 100 mL distilled water (based on Maes et al., 2017). The protocol was published recently https://doi.org/10.1016/j.envpol.2020.114298

# Questions and comments for the session 26.5\_Ma

denas (alAlicia Mateos Cáricia.mateoscardenas@ucc.ie) https://www.nature.com/articles/s41598-020-69635-2 Name: Aline Carvalho Question 1: Thanks again, Alicia! About the microplastic shapes, how did you categorize them?

Questions for Kennedy (k.bucci@mail.utoronto.ca)

Name : Alice Horton

Did you measure, or do you have plans to measure the types of associated chemicals and additives that might be causing the effects? Thanks for this question! Happy to discuss further if needed :)

to Bucci - what is an "ecotoxin"? That's a new term to me. Do we need it? We used "eco-toxin" in Rochman et al. 2020 for all the sorbed chemical contaminants from the surrounding environment. I tend to use "sorbed environmental contaminants" but kept eco-toxin in the first figure i presented as that's what it is called in the paper :)

Aline Carvalho (carvalho@chimie.ups-tlse.fr) thanks :)

To Mariacristina Cocca from Judith Weis

A talk a few days ago showed that most of the microfibers in the environment are actually cottoon and not plastic. Have any studies been done on shedding from cotton fabrics while they are being worn?

# Questions and comments for the session 26.6\_O

----Jonas:

----Hello Marillou thanks you, have you ever used the babylegs method before ?

-----Thanks you for this answer, one last question: DO you know what type of MPs is in the samples ?

yeah same here! Thanks for the tip -- i tried that, but i think my connection today is just slooooow... thanks for your patience and help everyone ! :) -- (From Bethany Jorgensen) Probably it's Thanksgiving Day her in USA! :)

Name:...Catharina Pieper Question 1:...Thank you for your talk. I was wondering what was your sampling method to collect stormwater? thanks! Thank you! We used an ISCO depth-integrated sampler. - Alice

Name:...Niels Mast Question 2:...Thanks Xia for that. Did you find >5mm particles in stormwater. If so, can you tell about it? Thank you! :) The vast majority of particles in stormwater were < 5 mm, but we did find the occasional macroplastic particle. - Alice Alice !! (NM:)

Name.... Jonas Question: Marilou, Have you ever used the babylegs method before this study ?

(To Speaker: Sluka Robert Plastic pellet pollution on florida's space coast – a citizen science approach ) Name:...Kyle K

Question:...Great to hear about what you and your colleagues have been doing. I am located in a little north in Volusia County. Definitely want to do similar type of work up here to compare with areas between north and south of Cape Canaveral! Hi Kyle! Would love to connect. My email address is bob.sluka@arocha.org

Thank you, Bob! I would love to. Happy Thanksgiving, BTW!

Name:...Diego Lelis (Federal University of Rio de Janeiro, Brazil) Question:...Great presentation! Could you talk a little about A ROCHA? Thank you. Happy to connect offline - you can see some about it at www.arocha.org Thanks :)

Name:...Catharina Pieper

Question:...Thanks for your talk! do you see differences on pellets degradation states found on the beaches after hurricanes?good question! Unfortunately, I didn't look at this.

Name: Ghezali Yousra

Question: Thank you for sharing with us!

what do you think of the fluctuation between the amount of microplastics found in the summer and winter season!!

I think it mainly is due to differences in wave patterns - much calmer in the summer and also no hurricanes. So storms seem to be important. Thank you, so mainly the amount of microplastic found in winter is higher that the one found in summer season!

Name: Thomas Maes

Q to Marilou (not really a question): Great presentation, I like the babyleg net and appreciate the topic: opening up microplastic research for citizenscience! It would be great to test its applicability in developing countries, here is my email: thomas.maes@grida.no

Name: Diego Lelis

Question: Marillou great presentation! Do you think that the type and size of the microplastic can vary with the period of the collection day? Thank you, our sampling campaign was carried out over only two days, it would be difficult to analyse the variation in the shapes of the microplastics. Many factors come into play, such as the types of rejects, distance to the source, UVs... Thanks :)

Vesna TEOFILOVIC

I like the idea of micro knowledge network Let me know how can I join it? You can have a look at our website : https://lapagaiesauvage.org/ We only accept samples collected in France, but we encourage everyone to develop this initiative in other territories, if you have any further questions you can send me an email at contact@lapagaiesauvage.org

From Bob Sluka - great presentation! How do the citizens give you the samples?

# Questions and comments for the session 26.6\_Me

Contact speackers : margotthibault@orange.fr

Questions for Leonardo: Plastic litter in the Patos Lagoon estuary and adjacent coast

Questions for Brenda and Karen: Microplastics monitoring for the implementation of public policies within the Cozumel Island Biosphere Reserve

#### Name:...Ana Lacerda, FURG- Brazil

Question 2:...Thanks for your talk, girls! :) Karen 1) have you found paint particles (probably from ships) in your samples? If so, have you considered these particles as floating plastics and put it into the Fragment category or excluded it from the analysis? 2) Are you planning to show the results in both number of particles and also weight?

Thanks for explaining it.

Karen, I've asked about paint particles because this is a controversial point on considering it as floating plastics or not (Many authors consider it as fragments, while others exclude it). This can lead us to problems on comparing the concentration of floating plastics worldwide. There was a presentation this week, (Clara Leinstenschineider), for example, where the authors did not consider paints together with "plastic particles". We also found 30 times more paints over "plastics" in Antarctica and considered it as different categories. Some of the paints were from our vessel (FTIR analysis comparing what we found in the water and paints from the hull and deck from our vessel), but others we suggested that were from other sources. You can have more details on https://doi.org/10.1038/s41598-019-40311-4

Ana, Gissell Lacerot here, CURE-Uruguay. Sorry for jumping on your question, but thought it was really interesting. We are also finding paint in Antartica (plastics resins), on marine samples, land, streams and ice. But we currently do not exclude it form the analysis (in our case it does not come from sampling ship). I was having this conversation with another colleague yesterday (Kirstie Jones-Williams). She took them out because it came from her sampling ship. Why did you decided to take them out?

Ana here. Hi Gissell! I did not considered it as floating plastics because I know there are some estimatives where people don't consider it as floating plastics due the potential source being their vessels, and the high density of those particles, so they should not be at the sea surface, BUT they were there problably due water tension or the properties that were altered, etc.... anyway, I decided to report it in the Antartic paper and not to keep it completely out of the results. But I guess we need to bare in mind that we should, at least, highlight what type of fragments we are considering as floating plastics.

Thanks for answering!, and for the recommendations. Yes, I agree paint might need at least a subcategory. ok, good. I already know your publication, but I will definitely give it another look. And please send me an email or a message on ResearchGate if you need anything! I'll be happy to discuss it.Great!, thank you!

Clara's presentation was very interesting when she showed that FTIR and X-ray were giving different resolutions. We have used FTIR, but now she improved the analysis using one more tool and based on that I guess we need to consider it in the future to estimate the sources of paint particles...

Questions for Margot Thibault: First characterization of marine litter collected on Fort Dauphin and Sainte Marie Island, East Coast of Madagascar

#### Name: Jakob Cyvin, NTNU, Norway

Question 1: Thanks alot for an impressive presentation! I'm vondering if you are thinking about using video analysis or photo analysis to do seasonal analysis of the areas?

-or in more general remote sensing?

I can email you - sorry :-)

Yes that was what I ment Alex

Thanks for the answer - the reason for my question was papers I have written from Japan, where stationary cameras tracked the amount of plastic on the beaches every day threw the year using automated object recognition to quantify the amount washed up.

Margot : Alright, I understood ;) For this first marine litter monitoring in Mada, we needed to have the brand of each litter identified :) so we needed to collect all marine litter beached.

Jakob: Thanks - I did not think about that part.

#### Amy Lusher

Thanks for a fantastic presentation, i really liked the way you put your video together it was so clever. One brief question, do you plan to look into different beaches, and i would be interested to see if you take into account other variables, such as the seasonality (jakob just wrote the same) .so second question, were you able to differentiate plastics types, or do you plan to do this in the future? Thank you for the clarification :)

#### Romain Tramoy LEESU, UPEC, France

Thank you for this amazing presentation. Do you have an idea of the quantity of litter per km? Or other quantitative data to enable comparisons with others? Sorry if i missed the information, I took the presentation on the way attracted by videos :)

Ok thank you :). Waiting for publication ;) Margot : Yeah, thanks for all your questions :D We are writing it :)

Questions for Jakob Cyvin: Citizen science for microplastic detection and analysis - A comparison with other fields of science- reasearch limits and possibilities in the field of microplastic.

# Questions and comments for the session 26.6\_Ma

Speaker: Raab Patricia Students' conceptions on the subject of microplastics - Raab Patricia, Bogner Franz X.. Paper number 334319, cc-by-nc-sa.

Name: Antje Wichels...

Question 1:...thanks Patricia, very nice talk, great results. May be I missed it, what was the diciplin of the students, since they were 20 years, they are already in an university

## Name: Maja Grünzner

Question 2: Great presentation! Are you going to follow up on these results e.g. compare the german students with students from other countries? / Thank you Patricia. It is great to see that you investigate the knowledge gaps before developing new information material.

Lisa Zimmermann:

Great and interesting talk! Which disciplines (natural vs. social science) did the students belong to? 2. Question: And what semester?

#### Elena Hengstmann

Are you going to ask these or others students again later to see whether answers will change with time?

Thank you!

#### Julia Pawlak

Q: Thanks for your presentation! Do you plan to perform more surveys with different target audience? Like younger or older people, different education status etc.?

