TAFT 2015

5th Trans-Atlantic Fisheries Technology conference
(45th WEFTA meeting)

Program Book

Organising committees

Scientific Program Committee

WEFTA Representatives
Torger Børresen
Mercédès Careche – CSIC, Instituto del Frio, Madrid, Spain
Françoise Leroi – Ifremer, Nantes, France
Joop Luten – Wageningen UR (Imares), IJmuiden, the Netherlands
Heidi Nilsen – Nofima, Norway
Ingrid Undeland – Chalmers University, Sweden
Véronique Verrez-Bagnis (chair) – Ifremer, Nantes, France

AFTC Representatives
Pierre Blier – Université du Quebec, Rimouski, Canada
David Green – North Carolina State University, North Carolina, USA
Heather Manual – Memorial University, Fisheries and Marine Institute, Newfoundland, Canada

SST Representatives
Lee-Ann Applewhite – Applied Food Technologies Inc, Alachua, Florida, USA
Mike Jachnke – Virginia Seafood Agricultural Research and Extension Center, Virginia, USA

Local Organizing Committee

Isabelle Adam – Ifremer, Nantes, France
Régis Baron – Ifremer, Nantes, France
Jean-Pascal Bergé – IDMer, Lorient, France
Véronique Bossière – Cap Aliment, Nantes, France
Mireille Cardinal – Ifremer, Nantes, France
Frédérique Chevalier – Ifremer, Nantes, France
Catherine Coriou – Ifremer, Nantes, France
Josiane Cornet – Ifremer, Nantes, France
Monique Etienne – Ifremer, Nantes, France
Françoise Leroi – Ifremer, Nantes, France
Sylvie Nouhant – Ifremer, Nantes, France
Sophie Pilven – Ifremer, Nantes, France
Patricia Thibault-Praslin – Ifremer, Nantes, France
Jean-Luc Vallet – Ifremer, Nantes, France
Véronique Verrez-Bagnis – Ifremer, Nantes, France

Registration and reception
12th October 2015

The welcome reception and registration takes place

Monday 12th october 2015 from 17:00 to 20:00

At La Cité (Nantes Events Center) - 5, Valmy street, Nantes, France

©La Cité Nantes, City center events

17:00 – 19:00	Registration
19:00 – 20:00	Welcome cocktail reception

CONFERENCE AGENGA

Tuesday, October 13

9:00 Opening by Véronique Verrez-Bagnis of Ifremer Nantes Centre

9:10 General introduction by Henri Seegers, Regional delegate of INRA Angers-Nantes Centre and representative of the food regional association "Cap Aliment"

9:25 WEFTA award winner speech by Horst Karl “Old problems and new challenges in research on new aquatic species in trade”

SESSION 1: Consumer behavior, nutritional value and functional foods

Chair: Sanja Vidacek and Michael Morrissey

9:50 **Keynote lecture:** Eating fish today: how combining naturalness and functionality - Gervaise Debucquet, Audencia, France (invited speaker)

10:20 Organic Aquaculture: Factors relevant for consumers - Themistoklis Altintzoglou, Nofima, Norway

10:25 Content of health beneficial compounds in Atlantic Mackerel during cold- and frozen storage – Turid Rudstad, Norwegian University of Science and Technology, Norway

10:30 Coffee break / posters

11:00 Bioaccessibility: a key for a better evaluation of the benefits and risks of fish consumption – Narcisa Bandarra, Portuguese Institute for Sea and Atmosphere (IPMA, I.P.), Portugal

11:20 Application of a sorting procedure for sensory evaluation of seafood products: comparison with conventional profiling – Mireille Cardinal, Ifremer, France

11:40 Sensory Characteristics of Food Products Made from Dulse (*Palmaria mollis*) – Michael Morrissey, Oregon State University Food Innovation Center, USA

12:00 Health and nutrition claims’ effects on consumer perception of omega-3 enriched convenience meals - Pall Amar Hauksson, Faculty of Food Science and Nutrition, Iceland

12:05 Biogenic amine content of fish products sold in Turkish market - Ali Serhat Özkütük, Faculty of Fisheries, Cukurova University, Turkey

12:105 Antioxidant and functional properties of shrimp hydrolysates and its application to fish tofu – Sunantha Ketnawa, Mae Fah Luang University, Thailand

12:20 Lunch

SESSION 1: Consumer behavior, nutritional value and functional foods

13:45 Fish protein hydrolysates processed with different natural antioxidants - Dana Rán Jónsdóttir, Matís, Iceland

14:05 Identification of peptides in a Prolyl Oligopeptidase-inhibiting fraction from trypsin hydrolysate of Pacific white shrimp (*Penaeus vannamei*) - Oscar Martínez Alvarez, Institute of Food Science, Technology and Nutrition, Spain

14:10 Determination of some functional properties of enzymatically hydrolyzed protein from head of sea bass (*Dicentrarchus labrax*) and its effect in whiting (*Merlangius merlangus*) mince - Şebnem Tolosa Yilmaz, Ege University, Faculty of Fishery, Turkey

SESSION 2: Bio-economy of aquatic resources

Chair: Heidi Nilsen and Régis Baron

14:20 **Keynote lecture:** Opportunities in the Blue Bioeconomy – Sveinn Margeirsson, Matis, Iceland

14:50 Replacement of surimi in restructured crab by raw crabmeat in restructured crab products – David Green, North Carolina State University, Center for Marine Sciences and Technology, USA

15:10 Study of the optimization conditions for hydrolysates production from *Scyliorhinus canicula* muscle and antioxidant activities - María Blanco, Grupo de Bioquímica de alimentos (IIM-CSIC), Spain

15:15 Coffee break / posters

15:45 Development of an ultrasound-assisted enzymatic hydrolysis process for the liquefaction of the red seaweed *Grateloupia turuturu* and its biomolecules recovery – Cécile Le Guillard, Ifremer and University of Nantes, France

16:05 An example of French strategy for the production of marine ingredients from salmon by-products: the Pesk&Co project, Margot Provost, University of Bretagne Occidentale, LEMAR UMR6539, France

SESSION 3: Aquaculture and new aquatic resources

Chair: John Fagan and Thomas Verhaeghe

16:10 **Keynote lecture:** Advancements in land-based brackish-water and marine aquaculture and their future in seafood production - Thomas Losordo (invited speaker), Pentair Aquatic Eco-Systems Inc., USA

16:40 Oyster refinement: effects of algae diets (*Skeletonema costatum* and *Rhodomonas baltica*) on the sensory characteristics and volatile organic compounds of Pacific cupped oysters (*Crassostrea gigas*) - Jasper van Houcke, HZ University of Applied Sciences, The Netherlands

17:00 Chemical and Microbiological Contamination in Japanese-clam (*Ruditapes phillipinarum*) from Tagus Estuary (Portugal) - Sónia Pedro, Portuguese Institute for Sea and Atmosphere (IPMA, I.P.), Portugal

17:05 New possibilities for coproducts from Atlantic salmon backbones and bits & pieces in fish spread – Grete Hansen Aas, AAlesund University College, Norway

Wednesday, October 14

SESSION 4: Seafood processing – incidence on quality and safety

Chair: Françoise Leroi and Sigrún Halldórsdóttir

9:00 **Keynote lecture:** Seafood Processing Innovation and Entrepreneurism: Challenges and Developments - David Green (invited speaker), North Carolina State University, Center for Marine Sciences and Technology, USA

9:30 Quality and safety of tropical yellowfin tuna (*Thunnus albacares*) steaks stored under air, vacuum or modified atmosphere – Adèle Dauchy, Ifremer, France

9:50 The effect of storage temperature on *Vibrio parahaemolyticus* numbers in Pacific oysters (*Crassostrea gigas*): Evaluating a predictive model under New Zealand conditions - Graham C Fletcher, New Zealand Institute for Plant & Food Research Limited, New Zealand

10:10 Cold Storage stability of minced fish added of grape antioxidant dietary fiber pomace - Helena M. Moreno Conde, Institute of Food Science Technology and Nutrition (ICTAN-CSIC), Spain

10:15 Quality improvement of cooked brown shrimp *Crangon crangon* trough detailed kinetic studies of the major quality attributes - Thomas Verhaeghe, Institute for Agricultural and Fisheries Research (ILVO), Belgium

10:20 The effects of nanoemulsions based on commercial oils (sunflower, canola, corn, olive, soybean, and hazelnut oils) on sensory, chemical and microbial quality of frozen sea bass fillets - Deniz Ayas, Mersin University, Turkey

10:25 Coffee break / posters

11:00 Investigation of ohmic heating for seafood processing - Stina Frosch, Technical University of Denmark (DTU), Denmark

11:20 Suppressive effect of ATP on autoxidation of tuna oxymyoglobin to metmyoglobin - Kota Inohara, Kagoshima University, Japan

11:40 Effect of heat treatments on mobility and in vitro infectivity of *Anisakis* L3 in hake muscle infected under controlled conditions - Isabel Sánchez-Alonso, Institute of Food Science (ICTAN-CSIC), Spain

12:00 Differences between *A. simplex s.s.* and *A. pegreffii*: in vitro infectivity and freezing tolerance - Mercedes Careche, Institute of Food Science (ICTAN-CSIC), Spain

12:05 Prevalence of nematodes (*Anisakidae*) in fish species most consumed in France – Véronique Verrez-Bagnis, Ifremer, France

12:10 Packaging, quality and shelf life of fillet products from live captured and stored Atlantic cod – Heidi Nilsen, Nofima AS, Norway

12:15 Heat resistance of the most isolated spore-forming bacteria in ready-to-eat brown crab meat - Santiago Condón-Abanto, University College Dublin, Ireland

12:20 Lunch

STRATEGY SESSION: FROM INDUSTRY TO RESEARCH (13:45 – 17:00)

Introduction of the strategy session: Jean-Pascal Bergé, IDmer (20 min), presentation of IDMer and introductive roundtable conference

Presentation of industry stakeholders participated in the roundtable: Copalis, Procidys, Gouessant, Diana, Olmix, Fleury Michon

First round table led by Copalis andProcidys on the general theme of contaminants and other undesirable compounds (heavy metals, dioxins, PCBs, antibiotics and pesticide residues ...) regarding analytical, technical, biological and regulation issues.

15:15 Coffee break

Second round table chaired by Fleury Michon on the sustainability of the sector and optimization of the value chain: sustainability, circular economy, environmental impact. Issues related to consumer perception and use of unvaluated sources will be addressed: regulation, processing, preservation, sensory characteristics, traceability ...

19:00 Gala dinner at "Les Fonderies", 25 Boulevard Vincent Gâche, 44200 Nantes

Nominations of WEFTA Award, Earl P. McFee Award and Lifetime Achievement Award

Thursday, October 15

SESSION 4: Seafood processing – incidence on quality and safety

Chair: Mercedes Careche and Jasper van Houcke

9:00 Minimizing the content of free and ester bound 2,- 3-MCPD and esterified glycidol in fish fingers - Sybille, Merkle, University of Applied Sciences Hamburg, Germany

9:20 Utilization and stability of cod liver during frozen storage – Effects of season, on-board handling and storage conditions - María Gudjónsdóttir, University of Iceland, Iceland

9:40 Conversion of lysine to cadaverine by cell-free supernatants (CFSs) from *Salmonella Paratyphi* A, and *Escherichia coli* in lysine enriched decarboxylase broth (LDB) - Fatih Ozogul, Faculty of Fisheries, Cukurova University, Turkey

10:00 Inhibitory effects of high pressure processing on *Morganella psychrotolerans* in herring (*Clupea harengus*) - Ilknur Ucak, Akdeniz University, Turkey

10:05 The effect of lactic acid bacteria isolated from fish on microbiological quality of silage made from fish processing waste - Ali Serhat Özkütük, University of Nigde, Turkey

10:10 Production of fish chips with using frozen saithe flesh (*Pollachius virens*) and determining its shelf life - Tolga Dincer, Ege University Faculty of Fisheries, Turkey

10:15 Comparison of the microbiological, chemical and sensory quality of plaice (*Pleuronectes platessa*) stored in flake ice and slurry ice – Karen Bekaert, ILVO, Belgium

10:20 Simplification of K-value measurement as an index of freshness of fish - Kunihiko Konno, Hokkaido University, Japan

10:25 Coffee break / posters

11:00 Quality evaluation of fresh, farm raised sea vegetables during refrigerated storage - Denise Skonberg, School of Food and Agriculture, University of Maine, USA

11:20 Quality improvement in trawling fisheries – CRISP; adaption of capture and handling practices to optimize catch quality and value - Heidi Nilsen, Nofima,

11:40 The role of blood for lipid oxidation and color stability of fish - Hanna Harrysson, Chalmers University of Technology, Sweden

12:00 Determination of quality differences between canned tuna and pouched tuna in different packing media – Asli Cadun, University of Ege, Fisheries Faculty,Turkey

12:05 Heat processing of pre-rigor Atlantic cod (*Gadus morhua*) - Svein Kristian Stormo, Nofima, Norway

12:10 Challenges in textural measurements of Atlantic salmon - Gine Ørnholt-Johansson, Technical University of Denmark, Denmark

12:15 Effect of crust freezing on the shelf-life of salmon (*Salmo salar*) stored at low temperatures under different packaging conditions – Selene Pedrós, School of Veterinary Medicine, University College Dublin, Ireland.

12:20 Lunch

SESSION 5 Innovative methods for characterization of seafood products and contaminants

Chair: Véronique Verrez-Bagnis and Guillaume Duflos

13:45 **Keynote lecture:** Latest advances for the characterization of seafood in a global market - Carmen Sotelo (invited speaker), Grupo de Bioquímica de alimentos (IIM-CSIC), Spain

14:15 Development of a qPCR method for the identification and quantification of *Thunnus obesus*, *Thunnus albacares* and *Katsuwonus pelamis* in canned tuna – Daline Bojolly, Université du Littoral Côte d'Opale, France

14:35 Species identification in samples containing fish mixtures: a targeted Next-Generation Sequencing approach - Kristina Kappel, Max Rubner-Institut, Germany

14:55 Development of a qPCR method targeting torA gene and application for the freshness monitoring of modified atmosphere-packed chilled whiting (*Merlangius merlangus*) – Alexandre Dehaut, Anses, France

15:15 Coffee break / posters

15:45 The application of near infrared spectroscopy to study physicochemical properties and quality degradation of seafood - Magnea Karlsdottir, Matis, Iceland

16:05 Use of fluorescence spectroscopy for monitoring whiting (*Merlangius merlangus*) fillets freshness stored under various conditions – Abdo Hassoun, Artois University, France

16:10 Detection of histamine in fish by Surface Enhanced Raman Spectroscopy using solid silver SERS substrates - Tibor Janči, University of Zagreb, Croatia

16:15 Toxicity of the Lessepsian pufferfish *Lagocephalus sceleratus* from Turkish Mediterranean coast and the species authentification by rapid PCR amplification method - Caner Acar, Tokyo University of Marine Science and Technology, Japan

16:20 Characterization of *Shewanella baltica* strains with usual and atypical H2S productions, isolated from a spoiled whiting (*Merlangius merlangus*) – Alexandre Dehaut, Anses, France

16:25 Highly active phosphatase is responsible for rapid loss of IMP nucleotide in cod muscle – Larissa Balakireva, NovoCIB SAS, France

16:30 Announcing WEFTA 2016 and TAFT 2018

Friday, October 16

SOCIAL PROGRAM (8:45 – 15:00)

Visit of the French Institute of Vine and Wine (Vertou, in the immediate vicinity of Nantes)

Visit of a regional winegrower and lunch at the wine producing place at Château-Thébaud (small town close to Nantes)

Meeting point "La Cité" 8:45 – departure by bus at 9:00

ORAL PRESENTATION ABSTRACTS

Old problems and new challenges in research on new aquatic species in trade

2014 Wefta award winner: Horst Karl

Max Rubner-Institute, Hamburg, Germany

Research on new aquatic species for human consumption has a long tradition in Germany.

Already in the early 1970th interest in underutilized marine species such as blue whiting, great silver smelt and Alaska pollock was increasing due to overfishing of traditional North Atlantic fish stocks. Blue whiting was discussed as substitute for deep frozen cod or saithe fillet blocks, but research revealed high Anisakis prevalence and abundance which stopped the production. The success story of Alaska pollock started. Today Alaska pollock is the most important marine fish species on the German market.

Between 1975 and 1985 utilization of Antartic krill (*Euphausiasuperba*) as possible protein resource for human consumption was in the focus of international research projects, including characterization of the raw material and product development. High fluoride content and endogenous enzymatic activitiesimpeded further product development and the interest of industry in commercializing of krill products was low at that time. Today krill oil is highly demanded asdietary supplement.

Within the last 10 years, research on new aquatic species focused more and more on the risks and benefits of tropical and subtropical species imported into the European Union. The variety of these species increases each year. Often no or minimal knowledge is existing on the chemical composition, species authentication, parasite status, sensory and storage characteristics and on possible risks (contaminants, natural toxins). Characterisation of imported deep frozen fillets of pangasius,tilapia or farmed barramundi showed that the nutritious value in terms of iodine and long chain polyunsaturated fatty acids is not comparable to traditional marine species or to rainbow trout and salmon from aquaculture. Further pangasius fillets, but also deep sea scallops and other imported fish species are often treated with an excess of water and water binding additives. On the other hand, species like cobia and barramundi are highly delicious and can contain considerable amounts of selenium.

The increasing import of tropical reef fish species like red snapperand grouper will increase the risk of ciguatera fish poisoning for European consumers.The global trade of new species as wet fish via air cargo will additionally increase the risk of food borne parasitic infections. Measures have to be evaluated to assure the safety of the consumer.

Eating fish today: how combining naturalness and functionality

Keynote speaker: Gervaise Debucquet

Audencia-School of management, Nantes, France

Sociologists have shown for a long time how social and cultural representations are associated with food influence perceptions, acceptation and avoidance. In this sense and in comparison with meat, fish and fish products appear to have a specific status, especially in the Judeo-Christian World. Initially associated with negative images and perceived as a "sub-meat", fish is nowadays promoted by nutritionists and fish industry as an essential component of a healthy diet preventing diseases. For several years, fish has become a 'natural functional food' and have certainly benefit from the context of the quick spread of Nutritional and Health Claim (NHC). At the same time, fast-expanding aquaculture with intensive and high technology techniques has introduced confusion in consumer's mind on quality of farmed fish, organic and others quality-related labels having brought only partial reinsurance. Increasing promotion and/or incorporation of marine nutrients with functional properties in foodstuffs could be a new step in the 'reification' of fish. Which impacts could have this technical-scientific-medical view on consumers' perception of fish and fish products? Could they lead consumers to refer back more to lay believes, to reactivate heuristics as naturalness or wildness and lastly to solve themselves the dilemma between eating fish for health and eating fish for pleasure and sharing? These points will be discussed in this lecture.

Key words: food culture – representations – food pleasure – healthy food - food dilemmas

Organic aquaculture: factors relevant for consumers

Presenting author: Themistoklis Altintzoglou

Co-authors: Pirjo Honkanen

Nofima, Tromsø, Norway

This study is part of the EU project OrAqua. The project's objective is to offer recommendations for the regulatory framework for organic aquaculture in Europe (EC Regulation 710/2009), based on a review of relevant scientific literature related to the current regulation. The emphasis has been on four main themes: a) feed, b) welfare, c) production systems and d) environmental impact of farming systems. Within OrAqua, consumers' perceptions of the elements included in the regulations are also taken into account. The objective of this consumer study is to of assess consumer perceptions, sentiments and understanding of organic aquaculture to promote consumer confidence and acceptance of organic farming principles based on scientific knowledge.

A thorough literature review was performed to identify existing scientific knowledge and reveal knowledge gaps. The review covered scientific publications and grey literature published since the year 2000. The found body of literature was filtered for relevance and evaluated for quality. With the areas of the regulatory framework in mind and knowledge gaps revealed by the review, a survey was developed. The survey included questions about consumer perceptions of the relevance of various issues to the official definition of organic fish, how these issues can influence the quality of the fish, attitudes about wild, conventional farmed and organic farmed fish, knowledge about organic fish, consumption behaviour and socio-demographic characteristics.

Consumers' knowledge of organic aquaculture is low, with no clear perception whether it is wild or farmed fish that can be called organic. Consumers associate natural living conditions with the definition of organic fish, but elements of the official regulation are ranked much lower. Consumers associate issues related to feed composition and utilization to the quality of the final fish products in general but not with organic aquaculture in particular. Fish welfare is relevant for a part of the population, but not all natural production practices lead to optimum fish welfare. When consumers are more informed and realise the balances between less-natural production decisions and their impact on the fish and the environment, their perception of aquaculture and expectations from organic production become more positive.

The consumers' perception of organic farmed fish is heavily influenced by their perception of fish farming in general. Therefore, a recommendation from these results would be to design and pre-test communication campaigns, which will have a positive effect on seafood consumption in Europe and create awareness about organic aquaculture within them.

Key words: Consumers, organic, fish, feed, regulations

Content of health beneficial compounds in Atlantic mackerel during cold- and frozen storage

Presenting author: Turid Rustad[1]

Co-authors: Inger Beate Standal[2], Revilija Mozuraityte[2], Ingrid Undeland[3], Hanne Digre[2]

[1]NTNU (Norwegian University of Science and Technology, Trondheim, Norway

[2]SINTEF Fisheries and Aquaculture, Trondheim, Norway

[3]Chalmers University of Technology, Sweden

Pelagic fish is considered a healthy food based on the content of marine lipids, proteins, vitamin D and other bioactive compounds. However, the quality of fish changes during storage. The aim of the present study was to examine the stability of health beneficial compounds and other quality parameters during cold and frozen storage of mackerel fillets.

Fillets of mackerel caught in February, were either chilled stored for 4 and 9 days or frozen stored (-27 ^{o}C) for 1, 7 or 12 months. The following parameters were measured: water, ash, water holding capacity, vitamin D, content and quality of lipids and proteins (oxidation of sarcoplasmic and myofibrillar proteins), and profile of low molecular weight (LMW) metabolites. Lipid oxidation was measured by peroxide value and thiobarbituric reactive substances. Protein oxidation was measured by contents of protein carbonyl and total thiol. Profile of water soluble low molecular weight metabolites was obtained by high resolution NMR spectroscopy.

The compositional analysis shows that amount of water, lipids, vitamin D and fatty acid profile is stable during storage. Lipid oxidation was not observed at the storage conditions. However, hydrolysis of lipids was observed, especially during storage of +4^{o}C. Proteins seem to be more influenced by the duration of the frozen storage. Frozen storage for 7 and 12 months significantly reduced the oxidative stability of sarcoplasmic and myofibrillar proteins in the muscle, compared to 1 month frozen and chilled storage. The results show that LMW compounds, such as taurine and inosine and inosine-monophosphate are well preserved during frozen storage. Time from catch to freezing is important when it comes to the level of such compounds. During cold storage (+4^{o}C up till 9 days) changes in composition occur that will influence the taste and safety of the product, e.g. the formation of biogenic amines and other degradation products.

Health beneficial compounds in mackerel, such as omega-3 lipids, vitamin D, and LMW metabolites were stable during frozen storage up till 12 months at -27 ^{o}C. Oxidative stability of proteins was reduced during long term frozen storage (7 and 12 months). The quality of the fish at the time of freezing, is a critical parameter. During cold storage (at +4^{o}C up till 9 days) changes in composition and quality occurs, namely hydrolysis of fatty acids and degradation of LMW metabolites.

Key words: frozen, storage, lipids, oxidation, quality

Bioaccessibility: a key for a better evaluation of the benefits and risks of fish consumption

Presenting author: Narcisa Maria Mestre Bandarra[1]

Co-authors: Cláudia Afonso[1], Sara Costa[1], Irineu Batista[1], Carlos Cardoso[1], Inês Coelho[2], Maria Leonor Nunes[1]

[1]Department of Sea and Marine Resources, Portuguese Institute of Sea and Atmosphere (IPMA, IP), Av. Brasília 1449-006 Lisbon, Portugal

[2]Food and Nutrition Department, National Health Institute Doutor Ricardo Jorge (INSA, IP), Lisbon, Portugal.

The bioaccessibility of the nutrients and contaminants in different seafood products is a promising field of research that may shed light into the real risks and benefits associated to their consumption. Instead of performing a risk-benefit analysis on the basis of the total contents of nutrients, such as selenium (Se), eicosapentaenoic (EPA), or docosahexaenoic (DHA) fatty acids, and contaminants, such as methylmercury (MeHg), the levels of these substances in the bioaccessible fraction (available to be absorbed across the intestinal wall) are used as inputs in the mathematical modeling.

Hence, the performed scientific work brings together two innovative areas of research, *in vitro* digestion modeling and advanced statistical treatment of the probabilities of exceeding the advised nutritional requirements and contaminant thresholds. The bioaccessible fraction of the main nutrients and contaminants in seafood was analyzed across a varied array of fish species and culinary treatments, encompassing wild and farmed products, and lean and fat fish.

Se bioaccessibility (ratio between bioaccessible content and total content before digestion) was always higher than 80 % with exception of canned tuna in water and olive oil which presented lower percentages (72 and 82 %, respectively). Of all studied wild and farmed fish species, farmed salmon was the one that displayed the highest EPA and DHA bioaccessibility, 71.1 % and 73.5 %, respectively. Compared to raw, EPA+DHA bioaccessibility usually decreased with the culinary treatment, especially after grilling (30-60 %). On the other hand, it was found out that Hg and MeHg bioaccessibility from raw fish ranged from 75 to 90 %. However, cooking or processing may cause a reduction of these contaminants bioaccessibility. Particularly, grilling reduced the bioaccessible concentrations of MeHg to 30-60 % of the initial contents. It seemed that the more drastic the heating treatment the lower the bioaccessibility (less than 20 % for processed tuna).

Risk-benefit assessment enabled to identify species and culinary treatments with a more favorable health impact, such as grilled fish or canned tuna.

Key words: farmed and wild fish; nutrients and contaminants; culinary treatment and processing; bioaccessibility; risk-benefit assessment

Application of a sorting procedure for sensory evaluation of seafood products: comparison with conventional profiling

Presenting author : Mireille Cardinal[1]

Co-authors: Josiane Cornet[1], Adèle Dauchy[2], Françoise Leroi[2], Régis Baron[1], Philippe Courcoux[3,4]

[1]Ifremer, Laboratoire Bioressources Marines et Bioraffinerie par hydrolyse enzymatique (BIORAF HE), BP 21105, 44311 Nantes, France

[2]Ifremer, Laboratoire Ecosystèmes Microbiens et Molécules Marines pour les Biotechnologies (EM3B), BP 21105, 44311 Nantes, France

[3]Oniris, Sensometrics and Chemometrics Laboratory, 44322, France Nantes, France

[4]LUNAM, University Nantes Angers Le Mans, France

These last few years, many papers have discussed the value of rapid sensory methods as an alternative or a complementary techniques to quantitative descriptive analysis, considered the reference tool for food sensorial characterisation (Varela, 2012; Dehlholm, 2012, Veinand, 2011). Among these methods, free sorting technique appears rapid for collecting data from the sensory panel, easy to understand, and gives generally a sensory map close to those obtained with conventional profiling. The interest and the possible use of this procedure has been tested on marine products in two different case studies.

The first case study aimed to evaluate the spoiling potential of 9 strains isolated from spoiled tuna (*Thunus albacares*), inoculated in sterile tuna matrix and stored 13 days at 8°C in plastic bags with air. The second one concerned the production conditions of enzymatic hydrolysates from salmon by-products and especially the effect of four processing parameters (time and temperature of hydrolysis, sugar and antioxidant addition) on the odour of the hydrolysates.

In these two case studies, we compared sensory map obtained with classical profiling (with trained panellists) and free sorting procedure performed with a mixed panel constituted of trained and untrained assessors in the microbiological study and untrained assessors in the process study.

Results show a rather good similarity of the two sensory maps and show the value of free sorting in the sensory characteristic description step, especially to avoid missing some descriptors. Moreover the free sorting can be used also to highlight effects of process parameters on sensory characteristics.

Results from these case studies showed that compared to quantitative and descriptive analysis, the holistic approach of sorting task could clearly constitute an interesting way for screening parameters in the first steps of a research project. The free sorting seems efficient for industrial applications and product development.

Key words: sensory method, free sorting, quantitative descriptive analysis, **odour, salmon by-product hydrolysate, bacterial spoilage**

Sensory characteristics of food products made from dulse (*Palmaria mollis)*

Presenting author: Michael T. Morrissey[1]

Co-authors: Jason Ball[1], Ann Colonna[1], Sarah Masoni[1], Chuck Toombs[2]

[1]Oregon State University Food Innovation Center, Portland, OR, USA

[2]OSU School of Business

There has been increasing interest in the use of seaweeds as a food source for human consumption. A relatively new area for research has been the use of seaweed in value added culinary preparations intended for commercialization.

In the present study, dulse obtained from a sustainable on-land cultivation system was used to develop five value-added culinary prototypes: salad dressing (DRE), chips (CHP), crackers (CRK), trail mix (TRM), and brittle (BRI). Prior to tasting, panelists were presented with a concept card for each sample. Concept cards presented nutrition data, images, health claims, and background information for each product sample. Sensory preferences were determined using hedonic scales amongst panelists (n=116) who were screened to be consumers and likers of seaweed and seaweed products. Additional comments regarding individual attributes were also collected.

Overall, the samples were scored favorably by the panelists. In terms of overall liking, the CRK ranked highest although there was no significant difference ($P<0.05$) between CRK and DRE, while the CHP scored the lowest. Significant differences ($P<0.05$) among prototypes for each attribute were also reported. Purchase intent varied, and was largely influenced by the taste and appearance of each samples as significant differences ($P<0.05$) were found for the purchase intent of each concept (prior to tasting) compared to after tasting. Furthermore, it was found that there was an inverse relationship between price and purchase intent. Impact of nutritional information, and importance of health claims and packaging information to consumers will be discussed.

Dulse grown by aquaculture methods was found to be readily accepted by U.S. consumers. This creates a strong potential for growing dulse and commercialization of dulse products for the US market.

Key words: dulse, seaweed, sensory evaluation, product development

Health and nutrition claims' effects on consumer perception of omega-3 enriched convenience meals

Presenting author: Pall Arnar Hauksson[1]

Co-authors: Kolbrun Sveinsdottir[2], Kyösti Pennanen[3], Fridrik Bjornsson[4], Emilia Martinsdottir[2]

[1] University of Iceland – Faculty of Food Science and Nutrition, Reykjavik, Iceland

[2]Matis ohf – Icelandic Food and Biotech R&D

[3]VTT Technical Research Centre of Finland

[4]University of Iceland – Faculty of Business

Health and nutrition claims are effectively used by food manufacturers to communicate health benefits, but such claims do not necessarily make products more appealing from the consumers' perspective. The aim of this study was to evaluate consumers' perception of omega-3 enriched ready meals with and without health and nutrition claims on the packaging label.

Participants (n=117) evaluated omega-3 enriched ready meals in a home-use-test. They were randomly assigned into two groups and received two different types of enriched ready meals (fish, vegetable) to be consumed within one week. One group received meals with conventional labels while the second group had labels with nutrition claims stating high content of omega-3 fatty acids with related health claims. The respondents answered questions on overall liking, perceived health benefits, product attractiveness and their willingness to pay in an online questionnaire after the consumption. Furthermore, the consumers answered questions regarding their attitudes concerning health, convenience and functional food. Sensory evaluation by a trained panel was performed on the meals using generic descriptive analysis.

Mean scores for both meals were consistently in favor of health claim labels for all questions. Group differences in attitudes towards health, convenience and functional foods were not found significant.

The results of this study will support functional food development and provide understanding on how receptive consumers are toward information conveyed by health claims.

Key words: health claims, nutrition claims, consumer behavior, functional foods, omega-3

Biogenic amine content of fish products sold in Turkish market

Presenting author: Ali Serhat Özkütük[1]

Co-authors : Esmeray Kuley Boga[2], Gulsun Ozyurt[2], Merve Erguven[2], Hakan Hayta[3], Serhat Ozkutuk[4]

[1]Department of Seafood Processing Technology, Faculty of Fisheries, Cukurova University, Adana, Turkey.

[2] Cukurova University, Faculty of Fisheries, Department of Seafood Processing Technology, Adana, Turkey

[3]Agriculture and Rural Development Support Institution, Mersin, Turkey

[4]Cukurova University, Vocational School of Yumurtalık, Adana, Turkey

Biogenic amines (BAs) in foods are mainly produced by the microbial decarboxylation of amino acids. Their presence is undesirable because of the toxicological effects to consumers The amount of BAs formed are influenced by factors such as microbial growth, availability of free amino acids, the presence of decarboxylase enzymes and elevated temperature conditions. Some technological processes such as salting, ripening, fermentation or marination can increase the possibility of formation of BAs. To protect consumer health, it is very important to monitor the concentration of biogenic amines in food. In this study, total seventeen fish products available on Turkish retail market were collected and investigated for the content of biogenic amines.

Fish products collected in Turkish retail market were frozen (Alaska pollock, anchovy fillets, sardine fillets, hake fillets, Pangasius fillets, Atlantic bonito, Atlantic salmon), smoked (mackerel, trout, Atlantic salmon), marinated (dried mackerel, anchovy, anchovy with red pepper sauce and anchovy with hot pepper sauce), smoked and marinated (mackerel and anchovy) fish samples. Sample preparation and derivatisation procedure for biogenic amine analysis were made according to Özogul et al. (2002). Biogenic amine analyses were performed using a Shimadzu Prominence HPLC apparatus. The column was a reverse-phase, ODS Hypersil, 5 µ, 250 x4.6 mm.

There are significant differences in biogenic amine content among fish products ($p<0.05$). Putrescine, spermidine, trimethylamine, spermine and dopamine were the most abundant BAs whilst phenylethylamine and histamine were minor BAs. Putrescine content was in range 3.60 mg/100g in frozen Alaska Pollock and 33.53 mg/100g in frozen Atlantic salmon. Fish products contained less than 19.87 mg/100g of cadaverine. The limit of 10 mg/100g for histamine in fish products as suggested by European Union (EU) were not exceeded in any samples analysed. Tyramine levels in fish products were below 10 mg/100 g. Frozen Atlantic salmon, marinated anchovy with hot pepper sauce and smoked marinated mackerel had the highest levels of tyramine, with corresponding values of 8.5, 5.27 and 3.5 mg/100 g.

Biogenic amine content in commercially produced fish products should be monitored as an important quality index. The mean histamine levels in all analyzed fish products were below the tolerance limit established by EU regulation. The application of international standards of food hygiene is crucial to ensure the quality and safety of fish products.

Key words: Fish products, biogenic amine, histamine, tyramine, Turkey

Antioxidant and functional properties of shrimp hydrolysates and its application to fish tofu

Presenting author: Sunantha Ketnawa[1]

Co-authors: Oscar Martínez Alvarez[2], Soottawat Benjakul[3], and Saroat Rawdkuen[4]

[1]Food Technology Program, School of Agro-Industry, Mae Fah Luang University, Chiang Rai 57100, Thailand (e-mail: saroat@mfu.ac.th)

[2]Institute of Food Science, Technology and Nutrition (ICTAN-CSIC), José Antonio Novais 10, Madrid, 28040, Spain

[3]Department of Food Technology, Faculty of Agro-Industry, Prince of Songkla University, Songkhla, 90112, Thailand

[4]Food Technology Program, School of Agro-Industry, Mae Fah Luang University, Chiang Rai, 57100, Thailand

Non-commercial cooked Whiteleg shrimp (*Penaeus vannamei*) was valorized by application of hydrolysis treatments with different proteases (Giant catfish viscera proteases, commercial trypsin, and Alcalase) for the obtainment of shrimp protein hydrolysates (SPH). Then application of the obtained hydrolysates on fish tofu was also investigated.

The hydrolysis of cooked shrimp was performed by using 8 units of each enzyme per protein content dissolved in buffer solution (1:3 (w/v) in 0.1 M Tris-HCl pH 8.0), at 50°C for 2 hours. Hydrolysis was controlled by using a pH-stat. Aliquots were collected at 0, 10, 20, 40, 90, and 120 minutes. After enzyme inactivation by boiling, the supernatants were lyophilized, stored at -20°C and referred to SPH. The hydrolysates obtained were determined for antioxidant properties including 2,20-azinobis-(3-ethylbenzothiazoline-6-sulfonic acid) scavenging capacity (ABTS), ferric reducing power capacity (FRAP), and metal ion chelating activity (MICA). The SPH at the levels of 0.05, 1.00, and 2.00% (w/w) were added into fish tofu. Quality attributes of fish tofu were monitored during storage at 4°C for 14 days.

SPH that were hydrolyzed by viscera proteases, commercial trypsin and Alcalase showed excellent solubility (>97%) in a wide pH range (3-10), good oil binding capacity (0.86-1.83 g oil/g hydrolysates), and discrete inter-facial properties. All SPH showed the activity in ABTS, FRAP, and MICA as 217-552 as ascorbic acid equivalents/g sample, 5.55-18.21 as µmol Fe^{2+}equivalents/g sample and 37-45 as µmol EDTA equivalents/g sample, respectively. In application study, the addition of SPH affected the texture profile of fish tofu by lowering firmness and toughness (P<0.05). For lipid oxidation by thiobarbituric acid-reactive substances (TBARS), fish tofu containing higher levels of shrimp hydrolysate had lower TBARS value (P<0.05). TBARS values ranged of 2.58 to 5.30 mg MDA/ kg sample during 14 days storage.

The enzymatic hydrolysis of non-commercial cooked shrimp could be a useful way to valorize with interesting biofunctional properties for food applications. SPH at a level of 2% (w/w) addition can be applied to improve the texture and extend the shelf-life of fish tofu by retardation of lipid oxidation. Thus, SPH could be an alternative natural substance for oxidative stability enhancement.

Key words: shrimp hydrolysates, antioxidants, functional properties, lipid oxidation, fish tofu

Fish protein hydrolysates processed with different natural antioxidants

Presenting author: Dana Rán Jónsdóttir

Co-authors: Sigrún Mjöll Halldórsdóttir

Matís ohf., Reykjavík, Iceland

Researches indicate that peptides processed from fish have interesting bioactive properties, such as antioxidant and blood pressure regulating properties, among others. Despite this, few products are found on the market. This is mainly due to quality problems connected to rancidity, and that the products tend to have characteristic fish taste and smell that are not desirable in food supplements. The aim of this project was to add value to by-products of fish processing by producing fish protein hydrolysates (FPH), containing bioactive peptides and maximize their quality in terms of sensory factors and bioactive properties.

FPH were produced from two different versions of isolated protein. The following antioxidants were tested: *Ascophylum nodosum* extract, *Fucus vesiculosus* extract, *Rosmarinus officinals* extract and a mixture of α-tocopherol and L-ascorbic acid. Hydrolysis were carried out with the enzyme Protease P from Amano and degree of hydrolysis was measured with the OPA method. Stability during storage was tested at three different temperatures; -80°C, 2°C and 22°C for 6 months. Oxidation measurements (PV and TBARS) were done every 6 weeks and sensory evaluation every 12 weeks. Bioactivity was measured at the beginning and at the end of the study using four different *in vitro* oxidation measurements, cellular antioxidant assay and ACE inhibitory assay. Dendritic cell assay was performed at the beginning of the study.

Results showed that addition of antioxidants slowed down oxidation during production of FPH. According to the sensory evaluation results, samples with added antioxidants gave better results than samples with no antioxidants. The results also showed that samples with added antioxidants had more antioxidant activity then control samples. FPH with added *Fucus vesiculosus* extract was the only sample that showed potential anti-inflammatory activity.

The results indicate that addition of antioxidants during processing increase quality and bioactivity of FPH.

Key words: Fish protein hydrolysates, enzyme hydrolysis, lipid oxidation, antioxidant activity, antioxidants.

Identification of peptides in a prolyl oligopeptidase-inhibiting fraction from trypsin hydrolysate of Pacific white shrimp (*Penaeus vannamei*).

Presenting author: Oscar Martínez Alvarez

Co-authors: Pilar Montero, María del Carmen Gómez-Guillén

Institute of Food Science, Technology and Nutrition, Madrid, Spain

Prolyl Oligopeptidase (PO) plays an important role in the degradation of biologically active peptide hormones and neuropeptides that contain proline. Different studies have suggested a relationship between increased PO activity and neurodegeneration and disturbances in memory and cognition. PO-inhibitors have been identified in plant extracts and also in different protein hydrolysates from fish or milk.

Pacific white shrimp are among the most widely cultivated shrimp in the world. Frozen head-on, head-off, and peeled shrimp were formerly the major products for export to the main global markets of United States of America, European Union and Japan. The trend now is for processing of shrimp to obtain value-added products. This processing generates different by-products such as muscle trimmings, carapaces, etc. that could be processed in mild conditions to obtain healthy molecules with interest as nutraceuticals. The objective of this study was to optimize the obtaining of PO-inhibiting molecules from muscle trimming of shrimp, and also to identify the peptides that could be implied in the PO-inhibiting process.

Muscle trimmings from Pacific white shrimp were hydrolyzed with trypsin at different pH values and temperatures, and the obtained hydrolysates were tested for PO-inhibiting activity. The hydrolysate that showed the best PO-inhibiting activity was selected and the peptides responsible for this activity were partially purified by SEC and RP-HPLC. UPLC-LTQ-Orbitrap and Uniprot database were further used to identify the peptides in the RP-HPLC fraction that showed the highest PO-inhibitory activity.

The best PO-inhibitor was obtained at pH 9 and 37ºC. The inhibiting potency of this hydrolysate was improved after fractionation by SEC and RP-HPLC. The fraction that showed the best activity was composed by low-molecular-weight peptides (482-1608 Da). This fraction was rich in hydrophobic residues (mainly alanine, proline and valine), serine and lysine.

Interesting PO-inhibiting hydrolysates can be obtained from Pacific white shrimp by using trypsin, and the peptides responsible for this activity contain at least 4 residues. The presence and position in the peptide chain of alanine, proline, valine, serine and lysine might play an important role in the PO-inhibiting ability of these peptides.

Key words: prolyl oligopeptidase, hydrolysates, trypsin, enzymatic inhibition, shrimp

Determination of some functional properties of enzymatically hydrolyzed protein from head of sea bass (*Dicentrarchus labrax*) and its effect in whiting (*Merlangius merlangus*) mince

Presenting author: Şebnem Tolasa Yilmaz

Co-authors: Şükran Çakli, Nida Demirtas Erol,Ömer Alper Erdem, Evren Burcu Şen Yilmaz,Arzu Burcu Yavuz

Ege University, Faculty of Fishery, Department of Fish Processing Technology, Izmir-Turkey

Aquaculture is one of the fastest growing industries in Turkey. Every year a considerable amount of processing leftovers that includes trimmings, fins, frames, heads, skin and viscera from aquacultered products are discarded. It is important to use those resources primarily for human consumption. Processing leftovers may be used as food ingredients due to the capability of their functional properties. The objective of this study was to determine the functional properties of the enzymatically hydrolyzed protein from head of sea bass and determine the effect of addition of FPH in different concentration on functional properties in whiting mince.

Fish protein hydrolysate was prepared from head of sea bass which cultered in Egean Sea by using alcalase according to the method of Bougatef *et al.*, 2010. Degree of hydrolysis was measured using the method of Benjakul and Morrissey (1997). In hydrolized protein some functional properties including; solubility, water-holding capacity, emulsification, and foam-forming ability and also textural changes were investigated. The resulted hydrolysate was added to whiting mince with different concentrations and the same functional properties.

There are many researches about fish protein hydrolysate in literature including; quality, functionality and antioxidative properties. The effectiveness of fish protein hydrolysate in controlling functional properties and texture changes dependent upon the concentration of hydrolysate which was added to fish mince. Hydrolysate from head of sea bass might be used in food systems as a natural additive.

- This study was supported by TÜBİTAK 112O925 project.
- Arzu Burcu YAVUZ as a member of the working team makes use of PhD scholarship program TUBITAK 2211-C for the priority fields.

Key words: protein hydrolysate, by products, functional properties, fish mince, sea bass, whiting

Opportunities in the blue bioeconomy

Keynote speaker: Sveinn Margeirsson

Co-author: Hörður Kristinsson

Matis – Icelandic Food and Biotech, Reykjavik, Iceland

While 70% of earth is covered with water, it´s a source of only 4% of our global food supply. There exist major opportunities with respect to more and better utilization of aquatic biomass. At the same time, we face numerous challenges with this valuable resource. Efforts to utilize aquatic biomass need to be sustainable and driven by science and market demand. The presentation will discuss past and present successes and failures in the Blue Bioeconomy, as well as focus on potential growth opportunities and how technical and market innovation in the blue bioeconomy may create new jobs and increase value creation and enhance regional development.

Key words: Blue Growth, Commercialization, Innovation, Business models, regional development

Replacement of surimi in restructured crab by raw crabmeat in restructured crab products

Presenting author: David Green

Co-authors: Tyre C. Lanier, Yiwen Thor

Food Bioprocessing and Nutrition Sciences, NC State University, Raleigh NC USA

The blue crab (*Callinectes sapidus*) industry in eastern USA has been declining due to the high labor cost of hand picking cooked crabmeat. Mechanically separated raw crabmeat can generate 3x higher yield at very low labor cost, but the product form is a paste, not the desired flaky crabmeat prized for making the meaty 'Chesapeake- style' crab cakes. US consumers buy over 200 million lbs/yr of analog crabmeat made from fish surimi; however, if this is used to make crab cakes they must then be labeled 'imitation' or as containing fish meat. If the heat-induced gelation properties of raw crabmeat were adequate, it could be economically substituted for fish surimi in restructured crabmeat production to eliminate 'imitation' labeling on restaurant menu items.

Heat gelation properties as affected by endogenous protease and transglutaminase (TGase) activity, and as augmented by added microbial TGase were assessed. EDTA was also used to inhibit endogenouse TGase. Protease/TGase activity were favored by different heating regimes. Gel properties at fracture were measured by a ball probe and changes to myosin heavy chain (MHC) assessed by SDS-PAGE. Crab 'analog' formulations typically used for surimi-based products were produced substituting raw crabmeat for surimi.

Raw blue crabmeat exhibited both endogenous protease and TGase activities. EDTA inhibited both of these, indicating that a metalloprotease was active. Added microbial TGase enhanced gel strength. Restructured crabmeat products could be made having comparable properties to those made from surimi with proper adjustment of the formulation and use of a thermal process that optimized TGase activity.

Gelling properties of raw crabmeat are poor compared to most commercial fish surimi but likely adequate to produce a restructured product which would not be labeled as 'imitation' crab.

Key words: raw, crabmeat, analog, gelation, enzyme

Study of the optimization conditions for hydrolysates production from *Scyliorhinus canicula* muscle and antioxidant activities

Presenting author: María Blanco[1]

Co-authors: Águeda Massa[2], José A. Vázquez[3], Carmen González Sotelo[1], Ricardo I. Pérez-Martín[1]

[1]Grupo de Bioquímica de alimentos (IIM-CSIC), Vigo (Spain)

[2]Instituto Nacional de Investigación y Desarrollo Pesquero (INIDEP), Mar del Plata (Argentina).

[3] Grupo de Reciclado y Valorización de Materiales Residuales (IIM-CSIC), Vigo (Spain)

Small spotted catshark (*Scyliorhinus canicula*) is a highly discarded species (100% intraspecific discard rate in some fisheries). Discards has been recognized as unsustainable fishing practices (EU Regulation 1380/2013) not only because of their impact on economic and ecological questions but also because they promote a significant waste of potential food resources. According to the Blue Growth initiative of the European Commission, it would be indispensable to have tools for recovering bioactive molecules from discards and other underutilized fishery resources such as byproducts, to support the sustainable growth of scarce natural marine resources and to increase the fish supply without an intensification of fishing pressure. Fish protein hydrolysates have been previously proposed as an alternative use for upgrading fish biomass. The aim of the present study was to optimize the enzymatic hydrolysis catalyzed by different enzymes for obtaining protein hydrolysates from S. canicula muscle wastes, and to study the antioxidant properties of the resultant hydrolysates. Muscle of S. canicula was enzymatically hydrolysed using Alcalase, Esperase and Protamex. The experimental development was performed using second order rotatable designs and evaluated by response surface methodology combined with a previous kinetic approach. The kinetics of muscle hydrolysis in each experimental condition were accurately fitted to the Weibull equation. Kinetic parameters obtained from this mathematical model (maximum hydrolysis and maximum rate of hydrolysis) were used as response variables of the factorial design. Results showed that Alcalase and Esperase were the unique enzymes successfully hydrolyzing the muscle of this species and their optimal values of pH and temperature were pH 8.77, 70.39ºC for Esperase and pH 9.4 and 64.63ºC for Alcalase (in this case only for the maximum hydrolysis). Antioxidant activities were studied using crocin and β-carotene bleaching methods in a single combined assay. The DPPH radical scavenging activity of hydrolysates was also tested.

Development of an ultrasound-assisted enzymatic hydrolysis process for the liquefaction of the red seaweed *Grateloupia turuturu* and its biomolecules recovery

Presenting author: Cécile Le Guillard[1, 2]

Co-authors: Jean-Pascal Bergé[3], Régis Baron[1], Justine Dumay[2]

[1]Ifremer Laboratory Biorafhe , Nantes, France

[2]University of Nantes Laboratory MMS, Nantes, France

[3]IDMer, Lorient, France

Among the numerous processes used in marine biotechnologies, enzymatic hydrolysis exhibits a great potential to improve extraction of bioactive compounds from seaweeds. In recent years, several articles in various research areas put forward the improvement and simultaneous stimulation of the enzymatic hydrolysis by ultrasound (UAEH). Indeed, the use of ultrasound in conjunction with enzymes can increase the yields of extraction of valuable biomolecules from plants, compared to conventional methods. Currently, there are very few works on this combined process on seaweeds, particularly wet ones.

Our work is focused on underexploited, proliferative alien red seaweed found on Brittany coasts, *Grateloupia turuturu.* The goal of our study is to develop an UAEH process using 4 industrial enzymes and an original ultrasonic flow-through reactor. The experiments of simultaneous combination enzymes/sonication (UAEH), sonication and enzymatic hydrolysis were conducted in triplicate, two temperatures were tested (40-22 °C).

Our results highlight the great potential of UAEH in seaweed liquefaction rather than enzymes or sonication alone. After 4 hours of this treatment at 40 °C, up to 90 % of primary material was recovered into soluble phase. Biochemical analyses of those resulting phases revealed an enrichment in carbon and nitrogen compounds, more precisely, carbohydrates and amino acids.

This original and innovative process is clearly an efficient procedure for the liquefaction of wet seaweeds, enabling the recovery of valuable components.

Key words: seaweed, enzymes, sonication, process, liquefaction

An example of French strategy for the production of marine ingredients from salmon by-products: the Pesk&Co project

Presenting author: Margot Provost[1]

Co-authors : Roxane Fagon[2], Marc Hémon[2], Vincent Gélamur[4] & Fabienne Guérard [1]

[1] University of Bretagne Occidentale, Laboratoire des Sciences de l'Environnement marin, LEMAR UMR6539, Rue Dumont D'Urville, Technopôle Brest Iroise, F-29280 Plouzané, France

[2] Yslab, 2 rue Félix Le Dantec, F-29000 Quimper, France

[3] SPF - DIANA, ZA du Gohélis, F-56250 Elven, France

[4] Meralliance, 55 avenue de Keradennec, F-29000 Quimper, France

Marine biological resources such as fish by-products are sources of valuable ingredients and bioactive molecules. Their exploitation using biotechnological tools is likely to lead to the development of new markets and industries, specifically in the areas of food, petfood, feed, cosmetics, and, in some cases, therapeutic agents.

For the first time in France, four companies located in Brittany on the West coast of France (Meralliance, Yslab, SPF-DIANA and AGH-Socofag, respectively) and the University of Bretagne Occidentale (Laboratory of Marine sciences - LEMAR UMR 6539) gathered their expertise to set up an integrative Research and Development project.

The Pesk&Co project aims to produce marine ingredients from salmon by-products such as structure proteins and functional peptides for food, aquafeed, cosmetics and biomedical applications.

The Pesk&Co strategy is based on :
(1) The implemention of a sustainable supply chain, fully traceable, and of biotechnological processes to embrace the total salmon by-products, from raw materials available to market.
(2) The creation of a Research and Development center based on efficient multidisciplinary cooperation between academic researchers and private companies from the 5 partners of Pesk&Co project.

Specific salmon by-products were chosen to apply this strategy. The first step was to find innovative processes at lab scale to allow, in a second step, a transfer at pilot and industrial scales.

We showed that in using a fresh controlled process we are able to extract a high quality native collagen with a clean treatment, which has allowed the removal of a patent. Simultaneously, analytical methods were developed in order to control quality and traceability of ingredients derived from production. In the longer term, the objective is to ensure compliance with the rules for commercialization and further developments. We demonstrated, what could be the definition and characterization of a high molecular weight native collagen from skins for several uses in cosmetics and medical devices, in a dried final product, and the capability to certify that each lot produced will be what it says it is.

On the other hand, hydrolysates were developed and incorporated into fish diet to improve larval development.

Results obtained, competences and technical installations developed within this project, will contribute to position the Brittany Region strategically within the overall market of innovative marine ingredients through a long term outlook.

Partners: http://www-iuem.univ-brest.fr/UMR6539/ http://www.meralliance.com/ http://www.diana-petfood.com/

http://www.yslab.fr/ http://www.groupe-glon.com/

Key words: salmon by-products, collagen, peptides, marine ingredients, innovative strategy

Advancements in land-based brackish-water and marine aquaculture and their future in seafood production

Keynote speaker: Thomas M. Losordo[1,2]

[1]Ph.D., Principal Scientist & Chief Engineer, Pentair Aquatic Eco-Systems Inc.

[2]Professor Emeritus, Department of Biological & Agricultural Engineering, North Carolina State University

Mailing Address: 0308 SW Montgomery St, Apt. 404, Portland, OR 97201, USA

Email: Tom.Losordo@Pentair.com

Mobile Phone: +1 919 264 3812

Aquaculture has become a major source of seafood over the past 40 years. A good example can be found in the Salmon farming industry. Major centers of production can be found in Northern Europe and Chile and to a lesser extent the Pacific Northwest of North America and Tasmania in Australia. Much of this production centers on large-scale, ocean-based net-pen production technologies for the on-growing of market sized fish. From the beginning of the industry Smolts, used to stock the net-pens have been raised on land in tanks using flow-through technology. Both of these technologies have encountered issues related to disease, environmental impacts and the overall sustainability of the production process.

Recirculating Aquaculture Systems (RAS) have been in development during the same 40 year period. RAS technology, designed correctly, has the potential to significantly reduce the environmental impacts from aquaculture. However, this technology has not found as wide acceptance as net-pens for the on-growing of fish. This is mostly due to substantially higher investment costs for RAS technology. Recently, there has been a shift to using RAS technology for the tank-based production of large so-called “Super-Smolts” that are grown up to 1 kg on land before placing them in ocean-based net-pens. This combination of technologies reduces the time “at sea” and reduces the overall environmental “footprint” of the production process.

Other species being reared in net-pens in Europe are the European or Mediterranean Sea Bass. New technologies are being deployed to reduce the environmental impact of these net-pen culture activities. This presentation will describe these technologies, new species of interest and associated environmental issues. We will discuss future directions of land-based RAS technology and how they can support or in some cases replace ocean-based technologies to lead to a more sustainable and diverse seafood production industry.

Oyster refinement: effects of algae diets (*Skeletonema costatum* and *Rhodomonas baltica*) on the sensory characteristics and volatile organic compounds of Pacific cupped oysters (*Crassostrea gigas*)

Presenting author: Jasper van Houcke[1]

Co-authors: Isabel Medina[2], Mireille Cardinal[3],Josiane Cornet[3], Jozef Linssen[4], Joop Luten[4]

[1]HZ University of Applied Sciences, Vlissingen, The Netherlands

[2]CSIC-IMM

[3]Ifremer

[4]WUR

Oyster refinement is common practice in France. The oysters are kept in land-based systems where they are usually fed with relative high concentrations of algae during a period varying from one till four months in order to increase the weight of the oyster tissue. A few studies have shown the impact of the diet on the sensorial profile and the volatile organic compounds (VOCs) of Pacific cupped oysters. We have studied the impact of the algae *Skeletonema costatum* or *Rhodomonas baltica* on the sensorial characteristics and on the VOCs of the Pacific cupped oyster originally from the important Dutch cultivation site Lake Grevelingen.