## Stephan Rohrbach:

Very interesting talk! Whats your opinion about the current knowledge of your asked students? Do you think the public should include this topic more in school or do you think the level of information gained from the other media are sufficient? Especially when comparing with other environmental topics?

Thank you for your question. In my opinion, the topic should definitely be given a higher priority in education. Media reports are often simplified or misrepresented. In school or university, pupils and students receive first-hand information. Compared to the probably biggest environmental topic of today, climate change, which has already found its way into schools, it is unfortunately not the case with the ecological consequences of microplastics.

Speaker: Winton Debbie dwinton@earthwatch.org.uk

Enabling individuals to understand, act on and reduce their microplastic footprint - Winton Debbie, Loiselle Steven, Marazzi Luca.

Paper number 334363, cc-by-nc-sa.

Name:...Johanna Kramm Question 1:...Is this tool available as app? The plastic on the go footprint calculator is available online, it is mobile friendly, but not an app https://plasticfootprint.earthwatch.org.uk/

#### Name: Delphine Lobelle

Question 2: Do you check whether the same products that are packaged in glass or cardboard versus plastic have a better overall footprint (carbon etc)? Amazing! Thanks for sharing that publication :) My email is d.m.a.lobelle@uu.nl- super interested in your project! Thank you for your interest, I will get in touch! Our study on this is published in PLoS One and available open access here https://journals.plos.org/plosone/article?id=10.1371/journal.pone.0236410 COmment: I think a huge impact would be achieved when reusing plastic materials which are not meant to be reused instead of buying freshly produced alternatives (plastic bag - cotton back for instance) (Stephan) Thanks Stephan, that is definitely an action we need to consider!

Ryan Shum: Really fantastic idea! Is there scope (or plans to) expand this tool for use by businesses and industry? Thank you! We hope this will be very useful for businesses, but more from application of the results we find, rather than for them to calculate their microplastic footprint. Though it could potentially be adapted for offices

Lisa Zimmermann: Very nice idea and presentation! Does the tracker only target microplastic release or also "larger" plastics? Thanks! The current tracker looks at macro plastic used "on the go", whereas this new concept focuses solely on micro as this is a real gap in footprint calculators/trackers

## Name: Tony Walker

Question 1: More of a compliment to Debbie Winton. Great talk. I never even knew there was such a thing as an: ONLINE PLASTIC FOOTPRINT CALCULATOR. Thank you for enlightening me. Looking forward to learning what the MP concept will achieve. Thank you so much! I hope you can have a look at the Earthwatch calculator and find out your own footprint! https://plasticfootprint.earthwatch.org.uk/

Speaker: Kramm Johanna Public risk perception of microplastics - Kramm Johanna, Völker Carolin, Werschmöller Simon. Paper number 334481, cc-by-nc-sa.

Name: Christoph Habel

Question 1: Thanks a lot - really interesting results. Are there any hypotheses why the educational background seems to play a role in the popularity of microplastics, e.g. the use of other media?

Name:...Gisela Böhm Question 2:...What was the full range of the rating scales?

Speaker: Miguel Isabel (no video file received to date) Plastic pollution: Overall knowledge, perceived impacts, and pro-environmental behaviours - Soares Joana, Miguel Isabel, Venâncio Cátia, Lopes Isabel, Oliveira Miguel. Paper number 334592, cc-by-nc-sa.

Name: Maja Grünzner

Question 1:Thank you Isabel for your presentation. Behavioural Science shows that societal factors (e.g. social norms) influence environmental "harmful" behaviour such as littering. Do you think that people could be biased when they answered that seeing the neighbours acting "pro-environmental" is not motivating? Hope that makes sense.

To all: Do you think individuals who are already motivated to change should be targeted or should "we as researcher" should try to target less motivated people?

Tony walker: An interdisciplinary session would be great YES!! (Maja)

Debbie Winton: Hi Isabel. In your talk you mentioned a study that found that social norms are more effective as an incentive to change behavior in younger demographics. Would you be able to share the study you were referring too? i'm really interested! Thanks!
## **Questions and comments for the session 26.7\_O**

Speaker: Hink Linda

Effects of different microplastic polymers on the terrestrial woodlouse Porcellio scaber and the associated gut microbiome - Hink Linda, Holzinger Anja, Bernstein Alina, Sandfeld Jensen Tobias, Schramm Andreas, Feldhaar Heike, Horn Marcus A..

Name:...Christina Bogner Question 1:...Thank you! I probably missed it. Did you mix the soil with MPs? Does % refer to soil fresh weight? It was not soil. And I didn't mention it :-) The mixture contained leaves, potato starch, and rabit food with and without x% MP (w/w) Thanks, this sound jummy :-). How large were your MPs? 75-120 µm That's cool. We use this size already for soil essays (B06). So, we can be sure that this size is "relevant" for soil dwellers. Thank you! You are welcome :-)

Speaker: Schirmer Elisabeth

MALDI mass spectrometry imaging in aquatic model systems - Schirmer Elisabeth, Treu Axel, Ritschar Sven, Schuster Stefan, Laforsch Christian, Römpp Andreas.

Name:...Christina Bogner

Question 1:...Thank you! Is this a "preparational" study to look into possible leachates from MPs? Or did I simply miss the MP link, sorry in this case.

Thank you. Yes you are right, this is a preparational study. Maldi MSI enables later on the detailed information on molecular changes in the mentioned samples

Can you later see the particles themselves, in the tissue?

No

How will you link to MP? :-)

we link it to other methods, e.g. microscopy

Ok, so exposure to MP, added by microscopy. I think, I got it. Thanks

Speaker: Cachot Jérôme

Contamination and effects of microplastics in the Seine estuary's food web. First results from the Plastic-Seine research project - Cachot Jérôme.

Speaker: Carreras-Colom Ester (contact info: ester.carreras.colom@uab.cat // maite.carrasson@uab.cat)

A European assessment of plastic ingestion in the Norway lobster (Nephrops norvegicus) -Carrassón Maite, Carreras-Colom Ester, Cartes Joan E., Rodríguez-Romeu Oriol, Constenla María, Welden Natalie, Soler-Membrives Anna. Speaker: Justino Anne (If you require any further information, please feel free to contact me at anne.karen@hotmail.com ! Thanks)

Ingestion of microplastics by fishes of an estuarine trophic chain in the Western Atlantic - Justino Anne, Lenoble Veronique, Pelage Latifa, Ferreira Guilherme, Passarone Rafaela, Frédou Thierry, Frédou Flávia.

Speaker: Esiukova Elena

Microplastics in marine macrophytes on the underwater slope of the Sambian peninsula (the Baltic Sea) - Esiukova Elena, Lobchuk Olga, Volodina Aleksandra, Kupriyanova Anastasia, Chubarenko Irina.

# Questions and comments for the session 26.7\_Me

Name:...Lígia Santana Question 1:...For Ana Patrício, why 3 hours of exposure? Is it a new protocol that you are developing? Thank you for th nice presentation!

# Questions and comments for the session 26.7\_Ma

Name:...Robby Rynek Question 1:...Did you directly take the aliquots from the sample or did you homogenize it before?

Name: Trang

Question 2: What the amount of reactants you used for Fenton reaction? how is the effiiency of this treatment step? Which solution used for density separation? Do you have any suggestions to analyse fewer aliquots but get representative results because analysis of all samples will take a lot of time Thank you, I would like to read your paper!

For Lisa:

Cleo: you show in the results different MP sizes smaller 0,335mm but say in the methods you filtered it on 0,335mm. So have you looked also on smaller MP? Thank you, great work also.

For Lisa

Trang: in your opinion, which sampling method should be used for moving water bodies (e.g., channel, stream, etc) with effects from peak events like runoff/stormwater?

Contact info for Lisa Watkins: ltw35@cornell.edu

## **Questions and comments for the session 26.8\_O**

#### Name: Maria M

Question 1: For Valentina Fagiano: Thank you for the presentation. Do you think these findings could have an effect in the zonation of the MPA and in particular, the no-take zone? Thank you Thank you very much for your question. We did not find any difference between locations subjected at different protection levels. The highest MPs concentration was detected at Estels within the no take zone. MPs composition suggest far contamination sources as predominant. Data underline that even notake zone within MPA are not sheltered from MP.

#### Name:...Stephan Rohrbach

Question 2:...Suggestion: Would be really cool if you also check for Antibiotic contamination/ quantities in the respective sewage; analysis if antibiotics are adsorbed to the different types of MP. Very cool project.

Thank you very much Stephan, that's a nice idea, we would need a collaboration for this since we don't have the tools to analyze antibiotics but in Barcelona there are some groups doing it and I will try to include it. Thank you very much!

Jessica Song (Session 26.4\_O today) had a pretty similiar project regarding rivers in Germany. You can have a look maybe she faced some obstacles and can collaborate with you in some aspects.

Hi Jessica thank you very much. It must be a nice project then. Unfortunately I was lecturing and couldn't follow up many talks. I will check it an contact you (my contact is eballeste@ub.edu). **Questions and comments for the session 26.8\_Me** 

Name:...Stjepan Budimir, question for Gulizia Alexandra poster: Chemical digestion methods: What are the impacts on microplastics?

Question :... What was the concentration of the digestive reagents in the end solution. so 10 or 30?

Hi Stjepan, We used between 10-30 w/v% depending on the reagent. We have a publication impending which will detail this much better ;)

thank you!

Name: Amedeo Boldrini, University of Siena

Question 1: Thank you Agnes for this interesting poster. Did you also growth the algae in a free particles environment to explore effects of particles enriched vs free-particles systems? Sorry but I missed part of the presentation and maybe you have already said that.