Market-sized Pacific cupped oysters (60-80 g) were fed with either the diatom *Skeletonema costatum* or the flagellate *Rhodomonas baltica* for a period of seven weeks at 30 mg dry weight algae day^{-1} $oyster^{-1}$ as a refinement diet. The oysters were refined in land-based pond systems. In order to make comparisons with non-refined oysters reference oysters from Lake Grevelingen were sampled as well.

After four and seven weeks oysters were sampled for sensory evaluation as well as the analysis on VOCs. For the sensory evaluation a Quantitative Descriptive Analysis with a trained panel (n=15) has been used. Panelists were asked to score on 18 attributes on a 10 cm unstructured line. The VOCs were extracted by Headspace Solid Phase Micro Extraction and analyzed by Gas Chromatography-Mass Spectrometry.

Sensory scores showed significant differences ($P<0.05$) between reference oysters, *Skeletonema* fed oysters and *Rhodomonas* fed oysters for overall odor intensity (4.99, 4.14 and 3.26, respectively), fullness of the shell (3.95, 5.35 and 5.61, respectively) and algae flavor (1.71, 2.29 and 1.33, respectively) after 4 weeks of refinement, while significant differences ($P<0.05$) for color of the visceral mass (4.74, 2.74 and 3.15, respectively), fullness of the shell (3.49, 5.80 and 6.09, respectively), marine flavor (5.65, 4.79 and 4.04, respectively) and salt flavor (4.64, 3.20 and 3.71, respectively) were found after 7 weeks of refinement. Preliminary results of the VOCs showed a decrease in the total amount of VOCs over time. More detailed results about individual VOCs are under evaluation.

Refinement has an effect on the sensorial characteristics of Pacific cupped oysters. Odor, taste and appearance are different for the refined oysters in comparison with reference oysters. Differences between the sensorial characteristics of *Skeletonema* and *Rhodomonas* fed oysters are less apparent.

Key words: Pacific cupped oyster, refinement, volatile organic compound, sensory evaluation, quantitative descriptive analysis

Chemical and microbiological contamination in Japanese-clam (*Ruditapes phillipinarum*) from Tagus estuary (Portugal)

Presenting author: Sónia Pedro

Co-authors: Helena Lourenço, Patrícia Oliveira, Patrícia Presado, Fernanda Oliveira, Susana Gonçalves, Helena Silva, Maria Fernanda Martins

Portuguese Institute for Sea and Atmosphere (IPMA, I.P.) Av. Brasilia, 1449-006 Lisbon, Portugal

Tagus River Estuary (ETJ) is the largest estuary in Portugal and has several uses, including fishing and aquaculture, in spite of being subject to industrial and urban pollution, with evidences of lead contamination and high faecal levels.

The production of bivalve molluscs in this ecosystem has increased in recent years due mainly to the introduction of exotic species with high adaptability and growth in this habitat, particularly the Japanese-clam (*Ruditapes phillipinarum*). This outgrowth has contributed to the decline of the bivalve indigenous species populations.

According to the rules of hygiene of foodstuffs, the bivalve mollusc production areas are classified according to the level of bacteria of fecal origin, such as *E. coli* (Regulation 854/2004). Moreover, levels of chemical contaminants in bivalves for human consumption have to be lower than the legal limits, namely cadmium (Cd), lead (Pb) and mercury (Hg) (Reg. EC 1881/2006, with amendments).

Given the lack of information about this exotic species sanitary status, the main purpose of this study was to evaluate the chemical and microbiological contamination of Japanese-clam (*Ruditapes phillipinarum*) from ETJ in reference and new sampling points, identifying potential hot spots.

Preliminary results of Japanese-clam for Cd, Hg, and Pb revealed levels below the legal limits, respectively 1.0, 0.50 and 1.5 mg/kg, although some samples presented Pb levels close to 1.0 mg/kg. Concerning *E. coli* (MPN/100 g), levels between 1300 and 54000 were obtained in different sampling points. These data support the strategy of redefining ETJ limits in order to exclude hot spots, contributing to safer use of these resources.

The authors acknowledge project "AMETEJO" from PROMAR Program for funding.

Key words: bivalves, metal contaminants, microbiological contamination, Tagus river

New possibilities for coproducts from Atlantic salmon backbones and bits & pieces in fish spread

Presenting author: Grete Hansen Aas[1]

Co-authors: Trygg Barnung[2], Jan Thomas Rosnes[3], Wenche Bjørkeng[4], Dagbjørn Skipnes[3], Jorunn Hansen[3]

[1]Aalesund University College, Aalesund, Norway

[2]Møreforsking

[3]Nofima

[4]Sandanger

Based on results from an innovation process, it was concluded that fish spread could be a promising product in Norway. In a traditional Norwegian diet, both breakfast and lunch meals consist of bread and spread, but at the retail market there are few spread available based on seafood. It is documented that raw materials from salmon contain high amounts of protein and lipids. Therefore, a project was initiated between research institutes and seafood industrial partners, to develop new spreads based on material from fish, including elements in the value added chain from raw materials to the consumer.

Backbones and bits & pieces from farmed Atlantic salmon were used as a raw material for developing the new fish spread. The raw material was collected at a salmon slaughterhouse and frozen in blocks and stored. At slaughtering of salmon, a batch of backbones and bits & pieces were frozen in 20 kg blocks and stored at -20°C. Thawing and storage of raw material are important processes to conserve quality through the production line. For production of spread, these blocks were thawed and skin and bones were separated from the meat, to make a fish mince. The mince was sampled and proximate analyses of protein, lipid, water and ash were performed in addition to oxidation analyses by free fatty acids and thiobarbituric acid and total bacteria count.

During five months of frozen storage, the oxidation was low. FFA increased from 1% to 1.5% during storage and TBARS increased from 30 to 57. A preliminary sensory evaluation showed an acceptable, fresh odour after thawing, although the mince became paler after 5 months of storage. The total bacteria counts showed that the material was acceptable.

To include backbone and cuttings of salmon, it is of importance to stabilize the raw material to secure a certain content of lipid, protein and water to predict stable consistency of the mince.

New observations of the brown meat after deep skinning also results in a good raw material as fresh, but the stability during storage are challenging. The industrial partner developed shelf stable prototypes based on the minced products and tested them for different market groups. Optimalizations were performed in regards to water holding capacitites, consistency and fish odour.

During the presentation, the utilisation of salmon raw material besides the fillet will be given and presented in a sustainable and resource perspective with focus on possible new innovations.

Key words: Atlantic salmon, coproduct, oxidation, frozen storage, spread

Seafood processing innovation and entrepreneurship : challenges and opportunities

Keynote speaker: David Green

North Carolina State University Center for Marine Sciences and Technology Department of Food, Bioprocessing and Nutrition Sciences, Raleigh, North Carolina USA

World demand for seafood has never been greater. Consumer interests are being fueled by research on the longevity and good health of the people in Japan, Finland, Iceland, and Scandinavia largely due to the nutritional profile of fish and other seafood. Market demands, however, are being influenced by consumer perceptions in a number of areas including quality and safety concerns. This presentation reviews some of the challenges and opportunities in seafood processing innovation and entrepreneurship related to the incidence on quality and safety. Seafood industry needs are identified within six main drivers: safety; quality and value; nutrition, health, and well-being; resilience and efficiency; environmental sustainability; and skills and knowledge. Recent developments in seafood research to help increase the world's food supply and feed the nine billion people projected to inhabit the earth by 2050 are provided.

Key words: seafood processing, quality, safety, innovation, entrepreneurism

Quality and safety of tropical yellowfin tuna (*Thunnus albacares*) steaks stored under air, vacuum or modified atmosphere

Presenting author: Adèle Dauchy[1]

Co-authors: Jean-Jacques Joffraud[1], Sandra Adenet[2], Frédérique Chevalier[1], Nadine Therese-Basile[2], PARM, Katia Rochefort[2], Françoise Leroi[1]

[1]Ifremer,Laboratory EM[3]B, rue de l'Ile d'Yeu, BP 21105, 44311 Nantes Cedex 3, France

[2]PARM

In Martinique, Yellowfin tuna (*Thunnus albacares*), one of the main pelagic caught, is generally captured around the fish aggregating devices (FAD), gutted, iced on board and sold along the road sometimes more than 48 hours after fishing. The objectives of this study were to analyse the microbial ecosystem of *T. albacares* with metagenomic tools and to investigate the effect of packaging conditions on quality and safety.

A yellowfin tuna caught off the Caribbean coast of Martinique was bled and gutted on board, sliced and transported to the laboratory. Steaks were divided into three batches. Batch 1: steaks were individually iced in plastic bags (AIR). Batches 2 and 3: steaks were respectively vacuum (VP) and modified atmosphere (70%CO_2, 30%O_2) packed (MAP) and stored at 4/8°C. Physico-chemical, sensory, bacteriological and metagenomic analysis were carried out in triplicate at predetermined time intervals until samples were organoleptically unacceptable.

AIR and VP steaks were considered unacceptable after 13 days and MAP steaks after 15 days. At the rejection point, the total bacterial counts were 10^{6-7} cfu g^{-1} in all batches. According to pyrosequencing of the 16S-rRNA gene and Polymerase Chain Reaction – Temporal Temperature gradient Gel Electrophoresis (PCR-TTGE) analysis, *Brochothrix thermosphacta* dominated the microbial ecosystems of all batches. However, this species was associated with *Pseudomonas* spp. in AIR samples and with *Enterobacteriaceae* and lactic acid bacteria in VP products. The pH value remained stable in the 3 batches, ranging from 5.77 to 5.97. Total volatile basic nitrogen and trimethylamine productions were weak and not significantly different between trials. Lipid oxidation significantly increased in the samples containing O_2 with a maximum value of 6.00 ± 0.75 mg-malonaldehyde kg^{-1} for spoiled MAP products. At the beginning of the experiment, the concentration of histamine was high (75-78 mg kg^{-1}) and stable up to 8 days, but then significantly decreased in all batches to reach a value around 25-30 mg kg^{-1}. The bacterial counts were lower than what is generally found in spoiled finfish from temperate-water. Although the microbial ecosystems of the 3 batches were different, VP and MAP did not significantly increase the shelf life of tuna steaks. However, samples were stored at 4°C the first week and then at 8°C, according to the French standard shelf-life validation of perishable and refrigerated food, instead of 0°C for AIR samples, so packaging may allow a significant energy reduction during storage.

Key words: tropical fish, spoilage, microbial ecosystem, histamine, metagenomic

The effect of storage temperature on *Vibrio parahaemolyticus* numbers in Pacific oysters (*Crassostrea gigas*): Evaluating a predictive model under New Zealand conditions

Presenting author: Graham C. Fletcher

Co-authors: Cristina Cruz, Marta Chycka, Swapna Gannabathula, Gerald Dsa, Nicola Wei

New Zealand Institute for Plant & Food Research Limited, Auckland, New Zealand

Vibrio parahaemolyticus naturally occurs in seafood and when present in high numbers, some strains cause gastroenteritis. This study was carried out to evaluate the effect of storage at different temperatures on numbers of naturally occurring total *Vibrio parahaemolyticus* and to compare the results with those predicted by the Oyster Refrigeration Index software (Australian Seafood Cooperative Research Centre), a worst-case model intended to account for variability and to be pre-emptive towards protecting public health.

Three experiments were conducted over New Zealand summers (late February) in 2013, 2014 and 2015. In 2013, one set of samples (10-12 oysters) was assessed per time point, while in 2014 and 2015, two sets were assessed. Pacific oysters (*Crassostrea gigas*) were harvested from the same site located in the North Island of New Zealand. Samples were shipped cold (c. 10°C) to the laboratory. On arrival, oysters were incubated at different temperatures (5, 10, 15, 20, 25 and 30°C). Three sets of 10-12 oysters were sampled immediately (T0) and the remaining oysters were sampled on at least at four subsequent occasions. Multiplex real-time polymerase chain reaction (qPCR) analyses were conducted to confirm the identity of *V. parahaemolyticus*, and total and pathogenic populations of *V. parahaemolyticus* were estimated using the Most Probable Number (MPN) technique. The data were analysed using the DMFIT Model (ComBase).

Initial total *V. parahaemolyticus* populations averaged 160, 40 and 76 MPN/g in 2013, 2014 and 2015 respectively. *V. parahaemolyticus* showed minimum growth (or decline) when stored for up to 9 days at temperatures below 15°C. Numbers increased during storage above 15°C but not as fast as predicted by the Australian model. The observed growth rates increased with increasing temperatures: 0.0240, 0.0244 and 0.0597 $\log_{10}$ MPN/h at 20, 25 and 30°C respectively. We observed maximum increases of 2 $\log_{10}$ after 54 hours at 25°C and 2.5 $\log_{10}$ after 48 h at 30°C. The Australian model predicted higher *V. parahaemolyticus* numbers at 25 and 30°C, and lower numbers at 5°C and 10°C compared with those observed in our study. Predicted and observed data were similar at 15°C.

Naturally occurring *V. parahaemolyticus* populations under New Zealand conditions responded to storage temperature differently from those used to develop the Australian model. Risk from *V. parahaemolyticus* should not increase as long as product is stored below 15°C.

Key words: *Vibrio parahaemolyticus*, storage temperature, predictive model, growth rate, Pacific oyster (*Crassostrea gigas*)

Cold Storage stability of minced fish added of grape antioxidant dietary fiber pomace

Presenting author: Helena M. Moreno Conde[1]

Co-authors: Armando Solari Godiño[2], Jara Pérez Jimenez[3], Fulgencio Saura Calixto[3], A. Javier Borderías Juárez[1]

[1]Institute of Food Science Technology and Nutrition. Dpt. Products (ICTAN-CSIC). C/José Antonio Nováis 10, 28040 Madrid, Spain (e-mail address: hmoreno@ictan.csic.es)
[2]Instituto Tecnológico de la Producción - ITP. Dirección de Investigaciones Tecnológicas para la Transformación Pesquera –DGITTP, Carretera a Ventanilla Km 5.2. Ventanilla. Callao, Perú
[3]Institute of Food Science Technology and Nutrition. Dpt. Metabolisn and Nutrition (ICTAN-CSIC). Madrid, Spain

Wine industry produces a great amount of grape pomace, which is made up of peels and seeds. This natural by-product is rich in antioxidant dietary fiber (AODF), with high antioxidant capacity derived from associated phenolic compounds. The objective of this work was to add different proportions of this fiber in minced fish to preserve lipids from oxidation.

AODF was obtained from two Peruvian grapes (*Vitis vinifera)* varieties *Quebranta* (red) and *Torontel* (white) after the cool drying process of the grape pomace (using a dehumidifier at 18-30°C with 8,2 L per hour for 28-36 hours) after removing sugars and other soluble constituents. Dietary fiber content (DF), polyphenol content and associated antioxidant capacity by two complementary methods (FRAP, ferric reducing/antioxidant power, and ABTS, 2,2'-azino-bis(3-ethylbenzothiazoline-6-sulphonic acid) were determined. An initial Threshold Test was conducted in order to determine the maximum concentration of the AODF that would be acceptable. Semi-fatty fish muscle from horse mackerel (*Trachurus trachurus*), caught in June, was mince in a meat mincer. Then, AODF of both grape varieties at the selected concentrations (2% and 4%) was added to the muscle and then blended. Resulting product was kept wrapped in aluminium foil at 5ºC till the analysis day (1, 3, and 8). The analysis performed were: Water binding capacity (WBC), mechanical properties (hardness), color determination (L*, a*, b*) and antioxidant capacity by FRAP and ABTS assays.

AODF from grape pomace was characterized by high DF content (> 60%), mostly insoluble, as well as a relevant polyphenol content (> 30%). The incorporation of AODF (2-4%) to mackerel muscle induced an increase in the antioxidant capacity of the restructured product during the cold storage, as showed ABTS assay. On the other hand, WBC was not improved with the addition of AODF regardless the grape variety, concentration or storage period, so no extra water binding ability is exhibited probably because fiber is very insoluble. The increase in L*, a* and b* was in line with the increasing concentration of AODF. Hardness of the minced added of AODF is increased as the proportion of AODF is included.

AODF concentrates can be obtained by cool drying from grape pomace. AODF incorporation into minced fish showed potential to be used as an additive to retard oxidative degradation. This strategy may allow the revalorization of both grape pomace and fishery by-products letting to get enriched in fiber restructured seafood products.

Key words: antioxidant dietary fiber, cold dryness, fatty fish restructured products, fatty fish oxidative stability

Quality improvement of cooked brown shrimp *Crangon crangon* trough detailed kinetic studies of the major quality attributes.

Presenting author: Thomas Verhaeghe[1]

Co-authors: Geertrui Vlaemynck[2] , Jan De Block[1], Marc Hendrickx[3]

[1]Institute for Agricultural and Fisheries Research (ILVO) - Technology and Food Science Unit, Melle, Belgium
[2]Institute for Agricultural and Fisheries Research (ILVO) - Technology and Food Science Unit, Melle, Belgium
[3] Katholieke Universiteit Leuven - Department of Microbial and Molecular Systems (M2S), Leuven, Belgium

The brown shrimp (*Crangon crangon*) is in Europe amongst the top 5 species for the Dutch, German and Belgian fishery fleet and has a significant economic and social importance in the North Sea fisheries. Brown shrimp are traditionally cooked and cooled on board of the vessel by means of seawater (±3min, 80-100°C). As fishermen don't have a standard cooking process (considering cooking time, temperature and salt content), major quality differences and high yield losses can occur. Because by cooking, shelf-life is prolonged and a ready-to-eat product is created, changing the cooking parameters to optimize yield, holds the risk of losing other quality attributes. In this study, the thermal inactivation kinetics of the most important spoilage enzymes, proteases and polyphenoloxidase (PPO), were determined. The doneness of the product was determined by observing the heat-induced denaturation of muscle proteins. The denaturation kinetics of the muscle proteins were determined and protein stability was compared to cook loss and protein loss.

Enzyme inactivation kinetics and protein denaturation kinetics were determined by isothermal heating of enzyme extracts and muscle tissue, respectively. Cook loss and protein loss was determined by heating intact shrimp. Next to treatment time and temperature, influence of the salt content was determined on protein denaturation, cook loss and protein loss.

Enzyme activity was measured by adding the treated extracts to a substrate and by measuring the reaction products spectrophotometrically. Muscle protein denaturation was studied by differential scanning calorimetry (DSC), only actin was considered as most heat stable muscle protein. Cook loss will be determined by measuring mass differences and protein content will be determined according to the Kjeldahl method.

Both enzymes and muscle protein decay could be described by a first order model. The decimal reduction time for PPO was the lowest, indicating its low thermostability. For proteases, two stability fractions were found. Only actin denaturation was considered as it was the most heat stable muscle protein. All kinetic data show that, at high temperatures, proteolytic enzymes will be the most important boundary condition for further process optimization. Preliminary results show high cook losses when with increasing salt contents and cooking times.

The kinetic data presented, show that cooking parameters used by processors are higher than necessary for sufficient inactivation of proteases and PPO as well as for actin denaturation. For further process optimization, protease inactivation can be used as a boundary condition, reducing the total heat load of the process. In this way unnecessary yield losses can be avoided.

Key words: Quality, shrimp, enzymes, proteins, yield

The effects of nanoemulsions based on commercial oils (sunflower, canola, corn, olive, soybean, and hazelnut oils) on sensory, chemical and microbial quality of frozen sea bass fillets

Presenting author: Deniz Ayas[1]

Co-authors: Yesim Ozogul[2], Mustafa Durmus[2], Esra Balıkçı[2], Deniz Ayas[1], Saadet Gökdoğan[2], Yılmaz Ucar[2]

[1]Mersin University, Faculty of Fisheries, Department of Seafood Processing Technology, Mersin, Turkey

[2]Cukurova University, Faculty of Fisheries, Department of Seafood Processing Technology, Adana, Turkey

Nanotechnology focuses on the characterization, fabrication, and manipulation of biological and non-biological structures which are smaller than 100nm. The application area of nanotechnology in food industry is fortification of food and new product development. Recently, nanotechnology has been introduced to the food industry to address issues relevant to food and nutrition. Nanoparticules having antimicrobial properties prolong shelf life of food without damaging flavor and color changes. Although the potential use of nanotechnology by food industry is possible, studies in this field, especially for seafood are limited. Oil-in-water emulsions are important vehicles for the delivery of hydrophobic bioactive compounds into a range of food products. The preparation of nanoemulsions with small droplet size is of particular interest, leading to a creamier mouth feel and greater emulsion stability. Therefore, the aim of this work is to investigate the effects of oil-in-water nanoemulsions using sunflower oil on sensory, chemical and quality of frozen sea bass (*Dicentrarchus labrax*) fillets. This project was supported by Scientific Research Projects Unit of Çukurova University (Project No : SÜF2013BAP19)

Physical properties of nanoemulsion were analyzed in terms of viscosity, particle size of droplets, thermodynamic stability, refractive index, and surface tension. Sea bass were obtained from a local fish farm in İzmir, Turkey. They were immediately gutted and divided into two lots. One lot was treated with nanoemulsion and the other was used as the control. All samples were put in a freezer bag and stored in a freezer (-18°C). Sensory, chemical (total volatile basic nitrogen (TVB-N), thiobarbituric acid index (TBA), peroxide value (PV) and free fatty acids (FFA), and microbiological qualities (mesophilic aerobic bacteria, total psychrophilic bacteria, and total *Enterbactericaea* bacteria) of sea bass (*Dicentrarchus labrax*) fillets were investigated.

According to results of sensory analyses, the treated groups had higher sensory score than the control. Chemical parameters were observed to be significantly ($p<0.05$) lower in the treated groups compared to those of the control. TVB-N levels did not exceed the limit level (35 mg/100 g) during seven months of storage period. There were fluctuations in the levels of PV and FFA whereas TBA values were found lower than the recommended value for all groups. Nanoemulsion had also a positive effect in the delay of microbial growth.

The impact of nanoemulsion based on sunflower oil for the quality of frozen sea bass was strong, improving organoleptic quality parameters during storage period.

Key words: nanoemulsions, *Dicentrarchus labrax*, shelf life, frozen, quality

Investigation of ohmic heating for seafood processing

Presenting author: Stina Frosch

Co-authors: Søren Juhl Pedersen, Aberham Hailu Feyissa

National Food Institute, Division of Food Technology, Technical University of Denmark, Søltofts Plads 227, DK-2800 Lyngby, Denmark

Steam cooking is one of the main process steps in production of cooked shrimps. The natural variation in size and composition of shrimps within one batch pose a challenge to the traditional cooking process, as the small shrimps are overcooked to comply with legislation for the big shrimps. In short, the traditional thermal processing is far from optimal when treating food pieces of varying size in the same batch. Ohmic heating (OH) is one of the technologies potentially solving this problem by allowing volumetric heating of the product and thereby reducing or eliminating the effect of size. The application of OH has been studied for a wide range of foods (Kaur & Singh, 2015; Knirsch et al. 2010; Sastry, 2008; Vargheseet al. 2012). However, the research in relation to seafood processing is sparse and restricted to the use of OH for thawing of shrimps (Roberts et al. 1996). In this study the effect of different process parameters during OH of shrimps has been investigated. The measured responses were: 1) the heating time until the set core temperature (72 ^{o}C) of the shrimps were reached, 2) weight loss and 3) texture profile.

A batch ohmic heater was used for the experments which was built by BCH ltd. (UK). The OH unit consists of a holding cell with variable size adjustment and mountings for temperature loggers. Raw frozen shrimps (*Pandalus Borelias)* were supplied by Royal Greenland A/S (DK). Different mass ratios (mass of shrimps/mass of water) were used. The shrimps were cooked in brine with varying salt conc. and at different voltages, and the time to reach a core temperature of 72 °C were measured. Texture profile analysis and press juice (PJ) were performed with a Texture Analyzer XT.Plus (Stable Micro Systems Ltd. UK). The yield was calculated as the difference of the weight of the shrimps before and after cooking.

It was found that both salt conc. and voltage were significant for the processing time ($P < 0.05$) and there was no interaction between the two process parameters. In the statistical data analysis it was found that salt conc. were of significance ($P < 0.05$) for the PJ. Improved water-holding capacity and thereby lower weight loss seems to be influenced mostly by the content of salt in brine rather than the application of voltage ($P_{salt} < P_{voltage}$). When fixing the voltage and varying the mass ratio and salt conc. it was observed that the mass ratio had no effect ($P > 0.05$) on the processing time, only the salt conc. did. No major differences in the textural parameters were observed when comparing with varying process parameters. However, the textural measurements showed a comparable texture of the shrimps to that of conventionally cooked shrimps reported in other studies (Erdoğdu & Balaban, 2000).

The findings show a promising utilization of OH as a unit operation for the shrimp processing industries.

Key words : ohmic heating, cooking, shrimps, yield, texture

Suppressive effect of ATP on autoxidation of tuna oxymyoglobin to metmyoglobin

Presenting author: Kota Inohara[1]

Co-authors: Ikuo Kimura[2]

[1]Course of Biological Science and Technology(3rd grade student), United Graduate School of Agricultural Science, Kagoshima University, Kagoshima, Japan

[2]Faculty of Fisheries, Kagoshima University.

Myoglobin (Mb) is an oxygen-binding protein present in a wide variety of species. The discoloration of tuna meat from red to brown is induced by autoxidation of oxymyoglobin (MbO_2) to metmyoglobin (metMb) during storage in ice or frozen. On the other hand, the discoloration of highly fresh tuna meat could effectively be suppressed even if stored at -20˚C. However, the suppressive mechanism of the discoloration is not well understood.

The dorsal ordinary muscles of frozen specimens of southern bluefin tuna, which were caught and landed on deck in an alive state were purchased from a tuna fishing company in Kushikino, Japan and stored at -80˚C until they were used for experiments. Mb was isolated from dorsal ordinary muscle as reported by Ochiai *et al.* with some modification. The ratio of metMb to total Mb was determined essentially according to Bito by using the ratio of absorbance at 540 and 503 nm.

We have studied the effects of ATP on the autoxidation rate and molecular structure of tuna Mb measured with spectral perturbation in the soret region, quenching of fluprescence, dynamic light-scattering and zeta-potential measurements. concentration.

The autoxidation of southern bluefin tuna Mb at 25˚C was suppressed in the presence of ATP especially in acidic pH range. Mixing ATP with Mb induced a spectral perturbation in the soret region of Mb. This spectral perturbation was observed as a function of the ATP.

Quenching of Mb fluorescence was also caused by ATP, saturating at around 0.5 mM ATP. According to dynamic light-scattering measurements the molecular weights of tuna Mb changed from 15.5 to 11.3 kDa with ATP and zeta-potential measurements gave also a negative surface charge without ATP and a positive one with ATP, respectively.

The above results indicate that ATP-induces changes in the conformational structure of Mb. The effects of ATP on Mb could thus provide a possible mechanism to regulate the autoxidation of Mb.

Key words: ATP, autoxidation, metmyoglobin, suppression, tuna myoglobin

Effect of heat treatments on mobility and *in vitro* infectivity of *Anisakis* L3 in hake muscle infected under controlled conditions

Presenting author: Isabel Sánchez-Alonso[1]

Co-authors: Mercedes Careche[1], Margarita Tejada[1], Miguel González Muñoz[2], Noelia Carballeda-Sangiao[2], Alfonso Navas[3], Angel Mendizábal[4]

[1]Institute of Food Science, Technology and Nutrition, Spanish National Research Council (ICTAN-CSIC), c/ Jose Antonio Novais 10 28040 Madrid, Spain
[2]Hospital Universitario La Paz (HULP)
[3]Museo Nacional de Ciencias Naturales (MNCN-CSIC)
[4]Technical Unit of Mercamadrid, Public Health Institute, Madrid-Salud

The importance of Anisakis infection of fish is well recognized by the fisheries sector and food safety authorities, since consumers may be accidentally infected by larvae in the third stage (L3) after consumption of raw or undercooked fish or cephalopods carrying the parasite, causing anisakiasis and allergic sensitization. Anisakis L3 is moderately tolerant to heat stress and in order to mitigate the risk of the intake of live larvae in cooked seafood products is important to define with precision at which point the parasites are no longer infective after a heat treatment, since overcooking may decrease sensory acceptance of fish muscle whereas undercooking may lead to health problems. The aim of this work was to study the effects of heating rate and final temperature to find minimal thermal treatments to inactivate Anisakis simplex.

Live A. simplex L3 were obtained from heavily parasitized hake ovaries and viscera, washed with 0.85% NaCl and stored at 4 ºC until use. Hake muscle was infected by placing 15 L3 Anisakis on steaks of aprox. 1 cm thickness and covered with another steak of the same size (200g). Each sandwich was wrapped in aluminium foil and larvae allowed to migrate (4 ºC, 24 h). Sandwiches were introduced in plastic bags and heat treatments were performed (oven or water bath) in the range of 40-80 ºC, at different heating and cooling rates. Anisakis viability (motility and fluorescence) and in vitro infectivity (agar penetration test) were monitored.

In the oven (set at 200 ºC) none of the larvae were viable after 20 min heating and those which survived after 10 min were not able to penetrate into agar. When sandwiches were heated in a water bath (set at 95 ºC), no larvae were viable beyond 70 ºC; from those which survived in the range 55-65 ºC, none of them penetrated into agar whereas at temperatures close to 50 ºC, half of those showing mobility were able to penetrate into agar.

Although more studies are needed to define with precision the minimal thermal treatments, these results stress the importance of using not only mobility as a measurement of viability but also measurements such as the agar penetration test as an indicator of the larval penetrability, since this ability may play an important role for the invasion of the gastrointestinal mucosa.

Acknowledgements: This work has been financed by the Project FP7-312068 EU PARASITE.

Key words: *Anisakis simplex*, heat treatment, viability, agar penetration test, hake

Differences between *A. simplex s.s.* and *A. pegreffii*: *in vitro* infectivity and freezing tolerance

Presenting author: Mercedes Careche[1]

Co-authors: Isabel Sánchez-Alonso1, Margarita Tejada[1], Miguel González Muñoz[2], Noelia Carballeda-Sangiao[2], Alfonso Navas[3]

[1]Institute of Food Science, Technology and Nutrition, Spanish National Research Council (ICTAN-CSIC), c/ Jose Antonio Novais 10 28040 Madrid, Spain

[2]Hospital Universitario La Paz (HULP)

[3]Museo Nacional de Ciencias Naturales (MNCN-CSIC)

Freezing of fish is one of the most efficient technological treatments to kill *Anisakis* larvae in the third stage (L3), but, although data are available for *Anisakis s.l.* there has been no systematic studies of the effect on different *Anisakis* species. Previous results from our group have shown in experimentally infected mince fish that viability of *Anisakis s.s.* larvae is affected by freezing rate, final freezing temperature & storage time, so that at fast freezing rates all L3 can be non viable even at -10 ºC. The aim of this work was to compare the ability of penetration in agar of *A. simplex s.s. vs A. pegreffii* and to study the effects of freezing rate and final freezing temperature on the mobility of *A. pegreffii* in order to compare the possible differences in infectivity of this species with the previously reported *A. simplex s.s.*

Live *A. simplex s.s.* and *A. pegreffii* L3 were obtained from heavily parasitized fish viscera. They were washed with 0.85% NaCl and stored at 4 ºC until use. A total of 100 larvae were placed in flasks (10 L3/flask) containing 0.75% agar and a layer of simulated gastric juice. They were allowed to penetrate into agar in an incubator at 37 ºC in 5% CO_2. The number of larvae penetrated into agar was monitored at 1, 3, 6, 20, 24, and 92h. Minced hake muscle was infected with L3 *A. pegreffii* (10 L3/ 75g mince) and subjected to three freezing rates until final temperatures of -10 and -15 ºC were reached. The time temperature profiles were recorded per each experimental unit. After thawing, viability of larvae was monitored. Species identification was performed for all the batches studied.

The *in vitro* infectivity measured by the agar penetration test at 37 ºC showed that *A. pegreffii* had slightly lower penetration ability than *A. simplex s.s.* The percentage of survival of *A. pegreffii* decreased with increasing freezing rates as it was found previously for *A. simplex s.s.* but the viability at a given freezing rate or final time was lower for *A. pegreffi*.

These experiments show that *Anisakis pegreffii* is less resistant than *Anisakis simplex s.s.* in terms of penetration ability in agar and freezing tolerance.

Acknowledgements: This work has been financed by the Project FP7-312068 EU PARASITE.

Key words:*Anisakis simplex s.s.*, *A. pegreffii*, freezing, viability, agar penetration test

Prevalence of nematodes (*Anisakidae*) in fish species most consumed in France

Presenting author: Véronique Verrez-Bagnis[1]

Co-authors: Yuwalee Seesao[2-3], Anne Thebault[4], Mélanie Gay[2], Cécile-Marie Aliouat-Denis[3], Bruno Le Fur[5], Diane Cuzzucoli[4], Isabelle Cos[5], Marc Jérôme[1], Laetitia Kolypczuk[1], Sandrine Bruzac[1], Christophe Audebert[6], Eduardo Dei-Cas[3], Eric Viscogliosi[3] and the 'Fish-Parasites' consortium

[1]Ifremer-BRM-EM[3]B, Nantes, France
[2]Anses-Laboratory for Food Safety, Boulogne-sur-mer, France
[3]IPL-CIIL-BDPEE, Univ Lille Nord de France, Lille, France
[4]Anses-DER-UMA-MSA, Maisons-Alfort, France
[5] Plateforme d'innovation Nouvelles Vagues, Boulogne-sur-mer, France
[6]Gènes Diffusion, Lille, France
[7]Hôpital Cochin, Paris, France

Among parasites frequently present in edible fish, nematode larvae belonging to the *Anisakidae* family occur in numerous fish and cephalopods. These larvae may induce digestive or allergic symptoms in human. To better define the impact of fish parasites on consumers' health and to improve the safety of fish products, we set up the French national Fish-Parasites network (ANR-10-ALIA-004, http://fish-parasites.com/en/). One objective of this project was to determine *Anisakidae* prevalence in fish sampled on the basis of risk ranking.

Fifteen species of fish were sampled in North East Atlantic and Mediterranean Sea according to a risk-ranking analysis using prevalence data from literature and French consumer consumption data, and modulated with data on the proportion of sold fish. A total of 1795 fish were sampled during sea research cruises and inland. Anisakid identification relied on 2 methods: individual Sanger sequencing or pooled High Throughput Sequencing (HTS) based on Cox2 fragments. HTS method was developed and validated during the project.

No *Anisakidae* were isolated from 44 % of all the fish sampled whereas 34 % of all fish had Anisakidae only in their viscera and 24 % had *Anisakidae* either in their fillets only or in their fillets and viscera. The most infested fish species are saithe (*Pollachius virens)*, megrim (*Lepidorhombus whiffiagonis)*, cod (*Gadus morhua)*, anglerfish (*Lophius piscatorius)*, whiting (*Merlangius merlangius)* and hake (*Merluccius merluccius)*, but the *Anisakidae* infestation is highly dependent from the fishing area. Nematodes were mostly identified as belonging to the genus *Anisakis*, mainly *A. simplex*, but also *A. pegreffii*. Species belonging to other genera such as *Pseudoterranova*, *Contracaecum* and *Hysterothylacium* were also identified.

All these data were compiled in a specific database, and would be used subsequently in risk assessing and prevention strategies.

Key words: *Anisakidae*, Risk ranking, Nematodes, Fish, Prevalence

Packaging, quality and shelf life of fillet products from live captured and stored Atlantic cod

Presenting author: Heidi Nilsen[1]

Co-authors: Birgitte Moen[2], Marit Kvalvåg Pettersen[2], Solveig Langsrud[2], Anlaug Ådland Hansen (PhD)

[1]Nofima AS, Muninbakken 9-13, Tromsø, Norway

[2]Nofima AS, Osloveien 1, 1430 Ås, Norway

Atlantic cod is a key species in the Norwegian seafood industry, but the biology and migratory patterns imply large seasonal variations in catch. Additionally, consumers demand fresh fish with high quality throughout the year, capture-based aquaculture of cod has therefore received high attentions. Cod is processed into many different product forms (e.g., fresh and frozen fillets, stockfish, salted and dry-salted, i.e. clip-fish) and is exported to a wide range of nations and market segments. The domestic market for cod is also important, especially for fresh cod fillets. As one of the work packages in the "CATCH" project, an interdisciplinary project financed by The Research Council of Norway, the objective is to obtain new knowledge that can contribute to more market-oriented applications of different cod products related to packaging technology, product quality and shelf life. Characterization of the microbiota on fillets from live stored cod was studied, in addition to the spoilage potentials of selected bacterial isolates, and how packaging conditions can inhibit growth of these bacteria.

Atlantic cod, that had been live captured at end of May 2014 outside northern coast of Norway (Finnmark County), were 1) either slaughtered and pre-rigor filleted at site two weeks after catch; for detection of microbiota and bacteria isolates with spoilage potentials, or 2) slaughtered about 8 month after catch (post-rigor filleted at Nofima's laboratory) for artificial contamination of cod products using a selection of bacteria with spoilage potential. For the latter trial different packaging conditions were used. All handling were performed manually with an optimal hygiene. Microbiota profiling using high-throughput sequencing of bacterial 16S rRNA amplicons (MiSeq, Illumina) was performed, in addition to traditional cultivation method. Volatile components (dynamic headspace GC-MS), odor acceptance, liquid loss and texture were also measured.

Bacteria isolated from pre-rigor filleted samples mainly belonged to the *Pseudomonas* and *Photobacterium* genera. Selections of strains within these two genera were used to inoculate samples of the post-rigor fillets. Preliminary results showed that the choice of packaging conditions affected the growth of spoilage bacteria, presence of volatile compounds and sensory quality.

The results have shown that the choice of packaging conditions is important to preserve the quality and shelf life of cod fillets. Further analyses will show how the natural microbiota and the inoculated strains were affected by the different packaging conditions, and the correlation to sensory profile and volatile components.

Key words: modified atmosphere packaging (MAP), bacteria, volatile components, shelf life, cod

Heat resistance of the most isolated spore-forming bacteria in ready-to-eat brown crab meat

Presenting author: Santiago Condón-Abanto[1]

Co-authors: Federico Lian[1], Cristina Arroyo[1], Paul Whyte[2], Nigel Brunton[1], James-G Lyng[1]

[1]School of Agriculture and Food Science, University College Dublin, Belfield, Dublin 4, Ireland

[2]School of Veterinary Medicine, University College Dublin, Belfield, Dublin 4, Ireland

Brown crab (*Cancer pagurus*) is one of the most appreciated and consumed crustaceans in European countries, especially in the south countries such as France, Spain, Italy and Portugal. Although, this type of seafood has been traditionally sold alive, in more recent times, new ready-to-eat brown crab products and whole pasteurized crabs have been launched into the market. However, industrial practices have not evolved to keep pace with these developments, the net effect of which can be sub-optimal quality products. For this reason this research was performed to identify the microbial flora present at the end of the shelf-life (i.e total viable count >6 logarithmic cycles) in brown and white meat of entire edible crab which was pasteurized and stored under vacuum in chilled conditions. Subsequent to this, the thermal resistance of the main flora were determined with a view to developing superior heat processes. 16S rDNA gene sequencing technique was used to identify the microbial flora through shelf-life. Sequencing revealed that spore-forming bacteria such as *Bacillus mycoides*, *Bacillus weihenstephanensis* and *Psychrobacillus psychrodurans* are the most identified species in the pasteurized edible crab.

The heat resistance of these three species was determined at different temperatures ranging from 80ºC to 105ºC. Ginafit Excel tool was used to fit the Geeraerd equation "Log-linear+shoulder" to the microbial inactivation curves. While *B.weihenstephanensis* was found to be the most resistant species requiring 11.36 min to reduce 4 log cycles at 102.5ºC with a z value of 7.1ºC; *B.mycoides* was the most heat sensitive requiring 5.1 min at 90ºC to reduce 4 log cycles with a z value of 8.8ºC.

This study revealed that spore-forming bacteria from *Bacillus* genus are the most significant microbial contaminant in pasteurized edible crab. Therefore, new heat treatments for ready-to-eat brown crab products should be designed with these organisms in mind.

Key words: brown crab, Bacillus, spore-forming bacteria, heat resistance, ready-to-eat

Minimizing the content of free and ester bound 2,- 3-MCPD and esterified glycidol in fish fingers

Presenting author: Sybille Merkle[1]

Co-authors: Horst Karl[2], Jan Kuhlmann[3], Jan, Fritsche[1]

[1]University of Applied Sciences Hamburg, Hamburg, Germany

[2]Max Rubner-Institut, Department of Safety and Quality of Milk and Fish products, Hamburg, Germany

[3]SGS Germany GmbH – Weidenbaumsweg 137 – 21035 Hamburg, Germany

Free and ester bound 2- and 3-Monochloropropanediol (2-/3-MCPD) and esterified glycidol are food processing contaminants are formed during the production of thermally processed foods [1,2,3]. (Pre-) frying is an important process for the production of breaded and pre-fried frozen fish products. For frying, edible oils are used, representing another potential source of free and ester bound 2-/3-MCPD and esterified glycidol. Additionally breaded and pre-fried frozen fish products contain a relatively high salt content, which may also have impact on the formation of these process contaminants. Limited data on the occurrence of these process contaminants in foods other than edible fats and oils is published. Therefore EFSA is actually calling for additional occurrence data in foods including fishery products [4].

A modified method of Jira (2010) and Küsters et al. (2011) was applied for determination of free 2- and 3-MCPD. The validated AOCS methods Cd 29b-13 and Cd 29c-13 were modified in order to quantify ester bound 2- and 3-MCPD and esterified glycidyl esters in fishery products [5].

In the present study contents of free and ester bound 2-/3-MCPD and esterified glycidol in breaded and pre-fried frozen fish products have been investigated. Influences of different parameters such as the frying temperature, type of the frying oil, salt content and frying times on the formation of free and ester bound 2-/3-MCPD and esterified glycidol in breaded and pre-fried frozen fish products have been examined.

Data of free and ester bound 2- and 3-MCPD and esterified glycidyl in various fishery products will be presented.

[1] Weisshaar, R., Perz, R., Fatty acid esters of glycidol in refined fats and oils. *Eur. J. Lipid. Sci. Technol.* 2010, *112*, 158-165.

[2] Zelinkova, Z., Svejkovska, B., Velisek, J., Dolezal, M., Fatty acid esters of 3-chloropropane-1,2-diol in edible oils. *Food Addit. Contam.*. 2006, *23*, 1290-1298.

[3] Kuhlmann,. J., Determination of bound 2,3-epoxy-1-propanol (glycidol) and bound monochloropropanediol (MCPD) in refined oils. *Eur. J. Lipid Sci. and Technol*. 2011, *113*, 335-344.

[4] EFSA (European Food Safety Authority) (2013). EFSA Journal 2013;11(9):3381.

[5] Karl, H., Merkle, S., Kuhlmann, J., Fritsche, J. Development of analytical methods for the determination of free and ester bound 2-, 3- MCPD and esterified glycidol in fishery products. *Eur. J. Lipid Sci. Technol.* 2015, *117*.

Key words: ester bound 2-MCPD, ester bound 3-MCPD, esterified glycidol, fish fingers, food processing contaminants

Utilization and stability of cod liver during frozen storage – Effects of season, on-board handling and storage conditions

Presenting author: María Gudjónsdóttir[1]

Co-authors:Dang Thi Thu Huong[1], Hildur Kristinsdottir[1], Magnea Karlsdottir[2], Sigurjón Arason[1,2]

[1]University of Iceland, Faculty of Food Science and Nutrition, Vínlandsleid 14, IS-113 Reykjavík, Iceland

[2]Matis ohf, Icelandic Food and Biotech R&D, Vínlandsleid 12, IS-113 Reykjavík, Iceland

The utilization of rest raw materials from round fish processing has grown immensely during the last 20 years. These raw materials are very important for the industry since great economic, nutritional and environmental gain can be obtained by increasing the yield of raw materials in general. In Iceland, cod liver has been utilized mainly for production and exportation of cod liver oil, but canned cod liver has also gained more popularity in recent years. The quantity of exported cod liver oil from Iceland has increased about 68% during the last decade, resulting in 80% higher value. Demands for high quality raw material are therefore growing, but today a considerable amount of cod liver is imported for further processing. Processing of fish liver can be challenging due to the currently used storage conditions and handling. The liver is generally handled like a by-raw-material, often in a rougher manner than the high quality fish fillets. Therefore the fish liver is also generally more sensitive towards deterioration. The presence of highly polyunsaturated fatty acids in cod liver makes them highly susceptible to oxidation. Moreover, due to a high endogenous enzymatic activity, lipids are rapidly hydrolysed, leading not only to loss of essential fatty acids but also to loss of fat-soluble vitamins, which can have considerable effect on the quality of the final product.

In the current study lipid degradation of cod liver during frozen storage was studied, where the effects of storage temperatures (-18°C/-24°C), packaging methods (vacuum packing/regular plastic bag) and seasonal variations (April/July) were evaluated. Moreover, the effects of various fish bleeding treatments (bleeding performed in one or two steps or without bleeding) on the storage stability of cod liver during frozen storage were investigated. For this, lipid composition, lipid hydrolysis, lipid oxidation and colour were analysed. Increasing lipid hydrolysis and oxidation were observed for most of the samples throughout the frozen storage. Vacuum packing and lower frozen storage temperature had a stronger preservative effect on lipid degradation at both seasons. Higher lipid hydrolysis could be observed in cod liver captured in July than in its counterpart from April. Based on the present results, both the packing method and storage temperature have a significant effect on lipid hydrolysis and oxidation in frozen cod liver. However, the effect from the different bleeding methods on the storage stability of cod liver was negligible.

Key words: Cod liver; bleeding; freezing; temperature; packaging

Conversion of lysine to cadaverine by cell-free supernatants (CFSs) from *Salmonella* Paratyphi A, and *Escherichia coli* in Lysine enriched decarboxylase broth (LDB)

Presenting author: Fatih Ozogul[1]

Co-authors : Fatih Ozogul[1], Nurten Toy[2], Ilyas Ozogul[2]

[1]Cukurova University, Faculty of Fisheries, Department of Seafood Processing Technology, Adana, Turkey

[2]Feke Vocational School, Cukurova University, Adana, Turkey

Conversion of Lysine to cadaverine by *Salmonella paratyphi* A, and *Escherichia coli* was investigated in Lysine enriched decarboxylase broth (LDB) using cell-free supernatants (CFSs) obtained from *Leuconostoc mesenterodies* subsp. *cremoris, Pediococcus acidilactici, , Lactococcus lactis* subsp. *lactis, Streptococcus thermophilus*.

LAB strains, which are *Lactococcus lactis* subsp. *lactis* IL 1403 (Lc. lactis subsp. lactis), *Leuconostoc mesenteroides* subsp. *cremoris* DSMZ 20346 (Leuc. mesenteroides subs. *cremoris*), *Pediococcus acidilactici* ATCC 25741 (*Pc. acidilactici*), and *Streptococcus thermophilus* NCFB 2392 (Str. thermophilus) were obtained from Sutcu Imam University (Kahramanmaras, Turkey) in BGML stock culture. The four selected foodborne pathogens were *Escherichia coli* ATCC 25922 (*E. coli*), which was bought from American Type Culture Collection (Rockville, MD, USA), and *Salmonella Paratyphi* A NCTC 13 (*S. Paratyphi* A), which was purchased from National Collections of Industrial Food and Marine Bacteria (Aberdeen, UK).Two groups of cell-free supernatants (25 or 50%) and control (only LDB) were prepared to determine cadaverine and other polyamines formation by foodborne pathogens (FBPs). Biogenic amine analysis was achieved according to the method of Ozogul et al. (2002) and measured in milligram amines per liter of broth.

Significant differences ($P < 0.05$) were observed among the species for each amine. All of the CFSs reduced the formation of cadaverine by ≤89% for *S. paratyphi* A following ≤68% for *E.coli*. The production of cadaverine and putrescine was exactly affected by the presence of CFSs, with the samples inoculated with *S. paratyphi* A and *E.coli*. The difference in polyamine was found with respect to the control samples. Spermine was produced in lower amount in *E. coli*, Spermidine was produced in *S. paratyphi* A whereas spermine and spermidine following serotonin, dopamine, tyramine and increased drastically in the major part of the samples concerning the control. Agmatine was characterized by a marked concentration decrease in all of the samples, and tyramine (TYR) was accumulated in very low concentrations in the controls. therefore, the ability of bacteria to produce certain biogenic amines such as histamine, tyramine, putrescine, and cadaaverine has been studied to assess their risk and prevent their formation in food products.

The results obtained from this study concluded that the lactic acid bacteria (LAB) strains with non-decarboxylase activity are capable of avoiding or limiting biogenic amine formation by FBP.

Key words: food-borne pathogen, cell-free supernatants, biogenic amines, LAB strains

Inhibitory effects of high pressure processing on *Morganella psychrotolerans* in herring (*Clupea harengus*)

Presenting author: Ilknur Ucak[1]

Co-authors: Nalan Gokoglu[1], Stefan Toepfl[2], Martina Kiessling[2]

[1]*Akdeniz University, Fisheries Faculty, Antalya, Turkey*

[2]German Institute of Food Technologies Quakenbrück, Germany

Fresh fish is one of the most perishable product, which spoils faster than the other meats. The main cause of deterioration is the activity of typical spoilage microorganisms, provoking loss of essentially fatty acids, fat-soluble vitamins and protein functionality, reduction of biogenic amines, and formation of off-odors. *Morganella psychrotolerans* is known to be a psychrotolerant specific spoilage organism, which is common in the marine environment. The use of refrigerated temperatures represents a useful mean to achieve lower rates of microbial growth but it is not a sufficient mild procedure to control the microbial spoilage in fish. High pressure processing (HPP) is a non-thermal technology used in the preservation of many food products since it has the capacity to inactivate product-spoiling microorganisms and enzymes at low temperatures without altering most of the organoleptic and nutrient characteristics of the product. The aim of this study was to evaluate the influence of high pressure processing on the growth of *M. psychrotolerans* in vacuum-packaged herring during storage at 4°C for 21 days.

Herring (*Clupea harengus*) fillets were obtained from a local market in Germany (Quakenbrück) and transported under ice. Fillets were rinsed with sterile distilled water. *M. psychrotolerans* was cultured in Tryptone Soya Broth at 25°C for overnight. Herring fillets were dipped in the solutions for 1 min to inoculate bacteria onto the surface of the fish flesh. The samples were pressure-treated in batches at 100, 200, 300, 500 and 600 MPa for 5 min. One batch (control) was left untreated. Samples were stored at 4°C during 21 days.

During the storage period, there were no significant differences among control, 100 MPa and 200 MPa pressure treated groups. At the end of the storage, *M. psychrotolerans* counts reached 8.21 log cfu/g, 8.14 log cfu/g and 8.16 log cfu/g in control, 100 MPa and 200 MPa treatment groups, respectively, while remained lower in 300 MPa treatment group. Until 7th day of storage (1.93 log cfu/g) no bacteria growing was observed in 500 MPa treatment group, furthermore significantly lower populations were determined in this group at the end of the storage (6.34 log cfu/g). *M. psychrotolerans* could not grow in 600 MPa pressure treated group throughout the storage.

500 MPa (until 7th day) and 600 MPa (during storage) high pressure processing has inhibitory effect on *M. psychrotolerans* in vacuum-packed hering during storage at 4°C for three weeks.

Keywords: *Morganella psychrotolerans,* high pressure processing, specific spoilage organisms (SSO), herring, fish spoilage

The effect of lactic acid bacteria isolated from fish on microbiological quality of silage made from fish processing waste

Presenting author: Serhat Ozkutuk[1]

Co-authors: Gulsun Ozyurt[2], Mustafa Boga[3], Esmeray Kuley Boga[2], Serhat Ozkutuk[1], Mustafa Durmus[2]

[1]Cukurova University, Vocational School of Yumurtalık, Adana, Turkey

[2]Cukurova University, Faculty of Fisheries, Department of Seafood Processing Technology, Adana, Turkey

[3]University of Nigde, Bor Vocational School, Nigde, Turkey

Lactic acid bacteria (LAB) is used successfully in the production of fermented foods for thousands of years. Many strains of LAB produce specific compounds with antimicrobial properties such as lactic acid and acetate, hydrogen peroxide, diacetyl and bacteriocins. The effect of LAB strains on microbiological quality of silage made from fish processing waste was investigated during 3 weeks. LAB was isolated from *Sparus aurata*, *Dicentrarchus labrax*, *Mugil cephalus*, *Cyprinus carpio* and *Silurus glanis* muscle, skin and gut. This project was supported by Scientific and Technological Research Council of Turkey (TOVAG-213O166).

LAB was isolated from fish and identified by 16S rRNA gene sequencing and PCR assay. Group 1 (control) included formic acid (3%) without any LAB inoculation. Group 2, 3, 4, 5 and 6 were *inoculated with E. gallinarum, Streptococcus* spp., *Lb. brevis, Lb. plantarum* and *P. acidilactici*, respectively. After minced of fish processing waste, addition of 15% molasses and inoculation of %5 LAB strains (10^8 cfu/ml) were made. All silage groups were stored at room temperature in plastic jar with caps and stirred daily until ripening.

Total aerobic and anaerobic viable counts were made on PCA plates for 2 days at 30ºC and in anaerobic jars at for 4 days 20˚C, respectively. LAB were grown on MRS agar at 30°C for 3–*5 days*. Total coliform bacteria *were* enumerated on *VRB agar*. Total yeasts and molds analysis were done on potato dextrose agar at 25°C for 3–5 days. *E. coli* enumeration was made in TBX medium for 18-24 hour at 37ºC. *Staph. aureus* was determined by Baird Parker agar. The presence of *Salmonella* spp. and *L. monocytogenes* was determined as described in the ISO 6579:2002 and ISO 11290-1⁄1996 method.

E. coli, *Salmonella* spp., *Staph. aureus* and *Listeria* spp. did not detect any silage group. Moreover, growth of mold and yeast was not observed. Although total coliform count of raw material was 3.48 log cfu/g, silage significantly inhibited coliform growth. Initial total aerobic (4.29 log cfu/g) and anaerobic (3.72 log cfu/g) count reached maximum level for group 3 and group 2, with corresponding value, 9.57 and 10.00 log cfu/g. Lactic acid bacteria count was in range 3.60 log cfu/g for control silage and 10.56 log cfu/g for group 5.

The study results revealed that lactic acid bacteria naturally present in fish could be suitable LAB starter cultures for production of safe fish silage.

Key words: lactic acid bacteria, fish silage, fish processing waste, microbial load, formic acid

Production of fish chips with using frozen saithe flesh (*Pollachius virens*) and determining its shelf life

Presenting author: M.Tolga Dincer

Co-authors: Hulya Sargin, Arzu Burcu Yavuz, Burcak Pir, M. Cagil Ucok

Ege University Faculty Of Fisheries, Izmir,Turkey

This study is about the production of fish additive potato chips and determining its shelf life. The goal is to increase the seafood consumption rates in Turkey; with this aim integration of fish flesh to a highly consumed potato product was realized. Study was planned to develop and produce commercially viable products which will contribute to the national and international economy. The differences of the fried and oven baked chips was determined in many sides.

As a raw fish material frozen saith (*Pollachius virens*) flesh was used. The production was begin with evaluating the dough by using chopper, stuffing the dough in to case to make it uniform and slicing after particularly frozen than completed after frying or oven baked those chips slices. Modified atmosphere packs were used as a chips package and 100% nitrogen gas was filled. Storage of the packs was done in room conditions. To determine the shelf life of the chips chemical, microbiological and sensory analysis was carried out. At the same time colour and textural changes were also observed. Also to determine the health risk and the benefit, acrylamide levels, fatty acid compositions, atherogenic index and thrombogenic index levels and chemical composition were observed.

Fish chips were produced with just contain mash potatoes, fish fillets, corn starch, spices, salt, sunflower oil and the water. Without any chemical additives product gave us a shelf life more than 75 days up to now and expected shelf life will be more than 90 days. Study is still going on. On the other hand acrylamide levels were determined not more than commercial potato chips products. The difference of the fish contained chips came from its protein, fat and salt contents. Lower values in fat and salt content and high protein content were the differences when compared with commercial potato chips.