Thank you Amedeo for your question. Yes, we had reference conditions with no NPs in both experiments to compare with the "contaminated" conditions. Did I answer to your question? Yes, thank you. I would add what are the main differences? Did particles promote or not the growth? Not at all (at least in these 2 experiments), we did not see any difference in algal densities at the different times whatever the concentration of environmental NPs was.

Thanks again, very interesting

Thanks for your questions

Name: Sara

Question to García Ordóñez Marlid: what was the concentration of MPs you used? 25 or 50ug/mL? sorry, got a bit lost. great job with the fluorescence detection.

To Roger Sola: Matthias Völkl: Are the cells you are using primary cells or cell lines?

## Questions and comments for the session 26.8\_Ma

To Berit: Name: Cleo: Question: Very nice work. thanks for the poster. Did you see effect of density on the organic/inorganic matter settling?

Name:...Matthias Tamminga to Bjarne

Question 1:...Thanks for the interesting talk! Could you give further information on the specifc staining protocol you used? Thank you for the interest, please send me an email to bjarne.kvaestad@sintef.no so I can forward you to the correct people

Name. Elena Hengstmann to Bjarne

Question 2: Thank you, Bjarne! Did you try your automated detection also with environmental samples? In this case organic matter resiudals might hamper the correct detection process. **Questions and comments for the session 26.9\_O** 

For Sarmite: very interesting study: have you found many cellulosics fibers in you samples? -no, actually none ;), because we conducted Fentons oxidation which eliminates all natural fibers Ok great! thank you!

## Questions and comments for the session 26.9\_Me

Name:...Matthias Völkl to Joanna Gonzales

Question 1:...Thanks for the talk :) Which media did you use for the in vitro incubation? Yes thanks. Hi, thank-you so much. We used Dulbecco's modified eagle medium. Hope that answers your question.Yes thank you so much :) 10% FCS i guess? The DMEM we used has 4.5g/L D-Glucose, L-Glutamine and 110mg/L Sodium Pyruvate. I hope that helps.

Name:...Nina Paul to Joanna Gonzales

Question 2:...Thanks, Joanna, really interesting results over the 21 days! Do you plan by any chance to look into the energetic balance to probably get insight in shifts of energy towards e.g. repairing? Thank-you. It is something we are thinking of to better understand the results we obtained at 21 days. Do you have any recommendations? E.g. glycogen content/energetic reserves. Would also be nice to see if there is kind of metabolic arrest going on, maybe shifts in strategy, from kind of "acute response" to overdue the unfavorable conditions, but then come into more "semi-chronic" conditions.

I am actually working on similar analyses, so we may discuss and exchange via email in detail, really looking forward to! :) nina.paul@awi.de thank you, I would love to. my email is jmgoncalves@ualg.pt

Name : marte haave;

Q: thanks for a lovely presentation, do you think you might have had the same response and adaptation if exposing to a a control particle? could it be that they are stressed by the particulates? We also tried sand particles, also we got no response. Very similar results to wood particles. Thanks, this was actually a question for the previous speaker :) :)

Name : Marte Haave

Q: great work, love that you used a control particle. Would be very interesting to see a long term exposure. ANy thoughts about the outcome? Whats the lifetime of woodlice?

I am not sure, but it's few years.

so a long term (several months) is possible?

It is , but they do not really like the jar/soil exposure. Of course it is matter of optimising the protocol.

what kind of effects would you expect? can you make a "best guess"

Well, I know. Thinking. Perhaps it goes back to zero. Yes, or changes in cuticle causes inflammation difficult, I know.. just interested to see the adaptation.. but perhaps inflammation over time, or agglomeration of granulocytes with NP?

We actually do not expect these large ones passing into the haemolymph

sorry, didnt you say that it caused a highr level of granulocytes.. the picture wasn't clear..

Yes, but the increase was caused indirectly- meaning MP is not in direct contact with granulocytesthe change is caused from the gut lumen.. at least that is our hypothesis based on studies with nanomaterials..

interesting.. so they react to the "invasion"

we think they react to changed gut environment, there are studies that show animals react even to change in diet.

thanks great! Good presentation.

Thanks, at least I had some interaction during these 4 days:):)

MP transport in rivers and streams, feel free to contact me its jan.boos@uni-bayreuth de. Cheers

## Questions and comments for the session 26.9\_Ma

Name:...Ana Catarino

Question 1:...Zhiyue, interesting work! On the size distribution graphic, do you predict that the smaller sized microplastics would be less or are they more difficult to detect and that would explain why their frequency decreases?

Question to Zhiyue: Name:.Hazimah Question 2: Nice talk. Were there any differences in the size distribution before/after exposure? Secondly, what type of plastic specimens did you use (pellets/film) and do you think the type would affect your outcomes? cool. thanks. Question to Fleurine:

Thank you for your presentation. What plastic samples are you intending to analyse for phthalates? Nice! All the best!

Name: Ana Catarino Question presentation 2: Will you prepare a library of the times of detection of each additive? available for other researchers, yes, retention. thanks (ahhhaahah, thanks!)

Hello, thanks for your question,

in our data base, we will add the retention index (so they won't change depending on the matrix) for the availability of the data base I don't know yet. i actually don't know the process for "sharing" a data base. but if/when the method will be validated i hope that it will be possible. That's great, thanks! you welcome !

=> I can also share this database if you contact me directly by mail : fleurine.akoueson@anses.fr]]

Hazimah:

Asta, what do you think is the main source of these particles you found in the sediment column?

Stephan: Hello Asta very interesting and scarying results. Have you also tried to analyse icecores from glacial areas and if yes have you found similar findings? What size ranges have you found?

Ana Catarino: Ola Marina, very interesting work! Do you think that sediment transport can be responsible for pellets transport as well? And that's why the deposition is continuous? Sera transporte por sedimentos? (portuguese!)

Yes! Thanks! That's it! Obrigada Marina, era isso mesmo, obrigada pela tua resposta!

Thanks everyone! thank chairman

Marina Zimmer Correa:

Ana Catarino, thank you so much for the question! The sediment can transport and also bury the pellets. The pellets reach the beaches as they are transported by ship, and the loss of this material occurs during all the year.

Day 5/5 Pads

## **Questions and comments for the session 27.1\_O**

Name:...

Question 1:...Miguel Tamayo: Did you feel the people there awar of the plastic problem? Thank!! Francesco

Name:...Chloe Question 2:...Did you use chemical analysis to confirm particles were plastics?

arnaud Huvet: Great talk François. On what types of samples are you planning the ddPCR (plastics, natural debris, water... ?

## Questions and comments for the session 27.1\_Me

Name:...Katrin Wendt-Potthoff Question 1:...very nice work! Is there any quantitative information about the moecules/biofilm?

Questions for Alexander Tagg:

#### Name: Gholamreza Shiravani

Question: Could you tell me how deep were the sediment samples (peneteration depth of Van-Veengrab sampler in sediments/bed)? and the area of taken samples (covering area of each sample)? Thanks.

#### Winnie Courtene-Jones:

Question 1: Thank you for this presentation, i am really interested in your FTIR anlaysis of the paint particles. are you using polymer libraries to assist in ID? or have you made your own? i am interested to know more about how to confidently ID these in samples, if possible to chat more more contact is winnie.courtene-jones@plymouth.ac.uk

Question 2: Also, do you think the main driver of the differences of microbial communities is due to the metals in the paint flakes?

- it's hard to compare i appreciate. The way i understand is that every paint manufacturer has a slightly different formula, so there's a huge diversity of 'types' of chemical formulas of the paints. Very interesting work to dig into for sure! i will watch for the results with interest :) Tagg: Yep that's basically it. Pigments are pretty broadly used, as are resins, but the specific combination of additives (which typically includes metal oxides) are very unique. Plus, we have to consider these particles have been in the environment for some time and may have been fundamentally changed by oxidation etc since manufacture. The best way would be to use a huge variability of paints with some overlapping similarities to pin down which attributes are causing fuctional change. Essentially this is where I will start next

Stephan Rohrbach

Did you have to pre-process the particles prior to SEM-EDX like get rid of the organic matter beforehand? And if yes, how did you do this? Thank you

Katrin WP: Question to Alex: can the metal content be beneficial for the SRB? H2S detoxification? Roxane Danquigny:

Did you consider analysing metals via ICP-MS, in order to have a quantitative information? Tagg: Honestly not. It was a very exploratory study. We started with the FT-IR and raman, and moved on to EDX when these results were inconclusive. Ultimatly yes, a mass spec approach would be ideal (I considered SIMS) but we just didn't have enough particles to warrant further work; the sample size was just too small to really get to the bottom of something this complex. The good thing is I'll be researching this further and mass spec will definitly be a useful appraoch going forward; I'll look into ICP, thanks for the suggestion

Questions to Jean-Francois Ghiglione

Miguel Tamayo: Thanks for your presentation, which types of organisms colonize the PCL? Did you observe changes in the population that could be related to toxicity processes triggered by the PCL by-products? Thanks a lot, i'd like to make you one more question regarging the organisms that colonize PCL, do any fungi specie appear?

Katrin WP: How could you ensure sterile conditions in the treatment without bacteria? Can the particles be sterilized?

Erika Cedillo: Thanks for your presentation and interesting work! How did you measure oxygen generation? Can you say something more regarding the experimenting setup to perform thsi measurements?

Antje Wichels:

Ouestion: Did you make any attempts to enrich or even culture bacteria to obtain frew and new isolates... very diffcult but I find it superimportant?