Potato chips are mostly preferred and consumed between 18-40 age groups in the World. When we look at the last decade potato chips amount per capita consumption has a real increase in Turkey. Due to the sectoral reports Cereza market size was determined more than 800 million $ after 2012. So integration of fish protein to a highly consumed chips product will be an added value for the consumers' health and for the seafood sector.

Acknowledgment:

*This study was supported by TUBITAK Project No: 114O671. Used data were taken from the saithe production.

*Co-author Arzu Burcu Yavuz taking a PhD scholarship from the programme of Tubitak 2211C.

Key words: fish chips, acrylamide, oven baked, fried, saithe

Comparison of the microbiological, chemical and sensory quality of plaice (*Pleuronectes platessa*) stored in flake ice and slurry ice

Presenting author: Karen Bekaert

Co-authors: Daphné Deloof, Sara Maes, Lisa Devriese, Johan Robbens

ILVO, Oostende, Belgium

Plaice is a very important fish species in Belgian fisheries with yearly landings between 6000 and 9000 tons. Due to the remote fishing grounds, fishing trips from Belgian fishermen can last up to 10 days. Therefore, as fish is a very perishable food product, rapid cooling on board is essential for the shelf life of fish. The application of slurry ice, consisting of small ice crystals dispersed in seawater, is expected to prolong the shelf life of the fish. This was evaluated by storage of plaice in flake ice (FI) and slurry ice (SI). The quality of plaice was analysed by means of sensory, microbiological and chemical methods.

Plaice was caught in the English Channel in March 2015 by the beam trawler Z571 during a single trip. Plaice was divided into two badges of 50 plaice in SI and FI respectively and stored on board in a cool room. Fish was transported to the laboratory and kept in a refrigerator in each type of ice before analyses were carried out. Sensory (Quality Index Method (QIM), microbiological (Total plate count, H2S-producing bacteria and *Pseudomonas* spp.) and chemical analyses (pH, TVB-N and TMA) were performed every 2 or 3 days during 18 days.

Temperature of plaice stored in SI decreased from 5,7 °C to 0,3°C in one hour, while it took more than three hours in FI. Using the QIM, no significant differences were detected between the two storage methods. An increase in all microbiological counts was observed over time in FI and SI, reaching a count of 7,08 and 7,40 log CFU/g respectively at the end of storage for H2S-producing bacteria. TVB-N values reached 28,51 mg/100 g and 34,38 mg/100 g in FI and SI respectively while TMA values reached 15,73 mg/100 g and 22,31 g/100g. Hence microbiological and chemical analyses showed higher values for fish stored in SI than in FI at the end of the storage period, although these differences were not significant. In this experiment, no expected advantages of the use of SI could be demonstrated. Probably, the fast cooling capacity of SI is of limited importance in winter when the temperature of the fish is rather low.

It can be stated that SI is suitable for rapid chilling of plaice on board of commercial trawlers. However, no beneficial effect could be seen on the shelf life of plaice, probably due to low temperature of the fish when caught. Therefore, the experiment could be repeated in summertime.

Key words: shelf life, plaice, slurry ice, flake ice, quality

Simplification of K-value measurement as an index of freshness of fish

Presenting author: Kunihiko Konno

Co-authors: Yimeng Wu, Jiaqi, Zhang, Jili, Shi

Hokkaido University, Hakodate, Hokkaido, Japan

K-value is a widely used index for evaluating the freshness of fish. ATP in fish meat is progressively degraded into much simpler compounds by the endogenous enzymes during its storage and the progress is determined by the temperature and time. Thus, K-value tells the storage history of fish. Originally ATP and its related compound (ARC) were analysed on ion-exchange chromatography, now on HPLC. However, there is no attempt to simplify ARC extraction process. In this presentation, sample preparation was significantly simplified suitable for ARC analysis on HPLC. By using the method, K-value increase for various fish species was studied by changing storage temperature.

Flounder, herring, cod, greenling, and rock cod were the fish used. Their dorsal muscle wrapped with plastic film was stored at 0 and 10°C. ARC was extracted from 1 g meat with 10 ml of chilled 5 % perchloric acid (PCA). 1 M KOH (5.4 ml) was directly added to the mixture to give pH 2.5-3 without removal of denatured protein. Supernatant obtained by standing the mixture in ice was filtered through membrane (0.45μm) and was applied to HPLC (Shodex Asahipak GS-320HQ, 0.1 M phosphate buffer (pH 3)). IMP decomposing activity in fish meat was measured by using fish muscle homogenate as enzyme in reaction medium of 0.1 M NaCl, pH 7.5, 5 mM $MgCl_2$, at 25°C.

The conventional ARC extraction method required removal of denatured protein by centrifugation to obtain ARC in the supernatant, but the step is not necessary. A careful neutralization of PCA with KOH, which is the most difficult step in the original method, was simplified by adding a fixed volume of 1 M KOH to give pH 2.3-3 not 7. The volume of KOH needed is determined by the volume of PCA used. Adjusting pH to 7 rather re-dissolved some proteins, but no protein dissolved at pH 2.5-3. Clear supernatant obtained by standing of the mixture was used for analysis.

IMP accumulation and its slow decrease determined K-value increase. The increase was faster at 10°C than 0°C for all species studied. Cold water fish tend to show quick increase of K-value; Cod and herring showed the fastest increase (90% in 2days at 0°C) while flounder showed much slower increase.

Flounder showed the highest IMP decomposing activity among the species, so K-value increase in muscle contains other factors.

K-value increase was fish species-specific. Cold water fish tend to show quick increase of K-value; a quick loss of freshness.

Key words: ATP, freshness of fish, IMP degradation, K-value, storage temperature

Quality evaluation of fresh, farm raised sea vegetables during refrigerated storage

Presenting author: Denise Skonberg

Co-author: Dhriti Nayyar

School of Food and Agriculture, University of Maine, Orono, Maine, U.S.

Seaweed consumption in western countries has increased over the past decade, due in part to their nutritional benefits, including high levels of dietary fiber, minerals, and antioxidants. Aquaculture contributed approximately 21 million metric tons to the worldwide seaweed harvest in 2011. Only a small portion of the harvest is farmed in the U.S., however various species of seaweeds are currently being tested for their aquaculture potential in the Northeast (Maine). Although seaweeds are traditionally dried, recent interest in consuming more fresh and local foods has created opportunities to market fresh seaweeds, or « sea vegetables ». To develop distribution and marketing strategies for fresh sea vegetables, growers and processors need basic information about their shelf-life and quality loss. The specific objective of this study was to assess the value of chemical, microbial, sensory and physical analyses in determining the shelf life of fresh sea vegetables during refrigerated storage.

Thus far, three species of sea vegetables have been evaluated, including sugar kelp (*Saccharina latissima*), dulse (*Palmaria palmata*), and winged kelp, *Alaria*. Mature plants were harvested by hand, stored at 2ºC or 7ºC for 14 days, and subjected to quality evaluations every 2-3 days. Triplicate lots of sea vegetables were assessed for total plate counts, instrumental color (L*a*b*), texture (texture profile analysis), soluble protein, total volatile base nitrogen, and drip loss. A 12-member sensory panel rated aroma, texture, color, and overall quality of the samples using a 15cm line scale. Data were analysed by ANOVA for species and storage temperature effects, and linear regressions were determined between dependent variables and storage time.

Given the different intrinsic structures, compositions, and textures of the three seaweed species evaluated, the utility of each analysis varied with species. Sensory evaluation and TPA firmness were useful measures across all species, while soluble protein analysis was suitable only for *Alaria*, which exhibited a sharp decline throughout storage. The color of all species changed significantly, with sugar kelp exhibiting a sharp decline in a*, and dulse and *Alaria* a sharp increase in b*, during storage. Total plate counts ranged from 10^2-10^5 across all species, and were highly variable over time, while measurable drip loss occurred toward the end of the acceptable shelf life.

Results emphasize the need to understand quality loss during refrigerated storage of sea vegetables, and to tailor chemical, sensory, and physical analyses to the particular species of interest.

Key words: fresh, farm-raised sea vegetables

Quality improvement in trawling fisheries – CRISP; adaption of capture and handling practices to optimize catch quality and value

Presenting author: Heidi Nilsen

Co-authors: Stein H. Olsen, Torbjørn Tobiassen, Ragnhild A Svalheim and Kjell Ø Midling,

Nofima, Tromsø, Norway

During the last 30 years, the Norwegian trawl industry has carried out considerable structuring to improve profitability. Nevertheless, the technological progress in processing of whitefish on board trawlers has been very low. Target areas within renewal has been on reducing fuel consumption by means of vessel design, developing different catching tools as well as making the output more efficient (by use of well-known technology). However, not much effort has been invested into the improvement of catch handling. Therefore, one objective of the *innovative research program CRISP* is to emphasize the quality of fish, in order to establish and increase the value of trawl-captured fish.

The quality of fish harvested from the sea is influenced by several factors; seasonal variations in feeding, temperature and spawning. The quality is also strongly dependent on how the fish is handled during capture. Few crew members, combined with high capture efficiency, limits the ability to improve the quality. During trawling operations, fish are exposed to a number of stressors, such as swimming to exhaustion, crowding in the cod end, severe barotrauma, and lack of controlled killing and bleeding. Often, the last fish in the storage bin have been dead long before bleeding, and this leads to insufficient exsanguination and muscle discoloration. A swim tunnel has been custom-made, which serves as a trawl simulator, to provide an experimental setup for swimming trials with groups of large fish. This makes it possible to study the cumulative effect of swimming and crowding on the physiology and quality of fish in a controlled environment.

Controlled experiments performed in the swim tunnel as well as trials with commercial trawling and pilot-scale live storage, proves the possibility to perform trawling fisheries in a manner to maintain high quality of the catch. Trials demonstrate fish being exposed to less stress and consequently resulting in fillet qualities whiter in colour and with lesser blood and discolouration. In order to implement these techniques to produce new high-quality products it is essential to thoroughly implement the various mechanisms that govern quality.

Our large-scale trials demonstrate the possibility to conduct trawl fisheries in a manner to promote high quality fish products. Before this can be implemented onboard trawlers today, one should gain further knowledge to understand the fish tolerance, -fatigue, -recovery and -blood flow during catch and handling. However, implementation of techniques and practices may challenge requirements to both efficiency and profitability in trawling fisheries.

Key words: Trawl fisheries, quality, live storage

The role of blood for lipid oxidation and color stability of fish – evidence and prevention

Presenting author: Hanna Harrysson[1]

Co-authors: Michael Axelsson[2], Ingrid Undeland

[1]Chalmers University of Technology, Gothenburg, Sweden

[2]University of Gothenburg, Gothenburg, Sweden

Fish hemoglobins (Hb's) can trigger oxidation of the valuable long chain n-3 polyunsaturated fatty acids (LC n-3 PUFA) through a variety of mechanisms. From this reaction the nutritional value is reduced, rancid odor develops, and color is negatively affected e.g. through redness loss. To date, it is still unclear how significant the role of blood/blood removal is for the quality of fish muscle, in comparison with other known endogenous pro-oxidants like myoglobin and lipoxygenases. The aim of this study was to gain evidence to the full impact of blood for lipid oxidation development and color changes of fish muscle using perfused rainbow trout (*Oncorhynchus Mykiss*) as a model.

Three groups of rainbow trout were used; (i) unbled, (ii) bled by classic gill cut, and (iii) perfused with 0.9% NaCl to completely remove blood. De-skinned fillets from each group were pooled by mincing, fortified with anti-bacterial agent and stored for 3 weeks on ice while following rancid odor, color (L*, a* and b*-values), pH and primary/secondary lipid oxidation products (peroxide value, PV, and thiobarbituric reactive substances test, TBARS).

There were major differences in oxidation development between the three groups; rancid odor appeared after ~4 days in unbled and bled groups, with the intensity being twice as strong in the former. Perfused fish developed no rancid odor during the entire 3 weeks-period. PV/TBARS-data ranked the three sample groups in the same order as the sensory data; but some PV and TBARS development took place also in the perfused group. It was also clear that astaxanthin, the red pigment of rainbow trout, was bleached by co-oxidation in parallel with lipid oxidation.

The present findings give clear evidence to the importance of blood for lipid oxidation development in fish, especially for rancid odour development. Our results thus strongly support the development of novel efficient blood removing strategies for fish that goes beyond classic bleeding. Also efficient antioxidant strategies specifically targeting Hb are required in cases where blood removal is not practically feasible.

Key words: hemoglobin, blood, lipid oxidation, colour, rainbow trout

Determination of quality differences between canned tuna and pouched tuna in different packing media

Presenting author: Asli Cadun

Co-authors: Nida Demirtas, Arzu Burcu Yavuz, Evren Burcu Sen Yılmaz

University of Ege, Fisheries Faculty, Seafood processing department, Izmir, Turkey

Canned tuna is one of the most commonly consumed types of seafood in the world. Tuna in a pouch is a recently-developed product when compared with canning process. According to some researches, tuna in a pouch will replace canned tuna in a few years. As a matter of fact the aim of this study was to determine the differences between two differently packaged tuna with different packing medias.

All the samples were purchased from the local super markets. Seventy two of canned tuna and pouched tuna were purchased in: twelve were canned in olive oil, twelve in sunflower oil, twelve in water and twelve were pouched in olive oil, twelve in sunflower oil, twelve in water. TBARS and pH values, fatty acid composition, heavy metal content (arsenic, cadmium, mercury and lead), texture, colour measurements and sensory evaluation were determined to differentiate the samples and the quality of them.

Results and discussion: According to the results of this study, canned and pouched tuna samples have concentrations well below the permissible FAO/WHO levels for these toxic metals. According to the sensory parameters; overall qualities of the canned tunas were higher than the one in pouched tunas except canned tuna in water. EPA and DHA concentrations in canned tuna in water and pouched tuna in water were higher than the others.

According to fatty acid compositions, sensory and physical analysis some differences were determined between the groups.

Arzu Burcu Yavuz was supported by the program TUBITAK 2211-C for her PhD

Key words: tuna, canning, pouch, quality

Heat processing of pre-rigor Atlantic cod (*Gadus morhua*)

Presenting author: Svein Kristian Stormo

Co-authors: Dagbjørn Skipnes, Izumi Sone, Karsten Heia, Lene Kramer, Torstein Skåra

Nofima, Tromsø, Norway

Fish is very sensitive to thermal processing and will often become tough and dry when exposed to excess heat. Technologies that can ensure extended shelf life and food safety, while maintaining optimal sensory properties, are therefore of great interest. Even if intact fish muscle is believed to be sterile, once the fish is stored the bacteria on the surface will invade the muscle. We have previously shown that heating the surface of 3 days on ice (post-rigor) muscle is not effective to increase shelf life. In the present study we used very fresh material (pre-rigor) to investigate how shelf life is affected when surface bacteria are targeted in minimal heating processing regimes.

Wild caught Atlantic cod (*Gadus morhua*) was vacuum packed and heat treated in a circulating wather bath (80 °C 180s, 90 °C 97s, 90 °C 1100s, and untreated control). The temperatures were selected based on heat transfer modelling (COMSOL Multiphysics ®) that simulated temperature development in the filet during heat treatment at 80 and 90 °C.

Sensory analysis showed little difference between minimally heat treated samples compared to the (untreated) control. Heating at a high temperature for long time (industrial pasteurization) did have an immediate effect on sensory attributes. Microbiological analysis measuring total CFU, H_2S producing bacteria and psychotrophic bacteria reflected sensory results mainly emphasizing the difference between industrially pasteurization and all other treatments.

Estimation of shelf life based on monitoring microbial growth in combination with sensory analysis show that it is challenging to increase shelft life by heat-treating the surface even when the raw material is very fresh (pre-rigor).

Challenges in textural measurements of Atlantic salmon

Presenting author: Gine Ørnholt-Johansson

Co-authors: Flemming, Jessen, Maria Guðjónsdóttir, Stina, Frosch

Division for Food Technology, Research Group for Food Production Engineering, National Food Institute, Technical University of Denmark, Kgs. Lyngby, Denmark.

The yield from processing of Atlantic salmon relies heavily on the textural properties of the meat as the texture affects the mechanical handling during production. Soft fillets and gaping are two of the most common causes of downgrading (Michie, 2001), which in turn reduces the profit of the outgoing products. However, it is not only the direct profit that is affected as the soft texture also can cause down time during the production with additional reduced profit as an outcome. A quick on/at line determination of texture could reduce down time in the production and may help to establish necessary precautions for production in order to avoid downgrading of the products. The aim of this study was thus to identify proteins which can be used for prediction of texture and consequently used for future development of a rapid analysing protein sensor.

38 Atlantic salmon farmed in Norway and processed in Denmark were used for the analysis. Texture was assessed by TA.XT2 Texture Analyzer (Stable Micro Systems, Surrey, England) measurements using a flat-ended cylinder with a diameter of 35mm. Single compression with a penetration depth of 15% of the total fillet height was chosen after testing of several scenarios, as it was this solution, which affected the muscle fibres the least by not breaking them. Two-dimensional gel electrophoresis (2DE) has been made on all muscle samples and the protein response was assessed multivariatly together with the texture.

Texture was measured as peak force and ranged from 0.45 N to 4.84 N. The 2DE gels revealed 433 spots which were found on all gels. The spot volumes, each representing the amount of the individual proteins, were used for further data analysis. By a multivariate approach around 70 proteins have been found to correlate with peak force. Based on these proteins several predictive models of texture could be built and variable selection revealed that only around 20 proteins were needed to predict the texture.

By measurement of only few proteins in the salmon muscle it will be possible to predict the texture.

Reference: Michie, I (2001). Causes of downgrading in the salmon industry. In Kestin SC, Warriss PD, editors. Farmed Fish Quality. Oxford: Blackwell Science. 129–136

Key words: salmon, texture, proteome analysis, modelling

Effect of crust freezing on the shelf-life of salmon (*Salmo salar*) stored at low temperatures under different packaging conditions

Presenting author: Selene Pedrós[1]

Co-authors: José-Antonio Beltrán[2], James-G Lyng[3], Declan Bolton[4], Nigel Brunton[3], Paul Whyte[1]

[1]School of Veterinary Medicine, University College Dublin, Belfield, Dublin 4, Ireland

[2]Department of Animal Production and Food Science and Technology, Faculty of Veterinary, University of Zaragoza, Miguel Servet 177, 50013, Zaragoza, Spain

[3]School of Agriculture and Food Science, University College Dublin, Belfield, Dublin 4, Ireland

[4]Teagasc Food Research Centre, Ashtown, Dublin 15, Ireland

The aim of the present study was to assess the effect of "crust-freezing"(CF) on the shelf-life of fresh salmon fillets, using vacuum skin packaging and modified atmosphere packaging (MAP), and storage temperature of -1.5±1ºC.

Crust-freezing is a method to rapidly chill food products with subsequent storage at temperatures close to freezing point frequently used. Optimal treatment conditions were identified initially by generating freezing curves of temperature reductions in fish samples.

Treatments consisted of introducing samples into a freezing tunnel with an air temperature of -20ºC in order to freeze the surface only (maximum depth < 3 mm). Samples were prepared from whole fresh salmon (72h post-capture) which were cut into fillets and separated into 4 groups: 2 control groups (n=14) packaged in skin and MAP (40% CO_2 – 60% N_2), and two groups (n=14) of fillets treated inside the tunnel and then packaged under the same conditions. All sample groups were stored in a refrigerated cabinet at -1.5±1ºC for 14 days.

Sampling analysis was conducted every 48 h during storage by assessing several physicochemical and microbiological parameters such as: pH, lipid oxidation (TBARs), gas mixture analysis (in MAP), Total Volatile Basic Nitrogen (TVBN) and levels of a range of bacterial groups (Enterobacteriaceae, Total Aerobic Mesophiles, Total Aerobic Psychrophiles, Lactic Acid Bacteria, H_2S producing bacteria and *Pseudomonas* spp.).

CF treatments reduced initial contamination levels of several groups of bacteria below the limit of detection in the case of: *Pseudomonas* spp. (<100UFC/g), H_2S producing bacteria (<10 UFC/g), and Lactic Acid Bacteria (<10 UFC/g). However these groups were detected at subsequent sampling points during the shelf-life and grew normally. Salmon in skin packs had higher levels of bacteria and TVBN compared to equivalent samples under MAP conditions. Crust frozen samples spoiled more rapidly than untreated controls, irrespective of packaging typed used. In general, indicators of spoilage at the end of shelf-life showed that control samples in MAP packaging were most stable followed by CF-MAP, Control-skin and CF-skin respectively. Our study concludes that despite the initial reductions in microflora obtained from CF, this technology resulted in reduced shelf-life of salmon compared to non-crust-frozen samples.

Key words: crust freezing, shelf-life, salmon, skin, MAP

Latest advances for the characterization of seafood in a global market

Keynote speaker: Carmen Sotelo

Investigadora Científica del CSIC, Instituto de Investigaciones Marinas,Vigo (Spain)

Globalisation has brought some advantanges to the seafood industry, such as wider range of different raw materials for the process, transformation and marketing of seafood with a reasonable cost. However, it has also involved different challenges related with raw material characterisation, which usually involve answers to several questions such as identity, origin, presence of contaminants and quality. Time of response, non destructive methods and accurateness are some of the key issues regarding these questions This talk will address some of the recent methodological advances which meet the current demands of industry under the light of these questions.

Development of a qPCR method for the identification and quantification of *Thunnus obesus*, *Thunnus albacares* and *Katsuwonus pelamis* in canned tuna

Presenting author: Daline Bojolly[1, 4]

Co-authors: Véronique Verrez-Bagnis[2], Bruno Le Fur[3], Urania Christaki[4], Thierry Grard[1], Périne Doyen[1]

[1]Université du Littoral Côte d'Opale, Institut Charles VIOLLETTE (EA 7394), unité Biochimie des Produits Aquatiques (BPA), Boulogne sur Mer, France

[2]Laboratoire Ecosystèmes Microbiens et Molécules Marines pour les Biotechnologies (EM[3]B), Ifremer, Nantes, France

[3]Plateforme d'Innovation Nouvelles Vagues, Boulogne sur mer, France

[4]Université du Littoral Côte d'Opale, Laboratoire d'Océanologie et de Géosciences, UMR CNRS LOG 8187, Wimereux, France

Tunas represent the third most fished species worldwide. *The regulation* for *canned tuna labels* mentions that a can must contain only one tuna species. However, this could be difficult to respect due to the high degree of similarity among the tuna species. Indeed, it can be difficult to authenticate the tuna species when the external morphological characteristics are removed due to filleting before canning. Consequently, two specific issues are encountered: i) unintentional substitutions could take place during sorting of tuna on fishing vessels, and ii) voluntary substitutions could occur between species, with different economic values.

Application of heat treatments during industrial processes, such as cooking and sterilization, induces protein degradation and DNA fragmentation. Therefore, very short sequences of genes are more adapted to identify tuna in canned products by DNA-based methods. Consequently, in this study short mitochondrial gene fragments were used to authenticate the three most consumed species in France: *Thunnus albacares* (yellowfin tuna), *Thunnus obesus* (bigeye tuna) and *Katsuwonus pelamis* (skipjack tuna).

Specific TaqMan probes have been designed for yellowfin, bigeye and skipjack tuna species identification by qPCR.

Quantification method was investigated on canned products specifically processed with different tuna species percentages.

Authentication of *Thunnus albacares*, *Thunnus obesus* and *Katsuwonus pelamis* was performed by TaqMan qPCR. The three species have been successfully identified in canned products. The first results on tuna species quantification in canned products are promising.

The development of this TaqMan real-time PCR method can provide a rapid tool to protect consumers and fish industries from frauds and to contribute to control compliance with existing legislation. The further step of this study is to prepare the implementation of this method to an industrial analysis laboratory.

Key words: tuna, authentication, quantification, canned products, TaqMan

Species identification in samples containing fish mixtures : a targeted next-generation sequencing approach

Presenting author: Kristina Kappel

Co-author: Ute Schröder

Max Rubner-Institut, Department of Safety and Quality of Milk and Fish Products, Hamburg, Germany

Authentication of fish species in fish products is mainly achieved with conventional Sanger sequencing of appropriate gene fragments (e.g. Cytochrome b (cytb) gene or Cytochrome Oxidase I gene). However, this approach is limited to products consisting of only one species as mixtures lead to overlapping chromatograms. Next-Generation Sequencing (NGS) technology offers the advantage of simultaneous assessment of multiple sequences generated from complex samples. In order to explore the suitability of this technology for fish product analysis, a targeted NGS approach was developed and tested with samples containing mixtures of different fish species.

Nine mixtures with varying tissue percentages from five Scombridae species were prepared and subsequently analysed – together with four tuna can samples – with a cytochrome b gene targeted NGS method on an Illumina MiSeq (V3) platform.

Sequence recovery for tuna mixtures was very accurate and duplicate samples as well as two individual NGS runs gave very similar results in terms of sequence read values. Detection of false-negative and false-positive sequences was very low when applying an appropriate threshold and species constituting down to 1 % of mixture could be identified, depending on the species composition. Indeed, the species composition had a great influence on sequence read percentages with particular species being overrepresented compared to others. NGS analysis of market samples indicated the presence of different species mixed in tuna cans, although this does not comply with European legal requirements.

These results show that NGS represents a powerful technology for species identification in food products with mixed species' contents and can become an important tool for the authentication of tinned fish goods in the future.

Key words: fish species, authentication, mixtures, Next-Generation Sequencing, Cytochrome b

Development of a qPCR method targeting *torA* gene and application for the freshness monitoring of modified atmosphere-packed chilled whiting (*Merlangius merlangus*)

Presenting author: Alexandre Dehaut[1]

Co-authors: Frédéric Krzewinski[2], Thierry Grard[3], Marlène Chollet[4], Philippe Jacques[4], Anne Brisabois[1], Guillaume Duflos[1]

[1]Anses – LSAl – Département des Produits de la Pêche et de l'Aquaculture, Boulevard du Bassin Napoléon, 62200 Boulogne-sur-Mer, France

[2]Univ.Lille – CNRS, UMR 8576 – UGSF, Unité de Glycobiologie Structurale et Fonctionnelle, 59000 Lille, France

[3]Université du Littoral Côte d'Opale – Anses, Unité Biochimie des Produits Aquatiques, Boulevard du Bassin Napoléon, 62200 Boulogne-sur-mer, France

[4]Univ.Lille – Laboratoire Régional de Recherche en Agroalimentaire et Biotechnologie – Institut Charles Viollette – Equipe ProBioGEM, Avenue Paul Langevin, 59655 Villeneuve d'Ascq, France

Tracking the early decline of freshness is an important concern for the fishery industry to insure the best quality for foodstuffs highly liable to spoil. A lot of techniques have been developed like sensory, chemical and more slightly microbial analysis. Unfortunately, most of them have drawbacks of being subjective, or having the disadvantage of being either reliable once the freshness is lost or for the analysis of whole fish. The study presents the development of a qPCR method targeting a gene harboured by specific spoilage organisms (SSOs) of fish: *torA*. This gene encodes an enzyme responsible of the production of trimethylamine whose odour is characteristic of the spoiled fish.

The study aimed to develop a degenerate primer pair able to amplify *torA* gene in a wide range of SSOs. For that, numerous software and algorithms were used for a maximal reliability of the *in silico* design process. A first selection of pairs where tested *in vitro* to further characterization. Finally a primer pair was conserved for efficiency and selectivity tests. The selected primer pair was tested for analysing MAP-chilled whiting along a 15 days storage study. Methods such as total volatile basic nitrogen (TVB-N) and trimethylamine (TMA) analysis or total viable count (TVC) analysis were performed simultaneously to evaluate fish overall quality.

A degenerate primer pair was selected after six steps of *in silico* design and selection. It amplified *torA* gene of both *Vibrio* and *Photobacterium* with good efficiencies, ranging from 91.6 to 93 % regarding species, on 7-log DNA dilutions. The best conditions of annealing temperature and primer concentration were found to be 62°C and 1 µM. As regards with selectivity, the degenerate primer pair allows to selectively amplify *torA* gene of *Vibrio* and *Photobacterium* compared with other tested species. TVC study led to inconclusive results, probably because ISO standard used was not suitable for these kinds of food and storage. However, throughout the storage of fillets, the qPCR approach allows detecting an increase of *torA* copies. Additionally, good correlation between qPCR results and the evolution of known spoilage markers were established, such as -0.86 (TVB-N) and -0.81 (TMA).

This study, thanks to careful steps of primers characterization, allowed designing a primer pair able to amplify *torA* gene of both *Vibrio* and *Photobacterium*. A very promising, sensitive and time-effective qPCR technique is thus proposed to characterize the freshness of processed whiting.

Key words: qPCR method, *torA*, whiting, MAP fish, freshness

The application of near infrared spectroscopy to study physicochemical properties and quality degradation of seafood

Presenting author: Magnea Karlsdottir[1]

Co-authors: Sigurjón Arason[1,2], Hordur Kristinsson[1,3], Kolbrun Sveinsdottir[1]

[1]Matis ohf, Icelandic Food and Biotech R&D, Vinlandsleid 12, IS-113 Reykjavík, Iceland

[2]University of Iceland, Department of Food Science, Vinlandsleid 12, IS-113 Reykjavík, Iceland

[3]Department of Food Science and Human Nutrition, University of Florida, 359 FSHN Building, Newell Drive, Gainesville, FL 32611, USA

Nowadays, near infrared spectroscopy (NIR) has become the alternative quality control method in the food industry due to its advantages over other traditional analytical methods. It is fast and non-destructive method and requires little or no sample preparation. One of the strength of NIR is that it can provide simultaneous determination of multiple components per measurement with a remote sampling capability and hence, it can provide real-time information from processing lines.

NIR was applied to estimate physicochemical characteristics and quality degradation of cod liver and fish oil. Calibration models were developed, using partial least squares (PLS) regression, for total lipid content and composition, lipid hydrolysis (free fatty acids), primary and secondary oxidation products, as well as sensory evaluation. Coefficients of determination for calibration (R^2_{cv}) and root-mean-square error of cross validation (RMSECV) ranged from 0.82 to 0.99. The validation of the calibrations indicated that physicochemical properties and lipid degradation of cod liver and fish oil can be estimated by NIR with good accuracy. Overall, the results demonstrate the potential for use of NIR spectroscopy as an objective and non-destructive method to inspect the lipid characteristics and quality of seafood products.

Key words: NIR spectroscopy; seafood; quality; lipid degradation; physicochemical properties

Use of fluorescence spectroscopy for monitoring whiting (*Merlangius merlangus*) fillets freshness stored under various conditions

Presenting author: Abdo Hassoun

Co-author: Romdhane Karoui

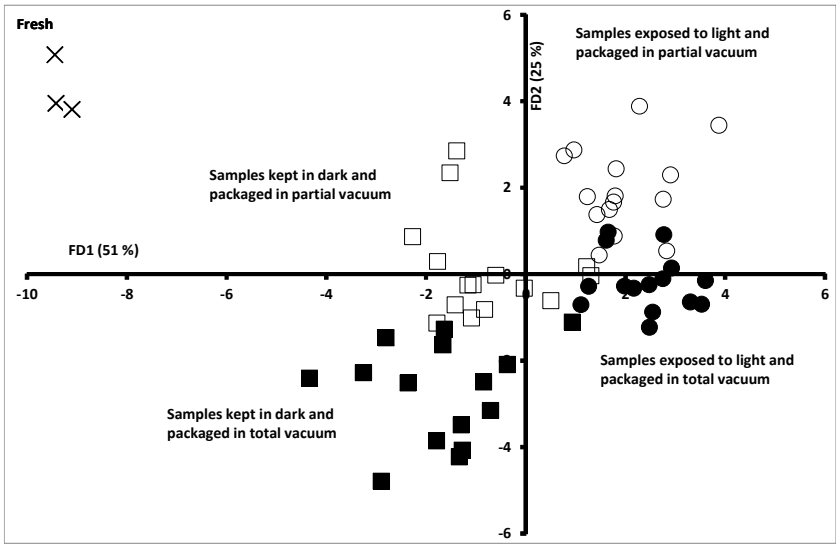
Univ. Artois, EA 7394, Institut Charles Viollette, Lens, F-62307, France

Fish is a highly perishable product, so adequate preservation techniques must be used to maintain the desired levels of quality and safety throughout storage. Storage of fish at low temperature and/or by using suitable packaging prolongs its shelf-life. The objective of the present study was to evaluate the potential of front-face fluorescence spectroscopy (FFFS) for monitoring whiting fillets freshness during 12 days of storage at 4 °C in the presence/absence of light and total/partial vacuum packaging.

21 whiting fish samples were taken at Boulogne Sur-Mer harbor just after unloading of trawler. The fishes were filleted and brought to the laboratory within a 2 h in iced condition. One sample was analyzed on day 1 and considered as fresh. The remaining fish samples were divided randomly to four groups (5 samples/group) in the: i) dark associated with total/partial vacuum packaging; and ii) light associated with total/partial vacuum packaging and analyzed on days 3, 5, 8, 10, and 12 of storage. Physico-chemical parameters and tryptophan and NADH (excitation: 290 and 340 nm, respectively) were acquired in triplicate.

Throughout storage, a general decrease of water content and an increase of TBARS and pH values were observed regardless of storage conditions. Fish samples revealed maximum peroxide values after 3 days storage.

Tryptophan emission spectra showed 2 maxima located at 371 and 336 nm. A red shift was observed for aged samples indicating changes in the molecular environment of tryptophan residues. The NADH spectra exhibited two maxima located at 395 and 460 nm. A linear correlation (R^2=0.7) was observed between TBARS and 460/395 ratio fluorescence intensity for samples exposed to light.



Factorial discriminant analysis with full cross-validation was applied to all the data sets. The similarity map defined by the discriminant factors 1 and 2 showed that all fish groups were mostly separated (**Figure 1**). Overall correct classification rate of 90.48% was obtained.

Figure 1: Factorial discriminant analysis similarity map of the leave one-out cross-validation determined by discriminant factors 1 and 2.

Fluorescence spectra showed some differences between whiting fillets according to their storage conditions. A high correlation between TBARS and NADH spectra was observed allowing the potential use of NADH as an indicator of fish level oxidation, particularly for samples exposed to light. Thus, it would be interesting to test the ability of NADH spectra on semi-fatty and fatty species. The FFFS could be used as a screening tool for determining fish freshness.

Key words: Whiting, freshness, oxidation, fluorescence, chemometric

Detection of histamine in fish by surface enhanced Raman spectroscopy using solid silver SERS substrates

Presenting author: Tibor Jančí[1]

Co-authors: Mile Ivanda[2], Lara Mikac[2], Nives Marušić[1], Helga Medić[1], Sanja Vidaček[1]

[1]Faculty of FoodTechnology and Biotechnology, University of Zagreb, Zagreb, Croatia

[2]Ruđer Bošković Institute, Zagreb, Croatia

Histamine fish poisoning is common problem that occurs worldwide and is probably the most frequent form of intoxication caused by consumption of seafood. Although large number of methods for laboratory histamine testing and laboratory screening of fish and related products has been developed over time they are often time and labor consuming and require expensive equipment and skilled personnel to conduct analysis. Consequently, there is still need for more simplified, fast methods suitable for short time on-field histamine testing of fish and related products.

Surface Enhanced Raman spectroscopy offers possibility of short-time analysis of different analytes at low concentration with minimal sample preparation. In our previous study, histamine has been detected using silver colloid SERS substrates in fish muscle extracts at concentration of $1x10^{-4}$ M which conforms to 111 mg/kg of fish. However, achieved level of detection is slightly higher than legislative set limits for histamine and results showed significant variations due to large number of factors influencing acquisition of SERS spectra, such as aggregation of silver nanoparticles and adsorption of histamine and other molecules present in extract on the aggregated nanoparticles.

In order to minimize variations of results and lower the limit of detection, Ag "film over nanospheres" (FON) solid SERS substrates which are known to have very uniform SERS enhancement of $>10^7$, were examined in this study.

Ag FON substrates were prepared by drop-coating of polystyrene nanospheres on a clean glass microscope coverslips which after drying formed close-packed array of nanospheres on the surface of coverslip. Ag was thermally evaporated in vacuum and deposited atop nanosphere arrays to form SERS active surface. Several size of polystyrene nanospheres and Ag film thickness were examined in order to achieve optimal histamine SERS signal enhancement.

Results obtained with different Ag FON substrates in histamine water solutions and histamine spiked fish muscle extracts will be presented.

Key words: histamine, Surface Enhanced Raman Spectroscopy, quality

Toxicity of the Lessepsian pufferfish *Lagocephalus sceleratus* from Turkish Mediterranean coast and the species authentification by rapid PCR amplification method

Presenting author: Caner Acar

Co-authors: Shoichiro Ishizaki*, Yuji Nagashima

Graduate School of Marine Science and Technology, Tokyo University of Marine Science and Technology, Minato, Tokyo, Japan

* Corresponding author, ishizak@kaiyodai.ac.jp

After opening Suez Canal in 1869, more than 300 marine species have migrated from Red Sea to Mediterranean system, termed as 'Lessepsian migration'. Several species of pufferfish are also found and adapted to Mediterranean biota. Since the first confirmed record of *Lagocephalus sceleratus* on 2005, the species has been collected from all around of eastern Mediterranean and sometimes caused food poisoning due to tetrodotoxin (TTX), one of the most potent neurotoxin. Therefore, Lessepsian pufferfish has been increasingly concerned as food hygiene and safety issues. In this study, then, we examined the toxicity of *L. sceleratus* and developed PCR-based rapid identification method.

Thirteen specimens were collected from Marmaris and Iskenderun Bay, Turkey. TTX was extracted from several tissues of the specimens with 0.1 % acetic acid and determined by LC-ESI-MS/MS. Total genomic DNA was extracted from the muscle tissue by a Quick Gene DNA Tissue Kit. In order to determine the species-specific regions existing in complete mitochondrial DNA (mtDNA) of *L. sceleratus*, the nucleotide sequence was aligned with other seven *Lagocephalus* species. Based on the result of the alignment, species-specific primer pair for the authentification of *L. sceleratus* was constructed.

All of specimens examined in this study contained TTX, although TTX contents varied among the individuals and tissues. One male specimen (body weight 1380 g, body length 48.0 cm) showed high toxicity. The intestine was the most toxic as high as 48.8 µg TTX/g, followed by the kidney (34.0 µg TTX/g), and the liver (25.4 µg TTX/g), estimated as 'strongly toxic levels'. It is notable that the flesh and testis had detectable amounts of TTX at 3.4 µg TTX/g and 2.6 µg TTX/g, respectively.

The alignment of mtDNA sequences among *Lagocephalus* species showed that the region of NADH dehydrogenase subunit 2 (ND2) was suitable for the designing of species-specific primer pair, which can distinguish *L. sceleratus* from other *Lagocephalus* species. Although other twenty puffer fish species were examined with the primer pair, positive results were only obtained from *L. sceleratus*. Method sensitivity was also determined by using various concentrations of template DNA (0.1 - 25 ng/µl), and the detection limit was as low as 0.1 ng/µl. Thus, the method was not only highly selective, but also very senstive. In conclusion, we confirmed the toxicity of *L. sceleratus* from Turkish waters and established a quick identification method for detection of potential contamination or accidental consumption.

Key words: pufferfish, *Lagocephalus sceleratus*, toxicity, PCR, Mediterranean

Characterization of *Shewanella baltica* strains with usual and atypical H_2S productions, isolated from a spoiled whiting (*Merlangius merlangus*)

Presenting author: Alexandre Dehaut[1]

Co-authors: Graziella Midelet-Bourdin[1], Jean-Sébastien Py[2], Benoît Gassilloud[2], Anne Brisabois[1], Guillaume Duflos[1]

[1]Anses – LSAl – Département des Produits de la Pêche et de l'Aquaculture, Boulevard du Bassin Napoléon, 62200 Boulogne-sur-Mer, France

[2]Anses – Laboratoire d'Hydrologie de Nancy, 40 rue Lionnois, 54000 Nancy, France

Shewanella putrefaciens has been described in numerous studies as the specific spoilage organism of marine fish conserved on melting ice. *S. putrefaciens* is more likely a bacterial group divided into four sub-groups than a single species; subgroup II being only composed by *Shewanella baltica*. In most cases, isolation of *Shewanella* strains involves the ability to produce hydrogen sulfur while growing on iron agar medium. Besides, among *Shewanella*, *S. baltica* has been described as the most H_2S producing bacteria during the storage of marine fish.

Four strains of *Shewanellaceae* were isolated from a spoiled whiting (*Merlangius merlangus*) using a phenotypic criterion (purple pigment production on TSA-YE medium). All the strains were subjected to growth ability, biochemical, molecular and proteomic analyses. Growth tests were performed in aerobic condition on TSA-YE NaCl medium from 4°C to 37°C. A test was also carried out anaerobically at 25°C on TSA-YE NaCl medium with 40 mM trimethylamine-N-oxide (TMAO). Biochemical patterns were observed on API 20E system incubated at 30 °C for 48H. Molecular analysis involved partial sequencing of both 16S rRNA and *gyrB* genes. MALDI-TOF analyses were carried out on proteins extracts with a Brüker LT Microflex device.

All strains were able to grow aerobically from 4°C to 30°C. Interestingly, they were also able to grow in anaerobic condition at 25°C with TMAO, the characteristic odor of trimethylamine (fishy off-odor) being ascertained. Biochemical characterization did not allow accurately identifying the four strains. This study allows emphasizing that a strain was unable to produce hydrogen sulfur. Two isolates synthetized an ornithine decarboxylase, making them potential producers of putrescine. Concerning the use of carbon source, different patterns were recorded among the strains. Genotypic characterization allowed identifying the strains thanks to both 16Sr RNA and *gyrB* sequencing. The four isolates were identified as member of *Shewanella baltica* species. Comparing the two sequencing approaches, *gyrB* sequences allowed a better discrimination of the strains. Proteomic characterization of the strains confirmed that the isolates belong to *S. baltica* species. MALDI-TOF study allows pointing out some slight differences between the proteome of the strains.
This study allows isolating a H_2S negative *S. baltica* from a spoiled fish. New studies will be necessary to estimate the prevalence of such a strain and its spoilage abilities. Thus H_2S production might not be a suitable screening parameter both to count *Shewanella* or to study fish spoilage flora.

key words: *Shewanella baltica*, H_2S, biochemical typing, genotyping, MALDI-TOF

Highly active phosphatase is responsible for rapid loss of IMP nucleotide in cod muscles

Presenting author: Larissa Balakireva

Co-author: Maria Hamdani

NovoCIB SAS, 115 avenue Lacassagne, 69003 Lyon, France

IMP nucleotide (natural flavour enhancer E630-E633), is an intermediate product of post mortem ATP degradation, responsible for umami taste typical of only fresh fish. This nucleotide is determinant for sensorial quality of seafood products and its concentration in fish muscle depends on two enzymatic pathways :

1) IMP synthesis from AMP, catalyzed by AMP-deaminase (AMP-DA). In salmon, the expression of AMP-DA 3 isoform was found in positive correlation with salmon firmness (*Larsson T* et al, 2012). In pork meat, this enzyme was also related to the duration of onset phase of rigor mortis and the texture (E.M. England *et al*, 2015).
2) The rate of IMP hydrolysis to inosine catalysed by phosphatase.

The aim of this study was to develop a method for measuring enzymatic activities responsible for IMP synthesis and IMP degradation in fish muscle. The method was further used to characterize tissue-specific activity of AMP-deaminase and IMP- phosphatase activity in cod and salmon muscles.

The nucleotides were measured in dorsal muscles of fish (cod and salmon) using a "PRECICE® Freshness Assay Kit". In this assay, IMP, inosine and hypoxanthine, are enzymatically converted to NADH and quantified by measuring the absorbance at 340nm using microplate reader (EPOCH, BioTek). The results were expressed in gram per kg and Ki-value. Phosphatase and AMP deaminase activity were measured as the rate of inosine and IMP formation from IMP and AMP substrates added to minced muscles, respectively.

Mean IMP concentrations in cod muscle after 3 days conservation on ice was found to be 0.033g of IMP per kg (K value 81%) which is ~24 times inferior compared to IMP concentrations found in similar salmon muscles - 0.80 grams per kg (K value 54%).

The comparison of phosphatase activity in cod and salmon muscles has revealed that IMP hydrolysis in cod muscle is very fast compared to the salmon. At 25°C cod muscle phosphatase is capable of hydrolysing 3.3g+/-0.5g of IMP per kg per hour. This process is 10-times faster than that in salmon (0.194g of IMP per kg per hour). In contrast, the activity of AMP-deaminase in cod was 1.7-times slower than in salmon.

Our results show that highly active phosphatase can be responsible for fast degradation of IMP in cod muscle. Since this enzymatic process is inhibited at low temperature, fast freezing of cod may result in higher content of IMP in cod muscle and better sensorial quality. In fact, the analysis of on-board frozen cod has shown that IMP concentration was as high as 1.011g of IMP per kg (K-value 27.3%).

Key words: nucleotides, IMP, K-value, cod, salmon

POSTER ABSTRACTS

Enhanced functional properties and antioxidant activity of rainbow trout *(Oncorhynchus mykiss)* by-product hydrolysates derived from microwave-assisted hydrolysis

Presenting author: Elizabeth Nguyen[1]

Co-authors: Owen Jones[1], Brad Kim[2], Fernanda San Martin-Gonzalez[1], and Andera Liceaga[1]

[1]Department of Food Science, Purdue University, West Lafayette, IN 47907, United States

[2]Department of Animal Sciences, Purdue University, West Lafayette, IN 47907, United States

Fish protein hydrolysates (FPH) have been widely used for better utilization of fishery by-products through the use of proteolytic enzymes to produce a wide range of functional peptides that can be used as food ingredients. With the use of microwave-assisted hydrolysis, rapid heating would be ideal for FPH production compared to slower conventional heating (CH) methods. The objective of this research was to investigate the effect of microwave (MW) heating during enzymatic hydrolysis on functionality and antioxidant activity of FPH.

Treatments consisted of adding Alcalase™ to trout by-products (frames only) at 0.02%, 1.75%, 3.52%, and 3.00%, enzyme substrate ratio (E/S), respectively. Treatments were hydrolyzed for 3.33, 10, 15, and 17.07 min reaction time using a microwave system (FISO Technologies Inc., Quebec, Canada) at 1200W, 20% power with 50% duty cycle and a CH method (water bath at 50°C), respectively. Degree of hydrolysis, protein solubility, emulsifying activity index (EAI), emulsion stability, foam capacity, foam stability and DPPH radical scavenging activity were evaluated. ANOVA with Tukey's pairwise comparison of means ($\alpha = 0.05$) was used to determine statistical significance of observed difference among means.

MW-FPH resulted in higher degree of hydrolysis compared to CH-FPH for all reaction time. Protein solubility was higher ($p < 0.05$) in MW-FPH compared to CH-FPH at pH 7, however no differences were observed between at pH 3 and 9. EAI was higher ($p < 0.05$) in MW-FPH compared to CH-FPH for 15 min (3% E/S) and 3.33 min (1.75% E/S). Emulsion stability had no differences among E/S treatments of MW and CH FPH. Foaming capacity was highest ($p < 0.05$) for 3.33 min (1.75% E/S) MW-FPH whereas foam stability showed no differences between MW and CH FPH. MW-FPH exhibited higher ($p < 0.05$) DPPH radical-scavenging activity compared to CH-FPH indicating better antioxidant property. Rapid heating of MW produced FPH with higher degree of hydrolysis, solubility and EAI, foaming capacity and antioxidant property.

The use of microwave heating is a rapid alternative to produce FPH with improved functionality and antioxidant activity. By-product hydrolysates derived from microwave-assisted hydrolysis show great potential as value-added food ingredients.

Key words: Fish Protein Hydrolysates (FPH), Microwave-assisted, Rainbow Trout, Functional Properties, Antioxidant Activity

Antioxidant properties of protein hydrolysates from marine by-products

Presenting author: Şükran Çakli

Co-authors: Arzu Burcu Yavuz, Nida Demirtas Erol, Ömer Alper Erdem

Ege University,Faculty of Fishery, Department of Fish Processing Technology, Izmir-Turkey

Large amounts of protein by-products from seafood processing industry are discarded without any attempt of recovery and valorization. The hydrolysis of marine by-products derived from seafood processing industries is a valuable process to increase the nutritional and bioactive performances of these protein sources. The demand for the sustainable use of marine by-product protein hydrolysate has led to the development of processes for the recovery and hydrolysis of proteins, the assessment of their functionalities, and application into different products. Protein hydrolysates are proteins which are chemically or enzymatically hydrolyzed into peptides into varying sizes. Several proteolytic enzymes are available from microbial, plant or animal sources and these are used for hydrolysing shellfish and fish proteins. Oxidation is an essential process in all living organisms and the formation of free radicals and other reactive oxygen speciesis unavoidable during the oxidative metabolic process. An antioxidant is defined as any substance that significantly delays or inhibits oxidation of a substrate when present at lower concentrations compared to that of an oxidizable substrate. Antioxidants are increasingly used as a means of enhancing shelf-life and to improve the stability of lipid and lipid-containing foods. Antioxidants from marine resources have attracted the attention of researchers as they are extracted from by-products of marine processing and do not have side effects. Under controlled enzymatic hydrolysis condition, the functional amino acid sequence could be released from the proteins, and high antioxidant activity from hydrolysates and peptides had been observed in the protein hydrolysates of yellowfin sole frame, Alaska pollack frame, mackerel muscle, Sphyrna lewini muscle, conger eel muscle, grass carp muscle, blue mussel muscle, lanternfish, monkfish, oyster, mussel and cod. These food-derived antioxidants are considered to be safer and without the side effects associated with the synthetic antioxidants. The antioxidant protein hydrolysates derived from various fish proteins have potential for nutritional, pharmaceutical, cosmetic and nutraceutical applications as a functional ingredient due to their health promoting effects.

Results of this study was supported by TUBITAK 112O925 (1002) project.

Arzu Burcu Yavuz as a member of working team makes use of PhD scholarship program TUBITAK 2211-C for the priority fields.

Key words: Fish protein hydrolysate, marine by-products, antioxidant properties

Proximate, fatty acid and amino acid composition of by products of the cultured rainbow trout (*Oncorhynchus mykiss*), sea bream (*Sparus aurata*) and sea bass (*Dicentrarchus labrax*)

Presenting author**:** Şükran Çakli

Co-authors: Ömer Alper Erdem, Nida Demirtas Erol

Ege University, Faculty of Fishery, Department of Fish Processing Technology, Izmir-TURKEY

In this study, proximate composition (protein, lipid, ash and moisture), fatty acid and aminoacid composition of by products (head, viscera and frame) of trout, sea bream and sea bass were investigated. After the results evaluated, production of value added product was aimed from these by products.

In this study, head, viscera and frame of the trout, sea bass and sea bream were used as raw materials. Proximate composition, fatty acid and aminoacid composition were investigated in these by-products. Differences between means were analyzed by one-way analysis of variance (ANOVA) followed by Tukey and Duncan tests. The results were presented as means ± S.D.

According to results the highest protein content respectively were investigated in head of sea bream (16,66±0,04%), frame of trout (16,31±0,0%) and head of trout (14,87±0,09%). The highest lipids were respectively investigated in visceral organs (sea bass 67,62±7,56 %, sea bream 44,69±1,89 % trout 34,85±2,07 %).

The highest EPA+DHA ratios were investigated in heads of sea bass (10,27272%) and sea bream (7,98222%). High EPA+DHA also were investigated in all other by-products. Also all by-products were important source of important essential aminoacids.

Results showed that by-products substantially contain lipid, protein, essential amonacids and fatty acids. The highest profitability are expected from bioactive components. These bioactive components can be obtained and purified by different technologies from simple to complex. These components can contain isolation of bioactive peptides, oligpsaccarides, fatty acids, enzymes, minerals and biopolymers for biotechnological and pharmaceutical application.

This study was supported by TÜBİTAK- TEYDEP 5135018 project.

Key words**:** Sea bream, Sea bass, Trout, Amino acid composition, Fatty acid composition

Reduction of sodium content of European anchovy (*Engraulis encrasicolus*) marinade with replacement of NaCl by KCl

Presenting author: Nida Demirtaş Erol

Co-authors: Ömer Alper Erdem, Arzu Burcu Yavuz, Şükran Çakli

Ege University, Faculty of Fishery, Department of Fish Processing Technology, Izmir-Turkey

Anchovy is important and most abundant fish species in Turkey. Anchovy is found in Black Sea. Different products can be made from anchovy. Marinade is one of them. Marinated products are generally eaten as appetizer. These products are made with salt and acid and it contains high amount of salt in other word sodium. High consumption of salty product can be harmful for human health. So that, many countries make an effort to reduce salt (sodium) consumption in foods. For this purpose, different substances such as KCl, CaCl, MgCl etc. can be used to reduce sodium content.

In this study european anchovy (*Engraulis encrasicolus*) was used as raw material. Anchovies were supplied from a local market as filleted. Filleted achovies were transported to Ege University, Faculty of Fisheries, Chemistry Lab in ice. In the same day, anchovies were put into process. For marinade production, food grade acetic acid, NaCl and KCl were used. Marinated anchovies were packed in plastic box with sunflower oil. Study was consist of five groups (**H0:** 100% NaCl; 0% KCl, **H1:** 75% NaCl; 25% KCl, **H2:** 50% NaCl; 50% KCl, **H4:** 25% NaCl; 75% KCl, **H5:** 0% NaCl; 100% KCl). In all groups, chemical and microbiological quality, proximate composition, sodium and potassium content, acid content in fish meat, acid and salt content in brine, sensory evaluation, TPA, cooking loss and color were determined.

In this study different replacement ratio of KCl were used to evaluate for reduction sodium content. 100% KCl showed that intense bitter taste and disintegrated texture occurred in product. Best results for sensory, texture and some other physical parameters were investigated in group H2 (50% NaCl; 50% KCl).

As a conclusion, this study showed that sodium content in ancovy marinade can be reduced by the replacemet of NaCl with KCl at 50% ratio.

- This study was supported by Ege University 13-BIL-032 project.
- Arzu Burcu Yavuz as a member of working team makes use of PhD scholarship program TUBITAK 2211-C for the priority fields.

Key words: anchovy marinade, KCl, NaCl, replacement, reduction

Recovery of nutritional protein and lipid fractions from Nordic shrimp byproducts

Presenting author: Serge Laplante

Co-authors: Piotr Bryl, Marie-Elise Carbonneau, Marie-Gil Fortin

MERINOV-UQAR, Gaspé (Québec) Canada

The Nordic shrimp processing industry in Quebec (Canada) generates about 11,000 tons of byproducts annually. Those byproducts could be processed to get a minced shrimp, which could find application as an animal feed. However, through the enzymatic hydrolysis of minced shrimp, higher value fractions for human or animal nutritional ingredient markets were produced. Among them, an hydrolysed protein isolate and a protein-rich solid fraction were obtained. The latter was defatted using supercritical CO_2 extraction to get a phospholipid and carotenoid-rich oil. From the byproduct, a yield of 13% minced shrimp was obtained. From the minced shrimp, 4% of hydrolysed protein isolate and 25% of solid fraction were obtained. The total lipid content in minced shrimp and solid fraction were 2% and 7.5%, respectively. Results present the flowchart of the process, including the yields and proximal composition of the obtained fractions. The niches of commercial applications are also discussed.

Key words: Nordic shrimp, byproducts, valorisation, fractionation, nutrition

Enhancement of ACE- and prolyl oligopeptidase-inhibitory activity of hydrolysates from sardine and tuna by-products by simulated gastrointestinal digestion

Presenting author: Oscar Martínez Alvarez[1]

Co-authors: Irineu Batista [2], Cristina Ramos[2], Pilar Montero [1]

[1]Institute of Food Science, Technology and Nutrition, Madrid, Spain

[2]Instituto de Investigação das Pescas e do Mar, Av. Brasília, P-1400 Lisbon, Portugal

Hypertension is a significant health problem worldwide. The treatment of hypertension is mainly based on the use of synthetic Angiotensin-Converting Enzyme (ACE)-inhibitors, but their consumption is often associated with undesirable side effects. Furthermore, Prolyl Oligopeptidase (PO) is involved in the maturation and degradation of peptide hormones and neuropeptides. Altered PO activity has been associated with autism spectrum disorders and various psychological diseases such as Alzheimer's disease or schizophrenia. Several PO-inhibitors are known and have been suggested as possible nootropic and antidepressant drugs. Tuna and sardine are industrially processed generating huge amounts of waste that could be an interesting source of PO- or ACE-inhibiting molecules. In this work, the ACE- and PO-inhibitory abilities of three protein hydrolysates from industrial by-products of tuna or sardine were evaluated, before and after simulated gastro-intestinal digestion (GID).