Miriam/HYDRA: Great data, thanks for sharing! What are your plans to include ecotox testing in your testing scheme? We are very interested to understand the effects of the intermediates / oligomers.

Cheers, Jean-Francois!

Stephan Rohrbach: Interesting talk! Sorry if I missed it, but can you specify what is your plan regarding your [3H]-aminoacids. Its impressive that apricots seem to be pretty durable, do you know if there are studies which tried to use them as substitutes for microbeads in common applications?

Name: Darshika Manral

Question: Great project! Coming from non-experimental background, I am curious to know the protocol for disposal of plastic based compounds post experiments?

Name : Juliana Ivar do Sul

Hi Lara! What is the implication of these communities on plastics to the salt march environments? (maybe good to check if there are works with other (natural?) particles in the area to see if the plastics are playing an important role!)

Lara: Hi Ju! Thank you for your question. We are planning to perform an field experiment by incubating plastics in the salt marsh to check which organisms will adhere and then check for any implications such as changes in plastic density/buoyancy or presence of environmentally important groups such as patogenic microorganisms etc. Also there is another work in progress in our group looking specifically at how biofouling affect plastic density/buoyancy in the Patos Lagoon so this information can be used for modelling plastic dynamics in the lagoon.

Stephan Rohrbach:

Thank you Lara for your talk: You conclude a lot from the environment to the extent of biofouling, but could it be that the time the plastic debris are in the area might have a great impact as well? Any plans to differentiate here?

Hey Lara hope you are still here:) My question was a bit misunderstood. So I rather wonder for instance you found only low biofouling on the fibres in the wet area and on the particles in the high marsh, but a lot biofouling in the mud area, right? The microhabitat definetly have a great impact on biofouling, but as far as I understand it, you have no idea if for instance the particles in the mud have been there for 2 years, whereas the tested fibres have been there for only 1 month? So maybe the time component is underestimated in your study. So maybe if you have the ressources you could try to differentiate between these two parameters

Lara: Hi Stephan, thank you for your question. Yes we think that time will play a role in the formation of the biofouling community so we are planning a field experiment to "watch" the biofouling over time at the different zones at the salt marsh. Perfect looking forward to see how this goes

Thanks!

Name: Kristian Madaschi Do you know if surface texture plays a role? Thanks, Lara :) Lara: Hi Kristian, thank you for your question. I think I answered your question on the live talk but yes, we do think (and there are evidences) that texture plays a role.

# Questions and comments for the session 27.1\_Ma

1. Christoforou Eleni - Effects of long-term exposure to microfibers on ecosystem services provided by coastal mussels Name:...Matt Cole Question 1:... Nice study. I missed what concentration of fibres you used, how'd this compare with environmental levels?

Name:...Charlene Trestrail Question 2:...Great talk, Eleni! How did you choose the ratio of fibers to Tetraselmis? Does this ratio change the mussels' responses?

Name: Camille Détrée

Question: Nice study!! Did you checked the size distribution of your MFs?

2. Michael Sturm - The potential of fluorescence dyes – Comparative study of Nile red and three derivatives for the detection of microplastics

Name:...Antje Wichels

Question 1:...Thank you Michael, very important results. Did you also try dilutions of the solvents, e.g. we had good results with a diluted EtOH (25%) we have tried a dark reader with blue light (440nm - 510nm), which worked pretty well. Did you also use other light regimes which might be comparable?

Name:...Alexandre Dehaut

Question 2:...What about the use of your new Nile Red to KOH treated samples? --> no pH issue?

Ok , thanks!

Name: Cleo STratmann Question: Thank you, very interesting! Is there a library for Nile red spectra to compare to standards, or do you that yourself? Which software you use?

Name: Eleni Christoforou Questions: Could Nile red dye help in the differentiation between organic and inorganic microfibers? Would they have a different absorbance? THank you!

3. Natalja Buhhalko - Sources of microplastic pollution and amounts of microplastics in zoobenthos and fish in the Baltic Sea. Name:...Marte Haave Question 1:...sorry, missed the size-range of the MP: could you repeat, please? Thanks. and nice presentation by the way :)

did you not find any PVC by this method? if i saw the results correctly they were not present..?

Name:...Marie Russell Question 2:...no questions - just to say nice presentation - lots of data - will you publish this at some point

Name: Julia Taylor Q: Did you analyze the outgoing water of the WWTP?

4. Rachel Coppock - Benthic fauna contribute to microplastic sequestration in coastal sediments Name:...Gholamreza Shiravani

Question 1:...Thanks for your great talk. Could you tell me which type of sediments did you use for experiments (sand dominant or silt/clay dominant sediments (mud))? and what do you meen with deeper layer? how deep ?

Name:...Alice Horton

Question 2:... excellent presentation! If some organisms transport microplastics upwards and some downwards, what do you think is the overall effect of the whole benthic sediment community - upward/downward/neutral?

Name:...Marte Haave

Question 1:.excellent presentation, great work. So... which sediment depth would you say, represents the recent deposition best? (monitoring concern). Is layering of small slices really representative, or is 0-5 cm best to show the input over last year? do we need to know the benthic community to decide, or can we take an average? (finished:) thanks a lot, Rachel:)

#### Name: Simone Lechthaler

Question: Thanks for the great presentation! Is there an approximate (or maximum) transport capacity through the organisms that can be estimated? I wanted to ask if it is possible to say how many millimetres/centimetres microplastic particles can be transported through the organisms? Hi Simone, so sorry I couldn't think quickly enough to answer this live! No worries :)

I don't think I can answer this from the work that we carried out, but it's a really interesting concept. Okay, but thanks for your answer!

Name: Valentina Fagiano. Do you think that based on functional traits it could be possible to individuate some proxy organism for the evaluation of plastics contamination? Thank you very much for your work and presentation

#### Name:...Marte Haave

Question :just of curiosity: Rachel: have you also found EPS in the sediments in natural environments? ever seen ingestion of these by brittle stars, or polychaetes? Hi Marte, I havne't found EPS but I also haven't been looking for it! When I filtered the 'fluff' layer, any potential EPS would have been washed away. Also, with regard to ingestion in the fauna from the natural sediment, I didn't digest or dissect these animals (I remember you warning me off in Micro2016!!). The sample analysis we carried out was very time consuming so we decided against looking for evidence of ingestion, but followed up with an experimental study instead.

Rachel: Thanks all for your questions, hopefully the study will be published soon :)

# Questions and comments for the session 27.2\_O

Similar size of microplastics and algae as food supply affects life history traits of two freshwater rotifers species. - Drago Claudia. (drago@uni-potsdam.de)

Name: Camille Richon

Question 1: Super interesting study, thanks! How do you explain the different results between the high and low food ? Is there food selectivity when food is high ? Do rotifers preferentially avoid MP when food is high?

Not sure it's well formulated sorry! But super interesting study and answers are clear thanks! :)

Particle size and hydrologic influence on microplastic accumulation in streambed sediments downstream of a municipal point source - Margenat Henar (henar.margenat94@gmail.com)

Name:Cleo Stratmann

Question 1: Great presentation! Gracias! Why you used 64µm as cut-off? Did you do procedureal blanks, as your extraction instrument was made from plastic, to account for contamination in samples? Why there was a increase in small MP about 200m after WWTP inlet? where it comes from?

Name:...Gael Le Roux

Question 2:...thanks for the presentation. Did you see fibers in your samples and any trends for this morphology?

Name:...Gholamreza Shiravani

Question 1:...Nice talk. Did you also compare the polymer-type of found MPs in sediments? Were the sediments mudy or sandy? Could the sediment and MPs aggregation to be a reason for settled/sinked MPs in sediments?

Prospective study on macroplastics and microplastics within dredging sediments - Constant Mel, Alary Claire, Billon Gabriel, Dumoulin David, De Waele Isabelle, Moreau Myriam. mel.constant@lilo.org

@Margenat Henar: Could you write your email here? I wanna ask detailed questions about MPs from WWTP in sediments.

#### Name: Cleo

Question 1: Thanks for this interesting study. What were the size classes of MP you considered (minimum)? Did you sampled replicated in each site; if yes how many? Your study implies that where there was dredging, MP can end up deep in soil sediments, actually. very interesting.

### Questions and comments for the session 27.2\_Me

#### Name:...Gholamreza Shiravani

Question 1:...Thanks for your great presentation. Were the plastic samples during the research of 6 weeks floating or were in bottom or both (settling after biofouling)? Do you know the thickness of biofilm over MPs or measure it?has the Polymer-type also an important role in biofilm formation? (Stefan Dittmar: Could you again try to answer or comment on the questions regarding thickness of the biofilm and floating/settling? I would be interested as well :)

Name:..Winnie Courtene-Jones.

Question 2:.Hi, thanks for the presentation. Perhaps i missed this (and sorry if i did), how did you measure the different pigments you refer to. can you explain these methods in a bit more details. Thank you

Miguel Tamayo to Gerardo Pulido: Are you able to produce this type of doped nanoplastics in the 1 - 100 nm range? And with kind of surfactans you use to disperse them? Thanks you a lot!! Great work!

Miguel Tamayo: Then, the sand treatment seems to be very usefull to remove nanoplastics, what about biological tretment (tipically used in most WWTPs)

Thomas Witzmann to Gerardo: Thanks for the nice talk! Do you think incubated Particles will show bigger/ lower retention than your special synthesized particles?

Erika Cedillo: Dear Gerardo thanks for your presentation. Can you tell something about the recovery of the colums? I mean, what happen with the nanoplastics that were recovered by the column?