Heads of sardine, heads of tuna and a mixture of muscle debris and viscera of tuna were hydrolyzed with Alcalase™. The molecular weight profile of each hydrolysate (FPH) before and after GID was determined by SEC-HPLC at 214 nm. ACE- and PO-inhibiting activities were evaluated by using Hippuryl-His-Leu and Z-Gly-Pro-pNA as substrate, respectively. The Hippuric Acid and nitroanilide liberated were quantified by HPLC. Simulated gastrointestinal digestion (GID) of FPHs was carried out by using pepsin (pH 2, 75 min.) and pancreatin (pH 6.5, 90 min.) at 37ºC.

All hydrolysates exerted moderate ACE and PO inhibition. The tuna heads hydrolysate exerted the maximum inhibitory activity. The inhibiting potency of FPHs against ACE and PO augmented during digestion with pepsin, and the best results were obtained with the digested hydrolysate of tuna heads. Interestingly, the inhibitory capacities of the FPHs slightly decreased or remained stable during subsequent pancreatin digestion. The enhancement of both PO- and ACE-inhibiting abilities of the hydrolysates during pepsin digestion coincided with the release of small peptides that were stable against further pancreatin digestion.

ACE- and PO-inhibiting hydrolysates can be obtained from industrial sardine and tuna by-products. Simulated GID (mainly that of tuna heads) increases the PO- and ACE-inhibiting potency of the hydrolysates, especially by digestion with pepsin. Tuna and sardine hydrolysates can therefore be additives with potential interest in functional foods and represent a healthy alternative to diminish the incidence of hypertension and neurological diseases.

Key words**:** ACE, Prolyl oligopeptidase, hydrolysates, gastrointestinal digestion, inhibition

Effect of barley on the antioxidant properties of rainbow trout fillets throughout the inclusion in the diet

Presenting author: Julia Pinedo Gil[1]

Co-authors: Ana María Larrán García[1], Daniel Rico[1], Miguel Jover Cerdá[2], Ana Tomás Vidal[2], Miguel Sanz Calvo[1], Ana Belén Martín-Diana[1]

[1] Subdirection of Research and Technology. Agro-Technological Institute of Castilla y León. Consejería de Agricultura y Ganadería. Finca de Zamadueñas. Ctra. Burgos km. 119, 47171, Valladolid, Spain

[2] Aquatic Resources Research Group, Animal Science Department, Polythechnic University of Valencia, Valencia, Spain

One of the major problems of fish is a high risk of quality loss due to oxidation. Lipid oxidation in fish-products leads to rancid taste and off flavor and development of many different substances from which some have even adverse effects to human health. This is more evident in fish rich in n-3 polyunsaturated fatty acids (PUFA) which is susceptible to peroxidation of PUFA resulting in restriction of storage and processing possibilities. Due to the positive health effects of long chain n-3 PUFA, there is an increased interest to produce fish products rich in n-3 PUFA. For this reason, the addition of antioxidants is necessary to increase storage stability, sensory quality and nutritional value of animal products. And this requirement will be more evident in fish with higher content of valuable fatty acids (FA). For animal foods there are always two possible ways to include antioxidants: Via the feed or post mortem during the processing. The inclusion of antioxidant on diet has many advantages since affect directly to the metabolism enhancing other properties that can be important for the final product. Barley is a cereal with and added potential of improving fish health due to the high β-glucan content and antioxidants present on their composition. The objective of this study was to evaluate the effect of inclusion of barley on antioxidant profile.

During twelve weeks rainbow trout was growth in two monitorised and controlled rooms using recirculation system at a temperature of 16 ± 0,7 ºC and 6 ± 0,34 mg/l of oxygen. Each room contains ten tanks and each tank 25 fish. Rainbow trout were fed using isoproteic and isolipidic diets (45% PB, 18% FB). Control diet was compared with diets enriched with barley which contain 5,20% of β-glucans. Each experimental diet contained different concentrations of β-glucan (0,0%, 0,18%, 0,37%, 0,75% and 1,5%). Each four weeks, samples were taken to study total phenols (TP), DPPH, ORAC, ABTS, FRAP and RACI.

The results showed an important effect on the barley dose in the antioxidant activity of the fish fillets. Fillets from fish fed with higher barley dose had higher antioxidant activity, than those fed whitout barley or low doses. These differences were more accused on fish fillets obtained at the end of the growth experiment. This would respond to an accumulate effect of the bioactive compounds during the growth and a significant reduction of free radicals on the products.

Barley inclusion in rainbow trout (*Oncorhynchus mykiss*) diets modified the antioxidant properties of fillets compared to control samples.

Key words**:** rainbow trout, barley, antioxidant ingredients

Effect of PSC hydrolysate, from skin of *Prionace glauca,* in the expression of collagen in human fibroblast cell cultures

Presenting author: Ana Cristina Sánchez Díaz

Co-authors: Begoña Correa García, Marta Pérez Testa, María Blanco Comesaña, Ricardo Isaac Pérez Martín, Carmen González Sotelo

Instituto de Investigaciones Marinas (CSIC), Vigo, Spain

Seafood discards and by-products are not often used as source for obtaining high value-added products. The Skin of *P. glauca* species is a potential raw material for collagen that could be used in diverse fields including food, cosmetic and biomedical industries (Zague et al. 2011).

The aim of present work is to test the effect of collagen hydrolysates, from skin of *P. glauca* species, on the collagen expression in human fibroblast cell culture.

Extraction of pepsin soluble collagen (PSC) from skin of *Prionacea glauca*: Extraction of collagen from *P. glauca* skin was carried out according to the methodology of Liu et al. (2012) with slight modifications. Basically, an alkaline treatment followed by an acid-pepsin extraction in which collagen is extracted by salt precipitation (PSC). The precipitated PSC is resuspended in acetic acid 0.5 N, dialyzed and lyophilized. **Hydrolysis of PSC:** Hydrolysis experiment were carried out in a stirred-batch reactor in deionised water with a lyophilized PSC proportion of 1:10 w/v. Reaction was performed with Alcalase at pH 8 and 55ºC during 3 h with an enzyme/substrate of 1 :20 (w/w). Finally the hydrolyzed PSC was subjected to two steps of ultrafiltration: 10.000 and 3.000 Da to obtain two peptide fractions: Permeate (<3000 Da) and (Retentate ≥ 3000 Da). **Cell Culture**: Human fibroblast cells (Innoprot) were incubated in a 24 well plates (50.000 cells/well) during 24 hours before adding each treatment of peptide hydrolysates (800, 500, 100 and 50 µg/mL of permeates or retentates). Then the plates were incubated 24 h and 48 h. **RNA extraction**: RNA extraction from fibroblast cell plates were carried out with the extraction kit "Cells-to-CT ™ 1-Step TaqMan® Kit" (Ambion). RNA concentration was measured by UV in a Nanodrop and concentrations adjusted to 35 ng/µL. **Collagen expression assay by real time PCR:** TaqMan real time PCR assays were performed with the TaqMan® 1-Step qRT-PCR Mix included in the Cells-to-CT ™ 1-Step TaqMan® Kit" (Ambion) follow the manufacturer's instructions. The specific and house-keeping gen systems used were COL_I and GAPDH whose primes and probe sequences are: COL_I-Forward: ATGCCTGGTGAACGTGGT, COL_I-Reverse: AGGAGAGCCATCAGCACCT, COL_I-Probe: 6-FAM ACCAGC ATCACCTCTGTC-MGB, GAPDH-Forward: GGAAGCTCACTG GCATGGC, GAPDH-Reverse: TAGACGGCAGGTCAGGTCCA and GAPDH-Probe:VICCCCCACT GCCAACGTGTC-MGB under the conditions.

Over expression was observed in fibroblast only when cells were treated with 800 µg/mL of hydrolyzed PSC, in the case of retentates the effect was observed already after 24 h, whereas for permeates 48 h of treatment was needed for observing the same effect. Besides, overexpression is also appreciated with 100 µg/mL of retentate treatment after 24 h. At 48 hours of incubation the collagen overexpression become more evident and increases as concentration rises, reaching a 20% of overexpression for the highest concentration.

These expression data were compared with those obtained for a commercial hydrolysate, Ana Maria de la Justicia, for which a small overexpression was observed in the permeate at 24 h with 100 µg/mL. Overexpression is more evident in wells treated with retentates of *P. glauca* hydrolysate. However, for commercial hydrolyzed, collagen expression increases with decreasing concentration of hydrolysate, contrary to what occurs with *P. glauca* hydrolysate.

Collagen overexpression is more evident in fibroblasts treated with retentate both whit *P. glauca* and commercial hydrolysate. While for *P. glauca* fibroblast, collagen expression increases as hydrolysate concentration increase, for commercial hydrolysate decreases as concentration increases. Both, *P. glauca* and commercial hydrolysates retentates have a molecular weight between 3,000 and 10, 000 Da although average molecular weight of commercial retentates was higher than *P. glauca* hydrolysates retentates. This could explain the difference in the collagen expression pattern.

Key words: collagen, RNA, hydrolysate, fibroblast, blue shark

Production of antioxidative protein hydrolysates from fishery processing by-products by alkaline proteases

Presenting author: Saroat Rawdkuen[1]

Co-authors**:** Sunantha Ketnawa[1], Oscar Martínez Alvarez[2], and Soottawat Benjakul[3]

[1]Food Technology Program, School of Agro-Industry, Mae Fah Luang University, Chiang Rai, 57100, Thailand

[2]Institute of Food Science, Technology and Nutrition (ICTAN-CSIC), José Antonio Novais 10, Madrid, 28040, Spain

[3]Department of Food Technology, Faculty of Agro-Industry, Prince of Songkla University, Songkhla, 90112, Thailand

[*]corresponding author, e-mail: saroat@mfu.ac.th

Alkaline proteases (AP) obtained from viscera of farmed giant catfish (*Pangasianodon gigas*) were used for production of protein hydrolysates from fishery processing by-products including skin gelatin, expired cooked shrimp, and shrimp cooking juice. Antioxidative activities of the protein hydrolysates (PH) were compared to those produced by commercial trypsin (CT) and Izyme AL® (IZ).

The hydrolysis was performed by using 8 units of each enzyme per protein content dissolved in buffer solution (1:3 (w/v) in 0.1 M Tris-HCl pH 8.0), at 55°C for 2 hours. Hydrolysis was controlled by using a pH-stat. Aliquots were collected at 0, 10, 20, 40, 90, and 120 minutes. After enzyme inactivation by boiling, the supernatants were lyophilized, stored at -20°C and referred to PH. The degree of hydrolysis (DH) of the PH was monitored then determined for antioxidant properties including 2,20-azinobis-(3-ethylbenzothiazoline-6-sulfonic acid) scavenging capacity (ABTS), ferric reducing power capacity (FRAP), and metal ion chelating activity (MICA).

AP provided the PH with the highest DH among CT and IZ for all starting materials used, excepting in cooked shrimp. For ABTS, the PH from skin gelatin exposed the highest ABTS when it was hydrolyzed by IZ whereas ABTS of the PH produced by AP and CT were similar (P<0.05). For cooked shrimp PH, the highest ABTS was found in the PH obtained with CT followed by those using IZ and AP. The PH from shrimp cooking juice that were obtained with CT and IZ showed the same ABTS whilst those hydrolyzed by AP provided the lowest value. For FRAP, CT and IZ seem to be the enzymes that hydrolyzed and produced a bioactive peptide pool with the highest FRAP. The PH from shrimp cooking juice exhibited the highest value following those from cooked shrimp and skin gelatin. For MICA, the PH from skin gelatin and shrimp cooking juice showed similar values while the lowest value was obtained from the PH from cooked shrimp. AP was the enzyme which led to obtain the PH from cooked shrimp with the highest MICA over those from and IZ and CT.

The alkaline proteases from giant catfish viscera showed possibility to replace the commercial trypsin, especially in Muslim markets. Moreover, this enzyme could be used for production of antioxidative protein hydrolysates from fishery processing by-products to obtain potential functional ingredients for any incorporation in food formulation.

Key words: alkaline proteases, antioxidative, cooked shrimp, gelatin hydrolysates, viscera

Control of sea lice in aquaculture using ultrasound – a feasibility study

Presenting author**:** Martin H. Skjelvareid

Co-authors: Atle Mortensen

Nofima, Tromsø, Norway

The sea louse parasite, especially the *Lepeophtheirus salmonis* and various *Caligus* species, poses a major problem for the salmon aquaculture industry. The sea lice migrate as free-swimming larvae and latch on to the skin of the salmon, causing physical damage and disease as they feed on their host. The concentration of sea lice can become very large in salmon farms, and lice spreading from farms are seen as a major threat to wild salmon.

Several methods for sea lice control have been developed, including special nets for fish cages, chemical treatment, flushing and brushing, and cleaner-fish which eat the parasites. However, the existing methods are either ineffective, too expensive, stressful for the fish or harmful to the environment.

Ultrasound has previously been used as a method to prevent fouling on ship hulls and in pipes, and is suggested as a possible method for inhibiting and/or killing sea lice. Ultrasound has the advantages of being inexpensive, easy to install in fish cages, and theoretically harmless to the fish.

In this feasibility study, we have reviewed the existing literature on ultrasound as a method for pest control, performed calculations on ultrasound propagation in sea water, and evaluated the feasibility of practical application of ultrasound in aquaculture.

The application of ultrasound can be divided into two major categories, based on the sound pressure involved. If the sound pressure is sufficiently high, ultrasound will create cavitation bubbles in the water, and the collapse of these bubbles results in a very violent environment with shock waves and locally very high temperatures and pressures. This could potentially kill sea lice. However, only a small volume of water can be treated, and cavitation is potentially harmful to the fish.

If the sound pressure is under the cavitation threshold, ultrasound cannot be expected to cause mortality for parasites like sea lice larvae. However, studies on barnacle larvae have shown that long-time exposure to medium-intensity ultrasound can cause inhibition of the larvae. If the same applies to sea lice, ultrasound could be used as a method to prevent attachment to the fish. Continuous treatment of whole fish cages may be possible.

Ultrasound is seen as a potential method for sea lice control, with several advantages over existing methods. Further studies are needed on the reaction of sea lice to ultrasound. Continuous insonification of fish cages with medium-intensity ultrasound is seen as the most promising mode of operation.

Key words: sea lice, salmon, ultrasound, aquaculture, pest control

Sustainable feed formulation for Swedish aquaculture from herring by-products

Presenting author: James Hinchcliffe[1,2]

Co-authors : Mehdi Abdollahi[1,3] , Elisabeth Jönsson Bergman[2], Susanne Erikson[2], Kristina Sundell[2] , Ingrid Undeland[1]

[1] Department of Biology and Biological Engineering, Food and Nutrition Science, Chalmers University of Technology, Gothenburg, Sweden.

[2] Department of Biological and Environmental sciences, Gothenburg University, Gothenburg, Sweden.

[3] Department of Fisheries, Faculty of Marine Sciences, Tarbiat Modares University, Iran

Aquaculture is expected to be the fastest growing food sector this century. Small pelagic fish such as herring, *Clupea harengus,* are currently used in fishmeal, a feed constituent that is used as the main marine source of protein in aqua feeds (FAO 2012). In Sweden, work is being done to establish a knowledge base for the development of sustainable marine aquaculture, focusing on two species: Spotted wolffish, *Anarhichas minor* and European lobster, *Hommarus Gammarus*. One of the aims is to develop new feeds for the two species during larval and juvenile on-growth stages. Although both wolffish and lobster larvae can be given formulated feeds at early weaning, previous studies have shown that live feeds are still superior regarding larval survival, growth and development. Currently, formulated feeds are highly dependent on raw materials from wild fisheries and are costly to produce. The development of diets composed from sustainable materials with suitable texture that increases survival and quality is an important challenge. In this study, pH-shift processing was used to isolate proteins from herring by-products, in order to investigate the potential incorporation of herring protein isolates as an alternative protein source in aqua feeds.

Herring by-products were sorted into back bones, heads and guts which thereafter, in different combinations, were subjected to pH-shift processing. For each raw material combination, the acidic version of the pH-shift process was compared with the alkaline process version to identify protein yield as well as potential functional and chemical differences in protein isolates, and how these could affect the potential of isolates as a feed ingrediant. Among studied factors were mineral content (e.g. calcium), fatty acid pattern, flotability and palatability.

Initial results show that the highest protein yields were given with heads and spines, and, also that the alkaline version of the pH-shift process generally gave higher yields than the acid version. Functional and chemical differences were found in isolates from different treatments and this will have consequences on future feed incorporation. For example, the acid process version generated higher calcium levels in the protein isolates than the alkaline version, something which is particularly important in lobster feeds. Also, preliminary feeding trials indicate satisfactory flotability and palatability of some isolates for both lobster and wolfish.

Key words**: a**quaculture, herring, by-product, fishmeal, pH shift, protein isolate

Growth hormone transgenesis influences muscle protein expression in Coho salmon (*Oncorhynchus kisutch*)

Presenting author: Flemming Jessen[1]

Co-authors: Robert H. Devlin[2], Daniel J. Macqueen[3], Dwight R. Causey[3]

[1]Division for Food Technology, Research Group for Food Production Engineering, National Food Institute, Technical University of Denmark, Kgs. Lyngby, Denmark

[2]Department of Fisheries and Oceans, University of British Columbia, Centre for Aquaculture and Environmental Research, West Vancouver, BC, Canada

[3]Institute of Biological and Environmental Sciences, University of Aberdeen, Aberdeen, AB24 2TZ, United Kingdom

Growth hormone (GH) transgenesis (GHT) leads to increased growth, feed intake and metabolic rate in fish. The genetic pathways involved include energy metabolism of carbohydrates, lipids, and protein, protein synthesis, stress and immune function [1]. These findings are consistent with previous data on GHT fish, showing alterations in many processes, including nutritional requirements, energetics, muscle fibre structure, and cartilage deposition (for references see 1). While gene expression studies can be conducted at both the mRNA and protein level, the resulting outputs are complementary rather than redundant, due to the non-linear relationship between protein expression and processes governing mRNA production, stability and translation rates. To date, analyses of GHT in fish have focussed on the mRNA level, via microarrays, qPCR and more recently RNAseq. However, to date, the impact of GHT on the proteome is poorly understood in fish – the present work aimed to fill this gap in knowledge.

Muscle samples from size matched non-transgenic (n=8) and GHT transgenic Coho salmon (n=8) were analysed by two-dimensional gel electrophoresis (2DE). Samples taken from freezer (-80°C) were immediately homogenized in buffer and centrifuged and the supernatant used for 2DE. Fixed gels were stained with Coomassie Brilliant blue, digitized (CCD camera), and subjected to image analysis (Progenesis SameSpots). Selected protein spots were identified by mass spectrometry.

More than 700 individual spots were common to all 2DE gels and included in data analysis. Close to 100 spots differed significantly ($p<0.01$) between the two groups. Approximately two thirds were present in higher amounts (spot volumes) in the GHT compared to the non-transgenic group, thus representing proteins upregulated by GHT. The remaining third of the spots represent proteins reduced by GHT. While we are yet to identify the selected spots, this will be done using Mass Spectrometry in the near future, allowing the nature of biological pathways altered by GHT to be discussed.

GHT caused marked changes in the muscle proteome of Coho salmon – the nature of the pathways underpinning these changes are currently being elucidated and will be described in this poster.

References: Devlin et al. PNAS 2009, 106, 3047–3052

Key words: growth hormone; transgenesis; muscle; proteomics; coho salmon

Pikeperch (*Sander luciopera*) from recirculating aquaculture systems – quality assessment and comparison with frozen products from retail

Presenting author: Monika Manthey-Karl[1]

Co-authors: Gregor Schmidt[2]

[1]Max Rubner-Institute, Federal Research Institute of Nutrition and Food, Department of Safety and Quality of Milk and Fish products, Palmaille 9, 22767 Hamburg (monika.manthey@mri.bund.de)

[2]State Research Center for Agriculture and Fisheries MV, Institute for Fisheries, Malchower Chaussee 1, 17194 Hohen Wangelin, (g.schmidt@lfa.mvnet.de)

Pikeperch (*Sander luciopera*) is considered to have a high potential for recirculating aquaculture systems (RAS). Constant high temperatures (22-24 °C) and a good water quality ensure satisfactory growth rates and enable a **production of 1 kg fish in approx. 15-18 months.** In Germany, pikeperch has a great consumer acceptance and is demanded the year round. The market value is high at 8.5 - 14 € /kg whole fish at farm gate. Its boneless flesh has an aromatic typical taste and is suitable for different forms of preparation. However, in supermarkets deep frozen skin-on fillets of wild pike-perch dominate, mostly available from Kazakhstan and the Russian Federation. Due to limited potential of the production in natural waters and ponds the expansion of pike-perch culture depends on the successful development in RAS. Within an EU financed project the influence of different formulated diets on the growth performance and the product quality of fish at market size was investigated. For comparison frozen fillets from retail were analysed.

The entire production cycle of the pikeperch was figured out in a commercial-scale aquaculture system. For grow-out two groups were fed different commercial diets usually used for sturgeon production over a period of four months, containing a high protein content of > 50% and a low fat content of < 15%. The fillets of fish at market size of 1.2 kg were analysed for proximate, fatty acid and free amino acids composition and selenium. A sensory panel assessed the sensory quality. Six products from retail were equally analysed and compared to aquaculture-products.

Irrespective of the diet, both groups of reared fish showed comparable growth performances and fish conditions. After four months fish achieved a mean weight of 1121,2 ± 237,95 and 1131,0 ± 213,89 g and a total length of 51,4 ± 3,47 and 51,4 ± 3,08 cm, respectively (n=25). The fillet composition of cultured fish was similar and comparable to wild pikeperch. The fat content of the muscle flesh slightly increased due to the feed applied. However, it remains lean with 1.6% compared to wild counterparts with less than 1%. High energy feed stuff did not increase intramuscular fat and led to excessive abdominal fat. In summary, amounts of docosahexanoic acid between 16.6% - 20.8% and eicosapentaenoic acid between 5.2% - 6.8% were estimated. The sensory quality was good. Musty/mouldy taste which is often appearing in fish from RAS was no problem and only found to a slight and acceptable extent. Frozen pike perch fillets from retail had a very different quality. Main deviations were an untypical smell and a sour to bitter aftertaste.

Both diets were suitable for pikeperch cultivation, no significant differences in growth performance or fish condition were determined. But based on the unpredictable uptake of adult pikeperch, it is necessary to avoid feeding losses with a very accurate feeding and control system**.** The fish quality of all groups was good. The tested formulated diets yielded similar production values. The partly bad sensory evaluation was the main difference of frozen fillets from imported pike perch.

Effect of dietary barley on rainbow trout diets in a stress challenge

Presenting author: Julia Pinedo Gil[1]

Co-authors: Ana María Larrán García[1], Cristina Tomás Almenar[1], Silvia Torrecillas Burriel[3], Mª José Caballero Cansino[3], Miguel Jover Cerdá[2], Ana Tomás Vidal[2], Miguel Sanz Calvo[1], Ana Belén Martín-Diana[1].

[1]Subdirection of Research and Technology. Agro-Technological Institute of Castilla y León. Consejería de Agricultura y Ganadería. Finca de Zamadueñas. Ctra. Burgos km. 119, 47171, Valladolid, Spain.

[2] Aquatic Resources Research Group, Animal Science Department, Polythechnic University of Valencia, Valencia, Spain.

[3] Aquaculture Research Group (GIA), Instituto Universitario de Sanidad Animal y Seguridad Alimentaria (IUSA), Universidad de Las Palmas de Gran Canaria, C/ Transmontaña s/n 35416, Arucas, Las Palmas de Gran Canaria, Canary Island, Spain.

Due to the rapid growth and easy accommodation to environmental conditions, rainbow trout (*Oncorhynchus mykiss*) is one of the most important freshwater cultured fish. European rainbow trout production represents 21% of worldwide production and Spain occupied 10%. These efforts are not only focused on the substitution of traditional sources of protein and oils, also new trends are focused on the inclusion of new ingredients with bioactive properties. In this sense probiotics, prebiotics, immunoestimulants and other ingredients have gain importance during the last years. For animal food there are two possible ways to include these compounds: as concentrate or additive including on the diet or as part of the raw material. The incorporation of these compounds through raw materials reduces the cost and many times help to revalorise byproducts from the agro-industry. Cereals are usually incorporated in aquaculture feeds as a source of starch and energy yielding substrate. Barley has not widely used as ingredient in fish feed, although few studies showed that its incorporation had not detrimental effect on growth parameters. One of the main properties of barley is the content on β-glucans and the acceptance of β-glucans as functional, bioactive ingredient has increased its popularity and its potential due to its immunoestimulant effect. Although many studies have been carried out in rainbow trout and the effect of different ingredients on the diet, few studies have been focus on the effect of β-glucans and less have been investigated in the inclusion of barley on the diet. The aim of this study is to evaluate the effect of dietary barley`s β-glucans against a stress challenge through the growth performance and liver morphology and histology.

During 78 days rainbow trout were growth in 2 monitorised and controlled rooms using recirculation system at a temperature of 14-17ºC and dissolved oxygen levels of 7-9mg/l. Each room contained 10 tanks and 25 fish per tank. At the middle of the experimental period fish were submitted to an acute stress by decreasing the concentration of oxygen to 4mg/l during 10 minutes. Rainbow trout were fed 5 isoproteic and isolipidic diets (45%CP and 18%CF) containing graded levels of β-glucans (00GLUC (0,0%), 018GLUC (0,18%), 037GLUC (0,37%), 075GLUC (0,75%) and 15GLUC (1,5%). Before and after the stress period final weight (Wf), specific growth rate (SGR), feed conversion ratio (FCR), survival, hepatosomatic index (HIS) and blood biochemical parameters (glucose, lactate and cortisol) were determined. Besides 3 individual fish liver per tank were histologically evaluated (40x, H&E) as described before by Torrecillas et al. 2007, 2011a, b. The concentration of barley showed a significant effect on the biochemical and histological parameters. Glucose and cortisol levels showed significant ($p<0,05$) effect of the diet meanwhile lactate did not show any significant modification regarding of the barley concentration. Glucose and cortisol increased significantly ($p<0,05$) during the stress period and this increment was more accused on fish fed diets with higher concentrations of barley, having the diet 075GLUC the highest level during the stress period. However, while cortisol decreased significantly ($p<0,05$) with higher concentrations of barley, glucose levels continue increasing till the higher concentration of barley experimented (15GLUC). These results were in agreement with Barton *et al*. (2000). Both, glucose and cortisol levels recovered after one week to basal values without significant differences between diets. No significant differences were observed during the experimental growth period before and after the stress challenge, however, significant differences ($p<0,05$) were observed on FCR immediately after stress associated to the diet. The lowest FCR value were observed in fish fed 075GLUC meanwhile fish fed without barley or 15GLUC showed higher values.Histological and morphometrical markers showed no significant differences during the experimental period before the stress challenge. However, hepatic lymphocytes loci increased significantly ($p<0,1$) after stress, presenting more number of lymphocytes loci those fish fed 075GLUC, while a higher concentration of β-glucan (15GLUC) presented no significant differences with the control diet (00GLUC). No differences in the hepatocytes areas and vacuolization were observed after stress.

The inclusion of barley in rainbow trout diets induce mobilization of immune cells due to the concentration of β-glucans but just until 0,75% β-glucans, above this level the low absorption by the high fiber content in the diet can decrease the mobilization effect of β-glucans. Those concentrations of barley also enhance FCR values, what means an improvement of growth parameters. This mobilization effect is known for other products with benefits on growth, pathogen resistance, etc. Further studies should be done to study the effect of the inclusion of barley in target organs such as intestine.