Lars Eitzen: Thank you for the great work! How can you make sure the Pd is still present in the particles after ozonation? And were there changes in the AC pilot plant filter after longer operation times ( to compare 'new' to 'colonized' material) Could you see the same pattern as in the lab-scale experiments?

(sorry for the length and thanks to Jean-Francois!)

Hi Lars, thank you for your questions! Happy to answer them. As I was saying, we have not performed experiments in the pilot plant with pristine AC, so I could not tell what it could happen. But, based on the column esperiments, we could expect that the nanoplastic retention is increased in comparison with aged material. Let me know if you have more questions! This is my email in case you woule like to reach me there: gerardo.pulidoreyes@eawag.ch

Hey Gerardo, I will simply contact you via email, thank you!

Question to Rico:

Stephan Rohrbach: Thank you for your cool talk! How does the calcification effect the potential of MPs to vector microbial communities, also regarding for instance antibiotic resistances and pathogenic organisms.

For instance is a calcification something like a coating which increase the attachement to the MP particles or the "stickiness".

Rico Leiser: The calcfication takes places right within the biofilm (next to the cyanobacterial cells), I'm not sure if this makes the surface coating stickier. But the calcite grains are definitly enclosed by the organic matrix of the biofilms. Ok thanks for sharing your opinion here :)

Thomas Witzmann to Rico: Thanks for the nice talk! Do you think polymer type will infuence the formation of calcite on MP? Or will it only depend on the cyanobacteria?

Question to Rico:

Nicole Posth: Nice talk! How did you expose your PE to the water column? Was it in tubes? How did you retrieve them after reaction time.

Erika Cedillo: Question to Kelsey: Are you considering to measure the oxidation of PE and PS plastics in a quantitative

way? Have you performed measures of carbonyl index, changes in molecular weight or something else?

Rico Leiser: Nice talk, very interesting.

Do you know which iron minerals formed on your plastics? Were the plastics incubated under anoxic or oxic conditions

Thanks! Our SEM-EDS analysis shows that there is iron and oxygen present, but it cannot tell the valence of the iron present. The oxygen measurement through our EDS is not quite sensitive enough either for us to tell which specific iron oxide we have. We are doing sequential iron extraction in the sediments, which may give us a way to connect what we see on the plastics with what is present in the sediments. Okay, thank you for the answer ;)

Camille Richon (To Kelsey): Very interesting study! Can you say anything about the role of MPs in metals transport in the ocean ? Do you think MPs may be vectors for metal dispersion in the ocean (both water column and sediments)? Cheers! :)

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## Questions and comments for the session 27.2\_Ma

Name: Erik van Sebille

Question 1: Nice talk Bart! In your four different standardisation steps, which do you think typically introduces the largest uncertainty/error? Do you also have a framework to keep track of these conversion uncertainties in the process of standardisation? OK thanks!

Miguel Tamayo: Thanks Koelmans, i wonder why are you conssidering the volume ingested insted of the " total surface exposed" ingested, thanks!

Bart: Hi Miguel, thanks for your question. For 'dilution of food' as an adverse outcome pathway (or mechanism of effect, MoE), you need volume; i.e. the volume (or mass) of food becomes less if a volume of low-caloric MP particles is co-ingested. Area could be relevant for nano-PL in case surface reactivity triggers the adverse effect (e.g. molecular level response, or inflammation). In other words, it is the MoE that determines the metric you have to keep track of. Does this make sense?

Tamayo:Thanks for answer. Yes but since you conssider a very wide size range, if the microplastics were covered by matter that could serve the organism as a carbon source, i suppous the dilution to vary as a function of the surface area exposed. indeed, if the microplastics are co-ingested with the usefull food, the microplastics will fill part of the stomac but also could adsorb part of this food, and therefore the surface could have a relevant role as well (they are only reflexions that came into my head during your presentation, they are probably worng but i prefer to ask you about them :)) Bart: thanks Tamayo, real systems indeed can be more complex than the lab environment. What I presented was a method to get 'clean' MP lab data aligned. But if MP is biofouled you indeed have to take that into account. We have developed a model for that, which we validated on empirical data, and then we see that there is a trade-off between food-dilution (volume-based) and extra nutritional value which indeed implies that surface area becomes a model parameter as well. But .. that's too much for 7 minutes!

Tamayo:Thanks a lot Bart! Impressionant model hoping to read the article!

Bart: it is the work of Georgiana Amariei; keep track of that name in the literature somewhere next months

I know here but we never find time enough to discuss this work :) Thanks again for your presentation.

Gissell Lacerot (for Bart Koelmans). Thank you for your presentation!, I need to go and check the article now :-). But is it possible to use your approach to check for underestimation, even if you counted a certain size range?. For instance, smaller particles might be difficult to count, and maybe be miscounted, even if you're including them in your counts.

Bart: Hi Giselle! Yes, this would be possible. Feel free to contact me/us.

Will do, it was great to see you, even in this virtual mode.

Agreed, haha!

Sinja Rist's presentation :

Name:...Matt Cole

Question 2:... Really clear talk, thanks Sinja. Curious about the relatively low uptake of "adaptations". Do you think this is because of "dilution" of papers given the massive growth in plastic-focussed papers in the last 10 years?

Name: Matthieu Mercier

Q: Thanks, very interesting! How do you take into account the "quality" of the method? What is also the recommendation for a better policy of uniformisation?

Name: Juliana Ivar do Sul

we are always looking for method harmonization but this includes size classes, matrices, maybe shapes and so many other variables! how in fact we could reach this dreamed harmonized methods in a short time (years)?

just curiosity, how many citations one highly-cited method paper has in our microplastic community? Sinja: I just checked our data... The highest number for one of our novel papers was 434 WOW!!! thats a lot for a method paper! thank you That was also rather the exception. But we had many between 200 and 300.

## Name: Garth Covernton

Q: How do you differentiate between development of completely new methods and refinement of methods? For example, would changing the time period or temperature for a digestion be a new method in your analysis? How much of that rapid increase is refinement (for example, biota digestion procedures are becoming more consistent, but it's often necessary to slightly change things based on the type of sample and with many more samples and studies this means a lot of variation). And thanks for this great talk!

Sinja: We did not take changes in temperature or time as new methods (otherwise it would have become way too many ;)), it really had to be new chemicals used (or new combinations) or device setups. Of course we see that many variations of e.g. digestion methods are necessary for a wide array of sample types (not one fits all), but in our view there are still many overlapping/competing methods that could be unified.

Great, thank you. That makes sense!!!

Franziska Fischer's presentation:

Name: Alexandre Dehaut (Hi François :-))

Q: Thanks for this interesting software and presentation.

--> You're presneting an application on water that is a (quite) simple matrix allowing small filters and pores. What about the applicability to larger filter and other filter composition (biota)

--> Is it possible to compare GEPARD and siMPle? Is it planned? What is better, what is less good?

Name: Julia Möller

Great Talk Franziska! Regarding validation, do you know if there ia a measure for false positive / false negative identification of polymer types in the automated system? Alright, thank you very much!

Name: Cristiane Vidal

Nice presentation! What's the rate of false positive/negative identification? I am thinking of fluorescence that covers Raman signal in some particles - Thank you!

Hi Cristiane, you are right, fluorescence is an issue. You could use either another laser with Raman to avoid/reduce this disturbance, or switch the spectroscopic methods. GEPARD can be used with, e.g. a Perkin Elmer Spotlight 400 microspectrometer as well, so by using FTIR, fluorescence would not be a problem.

Hi Franzi! particles based methods are complicated in all steps from sampling to characterisation. do you think we should move for mass based methods as other researchers working with organic pollutants?

Hi Juliana, I think it depends on the information you need. If a mass balance is enough, py-GC/MS etc. is a faster/more feasible option. yes I agree! I was just think whether would be a good idea to have mass-based methods and contribute with our colleagues studying POPs. I just sent you an email with some pics :)

## Questions and comments for the session 27.3\_O

#### QUESTIONS FOR MATTHIEU MERCIER

Name:...Matthias Egger

Question 1:...Thanks for this great talk, Matthieu! Curious to hear your thoughts on the sampling bias associated with Manta trawling? Surely, turbulent mixing leads to substantial underestimation, but considering that larger plastic objects have significantly more mass, and that they are not collected by Manta trawls, sampling with trawls only could substantially underestimate the floating plastic mass. Or could your new Wb approach alone explain the missing plastic? Thanks! :)

#### QUESTIONS FOR MARIE POULAIN-ZARCOS

Name:...Alice Horton

Question 1:...How would you take into account changing particle density due to biofouling or surface colonisation? We know that particle density does not stay the same (and also particles may change characteristics such as size due to fragmentation)

#### Name:...Cleo Jongedijk

Question 2:...You create your 'wind waves' with a very interesting wave maker, quite different from the classical wave flumes where wind waves (and sometimes wind) are created on the boundary and the sea state is developing far away from the boundary. Do you think there is an effect on the shape of your vertical turbulence profile by the fact that your 'wave maker' upwards and downwards moving grid is actually interacting with the flow in your turbulent area? Is the type of wave maker causing the upper few centimeters to have a constant profile?

#### NAme: Matthias Egger

Question 3: ...Great talk, thanks Marie! Curious to hear your thoughts on the possible impact of your new Sct on estimated concentrations measured at sea? (essentially the same question Erik just asked.. ;))

#### QUESTIONS FOR FANNY CHENILLAT

Name: Alberto Baudena

Question 1: Thanks for the very interesting presentation. How did you exactly initialised the particles on both scenarios? How much is the distance from shore from which they particles are released?

#### Name: Victor Onink

Question 2: Hi Fanny, thanks for your very interesting talk. I was wondering how you define whether a particle has beached or not, as there are multiple ways of going about this. Related to that, are particles considered to beach permanently or do they have the opportunity to resuspend?

#### Name: Matthieu Mercier

Q: Do you consider we can consider that the current oceans observations are on average 1/5th of the total input (due to beaching) or is it more complex if you allow the beaching to be reversible? How does this ratio change with the duration of the scenario (10years/50years)?

## **Questions and comments for the session 27.3\_Me**

Name:...Mauro Borsella

Question 1:... Hola!, Ola!, congrats for the presentation Décio, love it. There's any email to write to you? Thanks!

Question 2:. Nice talk!!! I may have missed the information. Have you characterised the type of the polymers in this sampling? Thanks for the trip to Ceara! Cheers from Campinas-SP

Name: Miguel Tamayo to Born Maximilian

Question :Great work! How do you measure the weight lost of the plastics conssidering the potential adsorption of minerals or metals from the sand? Thanks In my experience, the plastics become brown due to the interaccion with micro and probabely nano particles of sand that may make you understimate the real weight lost.

I will have a look into that! Thank you! But so far none of the plastics had any patina or something. Thanks for the answer :)

Name: Cleo Stratmann

Question: really cool work. regarding more realistic waves, there are wave-creating machines used for surfing indoor or inland, maybe that is an upscale idea :)

Thanks! Yes, it is actually in development right now, just a 90 days operation is a bigger problem at the moment.

Oh wait :D i got it wrong. Thought you ment the surf zone. I want to build a small wave generator for further model verification.

. ah really? cool where?

Actually there is a big wave park planned near Aachen. Maybe it would be interesting for future research :). But consturction hasn't even started.

There is one in cologne, a river wave I think, Berlin has an indoor installation wellenwerk I think it is called

Do you think your results/approach could be transferred to river water movements from ships /sluices somehow?

Mhh, i guess the transferability is not too good, since there is no wave action there and more a dragging over the sediment.

Actually the bigger ships here in PAris couse smaller waves on the shores.

Ah okay, that might be an interesting factor. I will take a closer look into that :

Name: Claudia premet

Question : thank you for your presentation and interesting work! did you use a simulated sea water or deionized water?

I used simulated sea water (3.5 % / 1025kg/m<sup>3</sup>)

thank you, and best of luck with further studies

Name: Cristiane Vidal

Question : To Diana, could you please share your e-mail? Thanks diana.rdg@gmail.com

TO Décio: Name: Cleo Question : Did you observe an increase in MP during/shortly after rain events independently of the low/high flow seasons? Did you see differnet shaped appearing more often in different seasons?

Name: Ghezali Yousra

Question : I saw in several studies shown that the winter season presents more MPs concentration that in the summer season, although it's more frequented by tourists and all? so basically is it directly related to tides, winds and storms!!!

## Questions and comments for the session 27.3\_Ma

Feel free to add your email address if you wish to continue the discussion later

Questions for Astrid Delorme: astrid.delorme@sigma-clermont.fr

Name:...Niels Mast Question 1:...can you tell something about the fragmentation process (as opposed to degradation)?

Questions for Messika Revel

Name:...nikoletta digka Question 1:...very useful information, thank you!. maybe i lost it, but did you use any procedural blanks to track mainly airborne fibers?

Questions for Jafar Al-Omari

Name:... Vincenzo Donnarumma Question 1:...Very interesting talk Jafar! Why do you think you've got more species richness in the attached bacteria in comparison with the free bacteria?

Name:...Tony walker Question 2:...Thanks Jafar for a great talk. It is great to see a study like yours helping to fill gaps on the freshwater Plastisphere.

Stephen Kneel HI Jafar good talk, would you consider SEM to examine surface structure of plastics? grooves for attachment for biofilm formation etc.

Questions for Racheal Miller

Name:...Anna Winkler

Question 1:...Thank your for the good talk! Great job! Maybe I missed it, but did you quantify the microfiber release into the ambient air of your clothes? Can you give a number, like X microfibers / kg dry weight of synthetic clothes per drying process?

There is one from this year in July... O'Brien et al. 2020 measured MPF release from dryers into the ambient air

Thank you! PS: Racheal, if you are interested, we also measured MPF release from domestic dryers that goes into the wastewater, check out my poster ;) 24.8\_Ma

Name:...Vesna Teofilovic

Question 2:...What is your recommendation what to do with fibers that stay in drying machine after the cycle?

But where to dispose those fibers after cleaning?

Name:...Sophia Murden

Question :...Do you think enough is being done to find out how to minimise fibre loss from textiles at their design stage?

Name: Kristian Madaschi

Do you think it then follows that it's best to use a drier without outdoor venting as opposed to airdrying outdoors? Where I come from there are no driers with ducting. There's only a lint trap. Reach out to I. Napper, whose conducted related research on condenser dryers. Our driers are not condenser driers either. The water just evaporates into the laundry

## Questions and comments for the session 27.4\_O

Name:...Stjepan Budimir

Question 1:...Great work! DO you think this changes much the current state of knowledge on microplastic pollution in Adriatic sea?

questions for Thorbjorn Joest Andersen

Name:...Winnie Courtene-Jones

Question 2:...Really fascinating work, thank you for the presentation. what did you use to measure the particle grain size? was this just a 'standard' particle size analyser? can i add a follow up: was any dispersant etc. added into the PSA? THANKS

Name: Matthieu mercier

Q: Same question than Winnie. How did you measure the grain size and the velocity distribution? (Winnie)- what was said for the velocity measurements? (i missed it :( and i am interested too ) So, the velocities were measured using settling tubes. It is a fairly simple way to measure settling velocities of bulk suspensions. The method was initially described by Owen, 1976 I think. We have a more recent paper by Sengupta et al., 2011 which also describes and use the method

Name: Joao Frias

Q: About the environmentally relevant concentrations. Where they estimated and/or calculated or based on literature review? How did you arrived at those concentrations? Thanks for this. Great talk, by the way.

### Name:Gholamreza Shiravani

Question: Nice work! What does the threashold sediment concentration for increasing the settling velocity of MP? Is it for all polymers the same? The settling velocity after floculation will be the same like sediments?

Yes, that threshold we do not know yet, its something we are looking into. It may also be that it varies across plastic types

Thanks so much. Could you write your email. I do the similar research in numerical simulation of flocculation of MP and fluid mud/cohesive fine sediments in the Weser estuary of Germany. Sure, my email is tja@ign.ku.dk - you are very welcome to contact me, we are interested in cooperation

Dan Wilson: Thank you for a really interesting presentation! The pellets you found, were they a range of colours (which could indicate multiple sources? Or a uniform colour (which could indicate one source such as a spill during pellet transport)

Name: Joao Frias

Question to Juan Pablo Lozoya: Thanks for your presentation. About the pellets: Did you considered doing GCMS analysis to trace back the origin of the oil that produced the pellets? That might point out on the potential source. Some studies like Mizukawa et al

https://doi.org/10.1016/j.marpolbul.2013.02.008 point out how to trace back the source of pellets.

JPLozoya: Thank you very much!

## Questions and comments for the session 27.4\_Me

@Speaker: Gopala Krishna Darbha

Interaction of nanoplastic debris with mineral substrates: application towards their removal from aqueous systems - Gopala Krishna Darbha, Singh Nisha, Tiwari Ekta (Paper number 333695, cc-by-nc-sa.)

Name:...Gholamreza Shiravani

Question 2:...Thanks for your great talk. We just hypothesized it for fine sediments and you prove it experimentally. Great work! How do you finde the clay aggregation with microplastic instead of nonoplastics?

Name: Erika Cedillo Question: Can you give further information regarding the main characteristics of the clays used for nanoplastics removal?

In hardwater ionic strength is very high which should result in aggregation of NPs as you said effect of ionic strength, then why removal is less

@Speaker: Rebelein Anja

Microplastic fibers in the water column do not influence fertilization and hatching success of sticklebacks - Rebelein Anja, Scharsack Jörn (Paper number 334210, cc-by-nc-sa). Qestions also to: anja.rebelein@thuenen.de

Name:Camille Détrée Question: Nice study! How did you prepared your fibers? What was their average size? Name: Xavier Cousin

Question: I had network issue and I missed your presentation... was this study performed in fresh or marine water ?

In freshwater - the sticklebacks around Bremerhaven migrate to freshwater/ estuarine channels for spawning and hatching of the embryos. Later on the larvae migrate back into the sea. Thanks !

Speaker: Primpke Sebastian

Towards monitoring of small microplastics via the rapid identification and quantification of microplastics by hyperspectral quantum cascade laser infra red (QCL-IR) imaging - Primpke Sebastian, Godejohann Matthias, Gerdts Gunnar. (Paper number 334299, cc-by-nc-sa).

Email: sebastian.primpke@awi.de Publication available here: https://pubs.acs.org/doi/10.1021/acs.est.0c05722

Name:Stephan Rohrbach

Question:Thank you very much for the interesting talk! Not sure if you have seen the talk about hyperspectral imaging from Andrea Faltynkova. Any comments on advantages/ disadventages in comparison of these both techniques?

Name:...Christina Bogner Question: Thanks for the talk. Does the sample preparation change compared to FTIR?

Name: Franziska Fischer

Question: Thank you very much for this interesting presentation! Did I get it right: the spatial resolution for the 36 min measurement is  $4.2 \mu m$ ? Thanks for the answer.

Name: Matthias Tamminga

Thanks for the nice presentation. I saw you had some issues with rubbers. Do you think rubbers (tire ware) are in principle measureable using this system? Thanks!

Name:...Christina Bogner

Question: Relating to my question above: Can we simplify the sample preparation for this technique compared to FTIR? Or do we still need all the steps (digestion, density separation etc etc) FTIR is "blind" to tyre wear. Does the laser "see" them? Tank you! Good bye guys was a pleasure

Name: Cristiane Vidal

Question: Thanks for the talk!

Considering spectra pre-processing, could you provide more information? Sorry if I missed the information, how did you perform model validation on polymer type, as HCA is an unsupervised chemometric model? Thank you!

Please, could you share your email? Very interesting! Hi Christiane, the email address is sebastian.primpke@awi.de and more details can be found also in the now published publication, which should be open access soon: https://pubs.acs.org/doi/10.1021/acs.est.0c05722. Thanks very much!!

# **Questions and comments for the session 27.4\_Ma**

FOR PRESENTERS, please indicate your emails below:

- Robin:robin.treilles@enpc.fr
- Adam:awcooper@ucsd.edu
- Ligia:ligiambms@ua.pt
- Chris: chw@pml.ac.uk

## QUESTIONS FOR ROBIN TREILLES

Name: Trang

Question 1: In your opinion, what are pros and cons of sampling method you used? Stacked sieves with bucket

Name:...Niels Mast Question 2:...For the larger fragments, could you recognize (product)origin in some cases?

QUESTIONS FOR LIGIA SANTANA

Name: Camille Richon Question 1: Nice presentation, thank you! Did you measure Cu dissolved concentrations? Is there any leaching from your particles? Cool thanks looking forward to the results :)

QUESTIONS FOR CHRIS WALKSINSHAW

Name:...Christine Knauss Question 1:...Thanks for a nice talk. Some of your mussels show decreased length between days. What is your explanation for this? Thanks

Name:...arnaud Huvet Question 2:...Nice talk, thank you. Did you plan to characterize chemicals released by fibers, cotton and polyester?Ok Thank you Chris !

Name: Charlene Trestrail Question: Do you know how old the mussels are at ~1cm length?

Name: Camille Détrée Question: Nice talk! Did you investigate the presence of MF in mussels tissues?

Thank you for this great talk! The effect on the growth appears potentially marginal according to your statistical analyses (p < 0.1), therefore do you expect some effects on the energy balance? Thank you Chris for your answer

Name: Christine

Question: Why do you think the respiration rates were different between the treatments earlier in your experiment and then normalized later in your exp

Thanks for the great questions everyone!

# List of Participants by first name from A to Z

## A

A. H. M. Enamul Kabir A.D. Forero López Aaron Beck Aaron Lim **Aaron Shultis** Adam Cooper Adam Porter Adelina Teodorescu Adil Bakir Adília Pires Adrián López-Rosales Agathe Bour **Agnes Feurtet-Mazel** Aileen Jakobs Albert Koelmans Alberto Hernandez Gonzalez Alberto Hernandez-Salinas Aleksandra Karapetrova Aleksandra Lewandowska Alessandra Cera Alessandro Nardi Alessio Gomiero Alessio Montarsolo Alethea Mountford Alexander Kunz Alexander Palummo Alexander Tagg Alexandra Abrahams Alexandra Ershova Alexandra Geisser Alexandra Gulizia Alexandra McGoran Alexandre Batista Alexandre Dehaut Alice Bartalini Alice Horton Alicia Mateos Cárdenas Alina Maltseva Aline Carvalho Altair Moreira Alvise Vianello

Amaia Igartua Amaia Mendoza Amalie Thit Amanda Dawson Amanda Valois Amandine Tisserand Amedeo Boldrini Amelia Piscitello Amna Abdeljaoued Amrutha K Amy Lusher Amy Uhrin Ana Amelia Franco del Pino Ana Carolina Ruiz-Fernández Ana Carrasco Ana Costa Figueiredo Ana I. Catarino Ana Liria Loza Ana Luísa Machado Ana Luzia Lacerda Ana Markic Ana Marta Gonçalves Ana Martinez-Rodriguez Ana Patricio Silva Anastasia Barsukova Andraz Dolar André-Marie Dendievel Andrea Binelli andrea carminati Andrea Faltynkova **ANDREA Santana** Andrea Torresan Andreas Eich Andreas Fery Andreas Greiner Andreas Held Andreas Kerstan Andreas Petzold Andreas Roempp Andreia Fernandes Andreia Rodrigues Andrés Rodríguez Seijo

Andrew Chetwynd Andrew Rogers Andrey Ethan Rubin Andy Booth Andy Cundy Angela Raffaella Pia Pizzichetti Angelo Maggiore Anika Mauel Aniket Choudhary Anil Kumar Anita Jemec Kokalj Anita tirkey Anja FRM Ramsperger Anja Holzinger Anja Rebelein Anja Rott Anja Vogt Anjaly Menon Anke Müllenmeister-Sawall Ann-Kristin Deuke Anna Bogush Anna Diem anna kukkola Anna Lewis Anna M Addamo Anna MacDonald Anna Micheluz Anna Palumbo Anna Robuck Anna Rotander Anna Sanchez-Vidal Anna Soler-Membrives Anna Winkler Annabel Mathiske Annalisa Sambolino Anne Goodwin Anne Justino Anne Kveberg Anne Mol Anne-Marie Boulay Anne-Marie Schmoltner Annegret Benke

Annegret Potthoff Annemette Palmqvist Annika Jahnke Annika Vaksmaa Antje Wichels Anton Astner Antonia Law Antonia Mosberger Antonia Praetorius Antònia Solomando Martí Antonia Weltmeyer Antonio Abreu Antonio Gallardo Campos Antonio Sarno Aquilino Miguelez Argiro Adamopoulou Arianna Bellasi Arianna Varrani Ariel Smith Arnaud Gallois Arnaud Huvet Arno Bringer Áron Mári Arto Koistinen Ashley King Ashley Morgan Asta Asmundsdóttir Astrid Fischer Atocha Ramos Martinez Aurelia Liechtenstein **Axel Peytavin** Aybüke Özdamar Azora Koenig Azziz Assoumani

# B

Barbara Beckingham Barbara Ciuha Bárbara De Feo Barbara Oßmann Bárbara Rani-Borges Barbara Scholz-Böttcher Bárbara Yolanda Abaroa Pérez Barbora Pinlova **Barbro Melgert** Batdulam Battulga **Beatrice De Felice Beatriz Fernández Beatriz Rios-Fuster** Begoña Jiménez **Benedicte MORIN** Bert van Bavel **Bethanie Carney Almroth Bethany Jorgensen** bettie cormier Bettina Firmke **Beverley Waller** Bhavani Narayanaswamy **Bianca** Novello **Bianca Saraiva Bianca** Unger Birte Höcker Bjarne Kvæstad Bjørn Einar Grøsvik **Björn Pötzschner** Boris Chubarenko Brenda Hernández Brenda Hernández Britta Baechler **Brittan Scales Brittan Scales Brittany Buirs** Brittany Cunningham Bruna Ramos Bruno Tassin

## С

Camila Andreussi Camila Vidal Camille Détrée Camille Richon Camilo Saavedra capuano tonia astrid Careen Krüger **Carine Lefevre** Carla Elliff Carla Palma Carla Spetter **Carlie Herring Carlos Barreto** Carlos Gravato **Carlos Sanz-Lazaro** Carlos Silva Carme Alomar-Mascaró **Carmen** Alvarez Carmen González-Fernández Carmen González-Fernández Carmen Melendez Carola Mazzoli Carola Murano Carolin Müller **Carolin Philipp** Carolin Völker Carolina De León Carolina De León García Carolina Rocha **Carolina Rodrigues Catharina** Pieper **Catherine Tsangaris** Cathy Hobbs Cátia Venâncio Cécilia Siri Cecilie Singdahl-Larsen Ceelin Aila Andersen Celio Alves Ribeiro Ceri Lewis César Cunha Chandrasekaran Natarajan Charlene Lujan-Vega **Charlene** Trestrail Charlotte Laufkötter Charlotte Laufkötter Charlotte Lefebvre Chelsea Rochman Cherie Motti Chiara Gambardella

Chiara Zucchi Chiyeun Cheung Chloe Way Chris Walkinshaw Christian Lott **Christian Scherer** Christian Schmidt **Christian Torres Christina Bogner** Christina Kieserg Christina Thiele Christine Knauss Christoph Georgi Christoph Habel Christoph Schür **Christoph Thomas Christophe Maes Christopher Pham Ciara Keating** Cintia Hernández-Sánchez Clara Leistenschneider **Clare Collins** Claudia Bess Claudia Cella Claudia Drago Claudia Halsband Claudia Heller Claudia Lorenz Claudio Marchesi **Claudio Pierard** Cleo Jongedijk Cleo Stratmann Cloé Veclin Coco Cheung **Conrad Sparks** Cordula Witzig **Courtney Robichaud** Crislaine Bertoldi Cristiane Vidal Cristiano Gerolin Cristina Andrés Barrao Cristina Cocca

Cristina Panti Cristina Romera-Castillo Cristina Villanova Solano Cyril Hachemi

# D

Daiva Jonuskiene **Damien** Cabanes dana lopez **Danae** Patsiou **Daniel Saunders Daniel Villodres** Daniel Wilson Daniela María Truchet Daniela Silva **Daniela** Thomas Daniela Zanetti **Danielle Marchant Danilo Pires** Darbha Gopala Krishna Dario Giani Darja Kranjc Darragh Doyle Darshika Manral Daura Vega-Moreno Dave Morritt Davi Fernandes **David Broere** david gonzalez ginas David Ortiz David Pettersen Eidsvoll De Witte Bavo **Debbie Winton** Debra Ramon Décio Semensatto Dede Falahudin Defri Yona Delfina Cornejo **Delphine Dobler Delphine Lobelle Denise** Mitrano Deonie Allen

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# Ε

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## F

Fabienne Lagarde Fabio Araujo Fabiola Saroinsong fabrice brunel Fan Liu Fangzhu Wu Fanny Caputo Fanny Chenillat Fanny Courric Federica Guerrini Federico accorsi **Felipe** Alves Felipe Branco Félix Avala Felix Pfeiffer Feride Oyku Sefiloglu Fernanda Avelar Fernanda Santos Fernando Sodre Filipa Bessa Flávia Silva Fleurine Akoueson Flora Rendell-Bhatti Florane Le Bihanic Florence Parker-Jurd Florencia Rodriguez **Florent Saudrais** Florian Breider Florian Pohl **Florian Steininger** France Collard Francesca Capanni Francesca De Falco Francesca Di Pippo Francesca Garaventa Francesca Malpei Francesca Modugno Francesco Parrella Francesco Regoli Francesco Saliu Francisca Ribeiro Franco Teixeira de Mello Francois Audrezet François Galgani Franz Bogner Franz Dichgans Franziska Fischer Frapiccini Emanuela

Frederieke Kroon Frederique Mongodin Freya Radford Friederike Gabel Friederike Ganster Fusar Poli Fusar Poli

# G

Gabor Bordos Gabriel Blázquez Gabriel Cardozo-Ferreira Gabriel De-la-Torre Gabriël Olthof Gabriela Kal?ikova Gael Forget Gael Le Roux Gaelle Darmon Gaëtan Cormy Gara Goñi Godoy Gariz Riveros Garth Covernton Gema Hernandez-Milian Gema Paniagua González **Georg Dierkes** Georg Papastavrou Georgia Holt Georgie Savage Gerardo Pulido-Reves Gerasimos Gkoutselis Gerson Fernandino Gert Everaert Ghanshyam Barman Ghezali Yousra Gholamreza Shiravani Giacomo Limonta Gian Bremm **Gilles Baratto** Gina López Giorgia Carratta Giovana Gimiliani Gisela Böhm Gisele Conceição
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#### Η

Habel Christoph Halina Binde Doria Hana Fajkovi? Hanna Cornelsen Hanna Kuhfuss Hannah Anderson Hannah De Frond Hannah Schäfer Hannes Imhof Hannes Laermanns Hannes Schritt Haroutioun Askanian Harriet Paterson Harrison Frost Harvey Harbach Hassan Elagami Heather Hunt Heather Trim Heinar Schmidt Helen Kaberi Helen Polanco Helen Wolter Helena Ruffell Helene Thomas Henar Margenat Henar Margenat Henry Arbaugh Hergen Haase Hilde Ervik Hilde Ervik Holger Kress Holger Schmalz Holly Nel

#### I

Ian O'Connor Ico Martínez Sánchez Ieva Uoginte Ifenna Ilechukwu Ignacio Gestoso Ika Paul-Pont Ilaria Degano Ilka Peeken ilkan Özkan Imari Walker Imisi Arowojolu **Imogen Smith** Ines Ruff Inga Kirstein Inger Lise Nerland Bråte Inger Lyngstad Inma Herrera Inta Dimante-Deimantovica Iordachescu Lucian

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#### J

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L

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Martin Löder Martin Mittelbach Martin Wagner Martina Vollmer Martinussen Kine Marzive Molazadeh Masayuki Kawahigashi Mateo Cordier Mathilde Falcou-Prefol Matteo Baini Matteo Galli Matthew Cole Matthew Ross Matthias Egger Matthias Ochs Matthias Schwarz Matthias Tamminga Matthias Völkl Matthieu Mercier Mattias Bossaer Maura Benedetti Mauricio Díaz-Jaramillo **Maurits Halbach** Mauro Borsella Max Döring Maximilian Born Maximilian Breitenbach May Gomez Maysan Nashashibi Mechthild Schmitt-Jansen Megan Grant Mel Constant Melanie Bergmann Melanie Fritz Melanie Poehlmann Melisa Fernandez Melissa Maurer-Jones Melissa McClure Meral Yurtsever Mercè Morató Trobat Meredith Seeley Merel Kooi

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Ν

Nadia Bouzid Naoual Ben-Charki Natalia Buzzi Natália Donato Natalia Sánchez-García Natalie Caughtry Natalie Hernandez Natalja Buhhalko Natascha Schmidt Natasha Mimpriss Nathaniel Clark Naveenkumar Yaranal Neda Farahbakhshazad Nedim Vardar neha badola **Nelle Meyers** Nereida Cordeiro Nevena Hristozova Nia Jones Nicholas Mackay-Roberts Nicola Shepherd Nicolas Grima Nicolas Keller Nicole Allison Nicole Baker Nicole Posth Nicole Zumbülte Niels Mast Niina Kärkkäinen Niko Krebs Nikoletta Digka Nils Kerpen Nina Paul Ninon Cefalu Nisha Singh Nithin Achutha Shettigar Niu Zhiyue Nora Meides Noreen Kelly Nourhene Rebai Nourhene Rebai

#### Nur Hazimah Mohamed Nor

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Oihana Latchere Oihane C. Basurko Olaia Zarketa Olga Lobchuk Olga Pantos Oliver Knoop Olivia Gerigny Olivier Lepaul Olubukola Alimi Orasai Faikhaw Oriol Rodríguez Romeu Oriol Uviedo Ortega Andres Oskar Hagelskjaer Otmar Geiss

### Р

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Pinja Näkki Pouyan Ahmadi Prasun Goswami

#### R

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#### S

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Simon Riedl Simon Werschmöller Simon Wieland Simona Mondellini Simone Lechthaler Simone Syhre Sindhura Stothra Bhashyam Sinja Rist sofi azilan aini Soledad Álvarez Soledad Muniategui Lorenzo Sonia Doblado Sonja Ehlers Sonja Oberbeckmann Sonya Moses Soo Bin Kim Sophia Murden Sophia Reichelt Sophie Bourgeon Sophie Schmiz Sören Gutekunst Soumaïa Tajouri sous damien Spela Korez Stacey O'Brien Stamatia Galata Stefan Dittmar Stefan Grass Stefan Lips Stefan Maday stefan peiffer Stefan Schweiger Stefania Di Vito Stefania Federici Stefania Gorbi Stefanie Ypma Stefano Magni Steffen Weyrauch Stephan Gekle Stephan Rohrbach Stephan Wagner **Stephanie Andrews** 

Stephanie Cieplik Stephanie Eytcheson Stephanie LaPlaca Stephanie Reynaud Stephen Burrows Steve Allen Steven Loiselle Stiffani Rominikan Stjepan Budimir Susana Agusti Susanne Belz Susanne Kühn Susanne Waaijers-van der Loop Sven Frei Sven Ritschar Svenja Mintenig Symiah Barnett

#### Т

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**Thomas Fröhlich** Thomas Maes **Thomas Steiner Thomas Witzmann Thorbjorn Andersen** Thorsten Hüffer Tillmann Lueders Tilman Durst Tim Lauschke Tim van Emmerik Tina Kunde **Tine Engelund Tingting Zhao** Tom Stanton Tom Sun Tomasz Burghardt Tommaso Giarrizzo tommaso nacci **Tong Yang Tony Kingsbury Tony Walker Torben Brecht Torsten Bettendorf** Trishan Naidoo **Troy Langknecht Tsering Tenzin** Tycho Bovenschen

#### U

Udit Pant Ula Rozman Ulgen Aytan Ulrich Meincke Urszula Aleksander-Kwaterczak Uwe Schneidewind

#### V

Vahid Pilechi Vaibhav Budhiraja Valentina Fagiano Valerie Barbe Valerie Jerome

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#### W

Walden Marcus Walter Waldman Wendy Alvarado Aguilar Wiebke Mareile Heinze William de Haan Willian Araújo Win Cowger Winnie Courtene-Jones Winnie de Winter Wolfgang Babel Wu Nan

#### Х

Xavier Abril Xavier Cousin Xia Zhu

## Y

Yang Liu Yann Patry Yasmin Yonan Yasmina Rodríguez Yen Vu Yoann Garnier Yolène Ledoux Yong Sik Ok Yoora Cho Yoshio Ikezawa Yu Li Yuanhu Zhang Yuanli Liu Yves Boulard Yvonne shashoua **Yvonne Staal** 

# Ζ

Zahara Martínez de Pedro Zara Botterell Zouxia Long

# **Closing words**

There you have it, the works presented at the 3rd edition of MICRO in an openscience nutshell. In publishing these proceedings we mark another milestone on the road to Micro 2022. Hope you will join us in laying the groundwork for this collaborative effort of the MICRO community.

The aim of the MICRO international conference series is to celebrate the burgeoning community of researchers, policy-makers, and the public concerned by Plastic Pollution from Macro to nano.

The MICRO conference series serves as a means to: (i) Identify current research limits and new challenges, (ii) Facilitate open access to the breadth of ongoing research, and (iii) contribute a collaborative effort to our continuously expanding community.

Many thanks to all who contributed, and see you at Micro 2022: Outside !

# MICRO2020 INTERNATIONAL CONFERENCE



23-27 NOVEMBER 2020 LANZAROTE AND BEYOND\*

FATE AND IMPACTS OF MICROPLASTICS: KNOWLEDGE AND RESPONSIBILITIES



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