Key words: rainbow trout, liver morphology, biochemical parameters and growth parameters.

Free amino acid content of Pacific cupped oysters (*Crassostrea gigas*) fed with *Skeletonema costatum* or *Rhodomonas baltica*

Presenting author: Jasper van Houcke[1]

Co-authors: Hanne K. Maehre[2], Jozef Linssen[1], Joop Luten[1]

[1]Wageningen UR, Dept. Food Quality and Design: Wageningen, The Netherlands and HZ University of Applied Sciences, Delta Academy, Vlissingen The Netherlands

[2]The Norwegian College of Fishery Science, UIT The Arctic University of Norway.

Free amino acids (FAAs) are known to have an effect on the sensorial properties of seafood. Concentrations of specific FAAs in oysters are known to be influenced by season and cultivation location. It is therefore hypothesized that the diet of the oysters plays a role in the FAAs present in oysters and could therefore influence the sensorial aspects of oysters.

We have studied the impact of the algae feed *Skeletonema costatum* and *Rhodomonas baltica* on the FAAs of the Pacific cupped oyster originally from the important Dutch cultivation site Lake Grevelingen.

Refined oysters were produced by feeding market-sized Pacific cupped oysters (60-80 g) with either the diatom *Skeletonema costatum* or the flagellate *Rhodomonas baltica* (both at 30 mg dry weight algae day^{-1} oyster^{-1}) for a period up to seven weeks in land-based pond systems. Oysters sampled from Lake Grevelingen were used as reference oysters (non-refined). Besides taking samples from the initial oysters (T0) samples were also taken after four (T4) and seven weeks (T7).

For each group, 10 individual oysters were freeze-dried for FAAs analysis. FAAs were extracted using hydrolysis and analyzed using a Biochrom 30 amino acid analyzer. The amino acids were chromatographically separated on an ion exchange column, followed by post-column derivatization with ninhydrin and detection of UV signal at 440 nm and 570 nm. UV-signals were analyzed by Chromeleon software and compared with physiological amino acids standards.

Results showed a significant ($P<0.05$) time effect for specific FAAs. The content of aspartic acid increased over time in *Skeletonema* fed oysters (3.8, 4.5 and 4.9 mg g oyster tissue^{-1}, respectively for T0, T4 and T7) and *Rhodomonas* fed oysters (3.8, 5.0 and 5.1 mg g oyster tissue^{-1}). While the content of alanine decrease over time in *Skeletonema* fed oysters (9.0, 6.0 and 4.7 mg g oyster tissue^{-1}, respectively for T0, T4 and T7) and *Rhodomonas* fed oysters (9.0, 4.2 and 4.0 mg g oyster tissue^{-1}). These changes might be explained by seasonal influences which have been reported in literature.

Only alanine content showed to be significantly different ($P<0.05$) between *Skeletonema* or *Rhodomonas* fed oysters at T4 (6.0 and 4.2 mg g oyster tissue^{-1}, respectively).

Feeding Pacific cupped oysters with a monospecific algal diet changes the FAA content over time. Differences in FAA content between *Skeletonema* or *Rhodomonas* fed oysters are small and in most cases not significant.

Key words: Pacific cupped oyster, algae diet, free amino acid, aspartic acid, alanine

Live storage of Atlantic cod (*Gadus morhua* L.) without feeding effects on fillet quality

Presenting author: Tatiana Ageeva[1]

Co-authors: Ragnar L. Olsen[2], Sjurdur Joensen[1], Stein Harris Olsen[1], Margrethe Esaiassen[2]

[1]Nofima, Tromsø, Norway

[2]Norwegian College of Fishery Science, UiT The Arctic University of Norway, Tromsø, Norway

Atlantic cod (*Gadus morhua* L.) is one of the most important commercial species in the North Atlantic and the peak harvesting season in Norway is from January to March. Live storage of cod is a promising way to extend the marketing season for fresh cod products. The aim of this study was to investigate quality changes in non-fed live stored wild cod from this period.

Atlantic cod were caught in Lofoten in March 2015 using “Danish seine” and stored alive without feeding in net cages for 12 weeks at the Aquaculture Research Station in Tromsø.

Fish (initial weight 6.8 ± 2.1 kg and length 90 ± 8 cm) were sampled after 0, 4, 8 and 12 weeks of storage by stunning the fish followed by cutting the gills and bleeding in cold seawater for 30 min before gutting. The fish was filleted 48 h post mortem and used for different quality analyses after ice storage for 7 days.

The results showed that most of the weight loss during live storage was due to weight reduction of gonads. The gonadosomatic index (GSI) decreased considerably during the first 8 weeks of starvation (from 13.2 to 1.8 %), but minor changes was registered during the next 4 weeks of storage. The decrease in hepatosomatic index (HSI), however, was evident during the last 4 weeks of live storage (from 4.8 to 3.0). Likewise, weight loss of muscle, measured as weight loss of gutted fish, occurred mainly during the last 4 weeks of storage. The weight loss was 3.3, 4.6 and 15.4 % after 4, 8 and 12 weeks of storage, respectively.

During the live storage, water content in muscle increased from 82.0 to 84.3 %. Water Holding Capacity (WHC) of muscle decreased from 90.5 % after 4 weeks, to 85.2 after 12 weeks of starvation. As measured by proportion of fish with atypical white muscle colour and gelatinous texture, quality defects developed mainly during the last 4 weeks of storage. The proportion of fish with quality defects concurred with proportion of fish with gutted K-factor < 0.6.

Live storage of cod is a promising way to extend the marketing season for fresh cod products. The results shows that cod caught in March could be stored live without feeding for at least 8 weeks without considerable quality changes.

Key words: lived stored cod, starvation, weight loss, water holding capacity, quality

Effect of freezing on the quality of hake stored in modified atmosphere

Presenting author: Juan Calanche[1,2]

Co-authors: Isabel Buisan[1], Selene Pedrós[3], Veronica Alonso[1], José Antonio Beltrán[3]

[1]Department of Animal Production and Food Science and Technology. Meat and Fish Science and Technology Laboratory. Faculty of Veterinary. University of Zaragoza. Zaragoza-Spain.

[2]Department of Food Technology. School of Applied Sciences of the Sea, Nueva Esparta Core, University of Orient, Boca del Río National Road, Nueva Esparta, R.B. Venezuela.

[3]School of Veterinary Medicine, University College Dublin, Belfield, Dublin 4, Ireland

Whitefish market is one of the largest segments in the commercial seafood market, and also one that has experienced profound change in recent years (Asche & Guillén, 2012). In Spain, and indeed in other European countries, gadoid fish represent an important percentage of overall fish consumption (MAGRAMA, 2013). In this sense, this research was aimed at evaluating the effect of freezing on the quality of Cape hake (*Merluccius paradoxus*) stored in modified atmosphere (MAP) and under refrigeration. Hake steaks previously frozen and fresh hake steaks (control) were packaged in modified atmosphere (60% CO_2/40% N_2) and refrigerated at 4°C. They were analyzed over time using physico-chemical, microbiological and sensory analyses. Total volatile basic nitrogen (TVB-N) showed no significant differences between samples, they exceeded the established legal limit after 96 h of storage. Both pH and the internal temperature (IT) were no different between samples. There was, however, significant differences ($p < 0.05$) during the storage. Mesophilic bacteria (MVC) and psychrotrophic (PST) counts were not different between fresh and frozen samples. However *Enterobacteriaceae* count (ET) was higher in frozen samples after 48 h while specific spoilage organisms (SSO) prevailed in fresh ones. Similar behavior exhibited proteolytic bacteria (PB) count that was always higher in fresh hake, while lactic acid bacteria (LAB) dominated in frozen hake. Sensory analysis of cooked hake did not detect significant differences for the quality index between treatments and the assessments were deficient after 96 h. Several sensory aspects (odor, flavor and texture) were deteriorated similarly in fresh hake highlighting the correlation of LAB with flavor while SSO did it with smell. Texture was the most affected aspect in frozen hake and it correlated well with TVB-N production and the growth of MVC and SSO. Shelf life of MAP hake was established at 96 h and taking into account results obtained, mathematical models were performed using partial least square regression (PLSR). Three equations were obtained for rapid assessment of important quality parameters like shelf life (SL) and microbiological count (MC) which exhibited R-squared and slope ≥ 0.98 and accuracy factor (*Af*) > 92% in an internal validation.

Key words**:** Gadidae, specific spoilage organisms, predictive model,shelf-life

Evaluation of lemongrass essential oil on the reduction of *Listeria monocytogenes* in artificially contaminated ready-to-eat food

Presenting author: Evren Burcu Şen Yilmaz[1]

Co-authors : Şükran Çakli[1] , Kitiya Vongkamjan[2]

[1]Faculty of Fisheries, Section of Processing Technology, Ege University, Bornova, Izmir Turkey

[2]Department of Food Technology, Prince of Songkla University, Hat Yai, 90112 Thailand

Listeria monocytogenes is an important foodborne pathogen that can cause a severe infection in human (listeriosis) from the consumption of contaminated food. Contamination of ready-to-eat (RTE) foods with *L. monocytogenes* can typically be traced back to post processing contamination from environmental sources. Various growth inhibitors or preservatives have been used for the reduction of *L. monocytogenes* by food producers. One of the natural antimicrobial essential oil (EO) obtained from lemongrass was evaluated against *L. monocytogenes* in this study. Lemongrass EO was used as a dipping solution at a ratio of 1/8 (EO/Water) for RTE sushi squid and cooked shrimp samples.

Each food sample (approximately 10 g, 5 x 5 cm^2) was inoculated with approximately 4 log CFU/ml of a mixed culture of two *L. monocytogenes* isolates (PSU-KV-033LM, LM33 and PSU-KV-120LM, LM120), previously obtained from the seafood processing plant environment (Vongkamjan et al., 2015). Inoculated RTE seafood samples were separately treated by dipping with lemongrass essential oil solution for 30 sec or 1 min. Enumeration of surviving *L. monocytogenes* counts was performed by duplicate platting on Modified *Listeria* Selective Oxford Agar (MOX), followed by inception at 30°C for 48 h.

L. monocytogenes showed significantly lower in sushi squid samples treated by a dipping method for 30 sec or 1 min in lemongrass EO solution compared with samples dipped in water and control group (only inoculated with *L. monocytogenes*). Similarly, in cooked shrimp inoculated with a mixture of LM33 and LM120, samples treated with with lemongrass EO solutions by a dipping method showed a reduction of *L. monocytogenes* as compared to treatment with water and the control group.

Our study showed that RTE sushi squid and cooked shrimps showed a reduction of *L. monocytogenes* after treatment with lemongrass EO. Results suggest use of lemongrass EO as an alternative control strategy against *L. monocytogenes* to reduce the incidence of food production contamination.

Key words: lemongrass essential oil, *Listeria monocytogenes*, ready-to-eat food, reduction, post-processing

Effect of high pressure processing on fish protein oxidation

Presenting author**:** Cécilia Arnaud

Co-authors: Laurence Pottier, Marie de Lamballerie

ONIRIS, Nantes, France

High-Pressure Processing (HPP) is an innovative technology for food preservation. This treatment permits to increase the shelf-life and to preserve the original taste as well to obtain healthier and environmental-friendly products. 13% of HPP applications in the world concern seafood and fish: ready-to-eat seafood meals, lobster meat extraction, clams, oysters and mussels shucking. The alteration of proteins by oxidation might occur and lead to textural, flavour and nutritional modifications. A treatment of 150 MPa destroys the parasites *Anisakis* and *Trichinella* whereas a pressure higher than 450 MPa are necessary to inhibit microorganism growth. The aim of this study was to evaluate protein oxidation on a lean fish (cod) and a fatty one (salmon) for two HPP levels: 150 or 450 MPa.

Cod and salmon fillets were vacuum-packed in polyamide/polyethylene plastic bags. High Pressure treatment was carried out using a vertical 3 L high-pressure pilot unit. Pressure of 0.1 (control), 150 and 450 MPa was applied and held for 5 min. The treated samples were stored at 4 °C during 14 days. Protein carbonyl contents were measured to quantify the protein oxidation via the DNPH-test (dinitrophenylhydrazine).

At atmospheric pressure, DNPH value for cod was 2-fold higher than for salmon and in accordance with literature [1]. Over the storage time, cod and salmon showed an approximate 1.4-fold increase in the carbonyl contents after 7 days and a stagnation for longer storage time.

After HPP, independently of the storage time, cod and salmon samples treated at 150 MPa showed protein carbonyl contents close to the control but 450 MPa-treated samples showed higher values (3-fold gain at day 0 and day 7). Globally, over 7 days of storage, HPP treated samples showed a 1.4-fold increase of carbonyl contents such as control. 450 MPa treatment increased protein oxidation while 150 MPa treatment did not oxidize more protein than the natural trends for both fishes. After 7 days of storage, cod and salmon are globally oxidized in a same rate. From 14 days of storage, protein carbonyl contents decreased for some samples indicating that complementary reactions might occur.

Protein oxidation is an interesting and emerging topic. HPP treatment increased protein oxidation of products. In future work, it will be interesting to study the potential correlation with the presence of lipids in both matrices.

[1] Tokur B., Korkmaz K., 2007. The effects of an iron-catalyzed oxidation system on lipids and proteins of dark muscle fish. *Food Chemistry*, 104, 754-760.

Key words : high pressure processing; protein oxidation; cod; salmon; storage

Changes in physicochemical properties of sea scallop myofibrillar proteins in response to low-temperature thermal processing

Presenting author: Denise Skonberg[1]

Co-authors: Jonelo Sobreguel[1], Jason Bolton[2]

[1]School of Food and Agriculture, University of Maine, Orono, Maine, U.S.

[2]Cooperative Extension, University of Maine

Traditional thermal processing can be detrimental to the texture of sea scallop adductor muscles, leading to the development of tough or rubbery meat texture. In contrast, scallop and other shellfish processed under vacuum at low temperatures tend to maintain their intrinsic qualities. In a previous study we observed significant differences in texture, water holding capacity, moisture content and cook loss in scallop meats subjected to small differences in processing temperature (55, 60, 65ºC) at appropriate pasteurization times. The objective of the present study was to more clearly understand the underlying physicochemical changes in myofibrillar proteins associated with these variations in scallop meat quality.

Scallop meats were sealed in plastic pouches (6 meats/pouch) under 95% vacuum and assigned to one of four treatments : raw (control), 55ºC (208 min), 60ºC (45 min), and 65ºC (10 min) that were processed in a circulating water bath in quadruplicate. Thermal processing time and temperature combinations for each treatment were calculated based on delivering a 6D process for *L. monocytogenes*, and timing began once scallop internal temperature reached the target set point. Scallops were immediately cooled to < 3ºC and subjected to salt soluble protein analyses. Myofibrillar protein was extracted for determination of Ca^{2+}ATPase activity, sulfhydryl group and carbonyl content, and muscle was stored at -80ºC for differential scanning calorimentry. Statistical differences in quantitative data were determined using one-way ANOVA ($P < 0.05$) and means separation performed using Tukey's HSD test.

As processing temperature increased, measures of protein denaturation and oxidation in scallop meats increased, although differences between the 55ºC (208 min) and 60ºC (45 min) treatment were not significant. In particular, salt soluble protein, an indirect measure of protein denaturation, was strongly influenced ($P<0.001$) by processing treatment, with values of 20.6, 10.7, 9.9, and 5.9 mg/g meat for the control, 55, 60, and 65ºC treatments, respectively. Data obtained from the current study strongly support the differences in scallop meat quality observed previously, with the 65ºC treatment displaying the greatest changes in protein structure, despite the very short processing time.

As consumer interest in minimally processed foods increases, the industry will need to balance safety with optimal food quality, particularly for high-value, specialty seafoods. These results can help processors to better understand the effects of low temperature thermal processing on the underlying physicochemical changes in scallop muscle and to deliver high quality products.

Key words**: s**callops, myofibrillar protein, thermal processing

Determining the differences of fried and oven baked fish chips, produced with sardine flesh (*Sardina pilchardus*)

Presenting author: M.Tolga Dincer

Co-authors: Hulya Sargin, Arzu Burcu Yavuz, Burcak Pir, M. Cagil Ucok,

Ege University Faculty Of Fisheries, Izmir,Turkey

This study is about the production of potato chips which includes 45% sardine flesh (*Sardina pilchardus*). The effects and the results of heat treatments; frying and oven baked cooking techniques were compared in last products. Two types of fish chips were produced and many parameters were compared. The question “which chips are healthier” was trying to answer. Acrylamide levels, fatty acid compositions, chemical compositions and shelf life of the products were compared.

As a raw fish material sardine (*Sardina pilchardus*) flesh was used. To prepare the dough mash potatoes, corn starch, spices, salt, sunflower oil, water and some less additives were used. The production was begin with evaluating the dough by using chopper, stuffing the dough in to case to make it uniform and slicing after particularly frozen than completed after frying or oven baked those chips slices. Modified atmosphere packs were used as a chips package and 100% nitrogen gas was filled. Storage of the packs was done in room conditions. To determine the shelf life of the chips chemical, microbiological and sensory analysis was carried out. At the same time colour and textural changes were also observed. And to determine the health risk and the benefit, acrylamide levels, fatty acid compositions, atherogenic index and thrombogenic index levels and chemical composition were observed.

As known sardine is a risky specie for oxidation, has a unique flavour and high lipid content. Although analysis are going on no oxidation and spoilage have been determined in 70 days and expected shelf life will be more than 90 days. The effects of frying and oven baked were shown in Omega 3 content and acrylamide levels of the products. But when compared with the commercial potato chips products still have lover levels of acrylamide and lower levels of lipids. Due to the sensorial panels, panellists decided that, this product may be preferred by consumers who like fish taste and fish smell. Lower levels of salt, fat and higher levels in protein contents were the profits when compared with commercial potato chips.

Seafood consumption in Turkey is still low when compared with the EU countries. Integration of fish flesh to highly consumed potato chips may be one of the solutions to give this consumption habit to the consumers. And also addition of fish, made potato chips product healthier in both fried and oven baked products.

Acknowledgment

*This study was supported by TUBITAK Project No: 114O671. Used data were taken from the sardine production.

*Co-author Arzu Burcu Yavuz taking a PhD scholarship from the programme of Tubitak

Effect of different heat treatment on lipid quality and fatty acid profile of seabass *Dicentrarchus labrax* fillet

Presenting author: Salah Selmi

Co-authors: Rana Guiloufi, Noureddine Ben Nacef

National Institute of Nutrition and Food Technology. Tunis, Tunisia.

Changes in biochemical and nutritional quality of seabass *Dicentrarchus labrax* fat during three cooking process (baking, frying and grilling) were investigated. N-3 and n-6 polyunsaturated fatty acids PUFA levels were 17.75 % and 14.8 % in fresh sample respectively in which docosahexaenoic acid C22:6 n-3 and linoleic acid C18:2 n-6 were the most abundant. Fatty acid profile was not affected by baking process, however, monounsaturated and polyunsaturated fatty acid levels significantly changed after frying method. Therefore, n-3 and n-6 PUFA decrease from 17.75 to 11.65% and increase from 14.8 to 28.12% respectively. Grilling process affect slightly the fatty acid composition with an increase of PUFA. N-3/n-6 index and PUFA/SFA ratio showed that frying method affect more the fatty acid profile. Thermal procedures induced only slight oxidative changes in seabass flesh immediately after treatment and do not generate *trans* fatty acids.

Key words: n-3 and n-6 polyunsaturated fatty acid, trans fatty acid, cooking methods, seabass.

Interactions of lipid and protein changes with bioelectrical measurements in frozen-stored hake and mackerel

Presenting author: Sanja Vidacek[1]

Co-authors: Tibor Janci[1], Ivana Ilic[1], Emira Jakupovic[2], Nives Marusic[1], Helga Medic[1], Igor Lackovic[3]

[1] University of Zagreb, Faculty of Food Technology and Biotechnology, Zagreb, Croatia

[2]Anamarija Company doo, Zagreb, Croatia

[3] University of Zagreb, Faculty of Electrical Engineering and Computing, Zagreb, Croatia

In our earlier studies, the potential of the bioelectrical measurements for differentiation the frozen fish fillets with low and high lipid content was evaluated. The results showed that the frozen fillets with low lipid content may be differentiated by bioelectrical measurements and that these measurements possibly reflect changes on proteins during frozen storage, while in fish with higher fat content, correlation between impedance measurements and protein changes (protein solubility) was moderate. As expected, it was observed that the protein solubility was strongly negatively correlated with some products of fatty acids oxidation in fish with high lipid content.

With the aim of studying the complex relationships among changes on proteins, lipids and bioelectrical measurements in frozen fish, in this study we measured changes on several fish species during 5 months of frozen storage. The HP 4294A Precise LCR meter was used to measure impedance magnitude (|Z|) and phase (φ) at 200 frequencies from 100 Hz to 100 MHz. The protein solubility, TVBN and the water holding capacity were determined. Unsaturated fatty acids and several aldehydes, alcohols and ketones were quantified. The results were statistically evaluated in SPSS with the aim of better understanding the bioelectrical phenomena in fish tissues and determining the best indicators of frozen storage deterioration.

The results on two fish species – hake and mackerel - will be presented.

Key words: freezing, bioelectrical properties, protein, lipid

Fatty acid compositions of cyprinid fish oils (*Carassius gibelio*) extracted from silage treated with formic acid and different bacteria strains

Presenting author: Serhat Ozkutuk[1]

Co-authors:Gulsun Ozyurt[2], Yesim Ozogul[2], Serhat Ozkutuk[1], Caner Enver Ozyurt[3], Merve Erguven[2], Yılmaz Ucar[2]

[1]Cukurova University, Vocational School of Yumurtalik, Adana, Turkey

[2]Cukurova University, Faculty of Fisheries, Department of Seafood Processing Technology, Adana, Turkey

[3]Cukurova University, Faculty of Fisheries, Department of Fishing and Fish Processing Technology, Adana, Turkey

Fish silage has promising potential to produce high quality animal food from discard fish and seafood wastes, preventing environmental pollution. Since fish meal is costly and unavailable in some countries, fish silage may be an alternative to fish meal. Fish silage can be produced either by organic and inorganic acid or by fermentation with bacterial cultures and a source of carbohydrate. It is important to separate lipid from fish silage, for the improvement of its shelf-life. This separated lipids can be use as a food supplement for human or animal consumption. Therefore, the aim of current study was to investigate fatty acid profiles of silage treated with formic acid and LAB strains. This project was supported by Scientific and Technological Research Council of Turkey (TOVAG-213O166).

Cyprinid fish (*Carassius gibelio*) was used for the production of silage. Fish were minced by a grinder and divided into six equal portions. The control group was treated with formic acid, and the rest of the groups were treated with LAB strains isolated from *Sparus aurata*, *Dicentrarchus labrax*, *Mugil cephalus*, *Cyprinus carpio* and *Silurus glanis* muscle, skin and gut. The LAB strains used for the fermentation of silage were *Enterococcus gallinarum*, *Streptococcus spp, Lactobacillus brevis, Lactobacillus plantarum* and *Pediococcus acidilactici*. Lipid of the silage was obtained by using a centrifuge and lipid samples were converted to their constituent fatty acid methyl esters by the method of Ichihara et.al. (1996). The fatty acids methyl esters were separated and quantified with a gas chromatograph.

Among fatty acids, myristic acid, palmitic acid, palmitoleic acid, stearic acid, oleic acid, linoleic acid, eicosapentaenoic acid and docosahexaenoic acid were the predominant fatty acids in all samples. The rates of SFA, MUFA and PUFA in raw fish were 22.88%, 31.77% and 14.40%, whereas their values were 21.87%, 39.68% and 14.57% in silage treated with formic acid, respectively. The ratios of SFA, MUFA and PUFA in silage treated with LAB strains were in the range of 22.18-22.97%, 39.07-42%, 14.45-15.53%, respectively. The results showed that the level of MUFA considerably increased in silage groups, slight increase in PUFA was observed compared to those in raw materials.

Fish oil extracted from fish silage treated with formic acid and LAB strains should be regarded as a healthy diet component for animal or human nutrition because of its high PUFA contents.

Key words: Silage, bacteria, formic acid, fatty acids, PUFA

Effect of red cabbage antioxidant extracts on lipid oxidation of fresh tilapia

Presenting author**:** Ayse Demirbas

Co-authors: Bruce A. Welt, Yavuz Yagiz, Changmou Xu

Gaineville, Florida, USA

Oxidation of polyunsaturated fatty acids (PUFA), eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) in fish causes loss of product quality. Oxidative rancidity causes loss of nutritional value and undesirable color changes. Therefore, powerful antioxidant extracts may provide a relatively low cost and natural means to reduce oxidation, resulting in longer, higher quality and higher value shelf life of foods.

In this study, we measured effects of red cabbage antioxidant on lipid oxidation in fresh tilapia filets using thiobarbituric acid reactive substances (TBARS) assay, peroxide value (PV) and color assesment analysis.

Extraction of red cabbage was performed using an efficient microwave method. Fresh tilapia filets were dipped in or sprayed with solutions containing different concentrations of extract. Samples were stored for up to 9 days at 4°C and analyzed every other day for color and lipid oxidation.

Results showed that treated samples had lower oxidation than controls. Lipid peroxide values on treated samples showed benefits through day-7. Only slight differences were observed between spraying and dipping methods. This work shows that red cabbage antioxidant extracts may represent an inexpensive and all natural method for reducing oxidative spoilage of fresh fish.

Keywords: antioxidant, shelf life, fish, red cabbage, lipid oxidation

Influence of high hydrostatic pressure and lysine addition on surimi gelation with low NaCl content

Presenting author: Deysi Cando[1]

Co-authors: Helena Moreno[1], Javier Borderías[1], Torstein Skåra[2]

[1]Department of Products, Institute of Food Science, Technology and Nutrition (ICTAN-CSIC), Madrid-Spain

[2]Nofima: Norwegian Institute of Food, Fisheries and Aquaculture Research, Stavanger-Norway

Following the dietary recommendations to reduce salt consumption, the reduction of sodium content is a technological challenge considering the important role of salts in the gelation of surimi proteins. For that reason the use of technologies such as high hydrostatic pressure (HHP) and the addition of additives to enhance protein gelation is an option. The objective of this work is to study the suitability of the combination of hydrostatic high pressure and the addition of lysine in order to enhance surimi protein gelation under reduced NaCl content.

To perform the study, two salt levels (0 and 0.3% of NaCl) and addition of lysine (0.1%), and the processing with two pressure levels (0 and 300MPa) were investigated. Analyses performed were: protein denaturation enthalpy by differential scanning calorimetry, water binding capacity (WBC) and mechanical properties as breaking deformation (BD).

Enthalpy values were lower in samples with 0.3% NaCl meaning a higher denaturation of proteins which would indicate an unfolding of protein molecules.

The addition of lysine significantly increased the BD, and WBC. These findings were supported by enthalpy value that decreases with the addition of lysine independently of NaCl content.

The application of high pressure also increased significantly BD and WBC. HHP processing induced protein unfolding, presenting lower enthalpy, which resulted in improvement of the protein network.

The combination of HHP processing and lysine addition, regardless of NaCl concentration,, resulted in an increase in BD and WBC indicating a synergic effect of lysine and HHP processing.

The addition of lysine (0.1 %) when samples are processed under HHP resulted in better gels, when low concentration or no NaCl is added. So that, that way of processing could be an alternative to make low salt content surimi-based-products with heathy properties.

Key words: surimi, NaCl-reduction, protein-gelation, lysine, high pressure

Seasonal and sexual changes of chemical composition in puffer fish (*Lagocephalus sceleratus*) caught from Northeastern Mediterranean

Presenting author: Deniz Ayas[1]

Co-authors: Ali Rıza Kösker[2], Fatih Özogul[2], Yeşim Özogul, Mustafa Durmus[2], Yılmaz Uçar[2]

[1]University of Mersin, Faculty of Fisheries, Department of Seafood processing Technology, Mersin, Turkey

[2]University of Çukurova, Faculty of Fisheries, Department of Seafood processing Technology, Adana, Turkey

Alien marine species migrate into the Mediterranean via the Suez Canal. Some alien species have economic value, although some species are harmful on ecosystem, native species and human health especially in the eastern Mediterranean. Pufferfish is regarded as one of the "worst alien fishes" of the entire Mediterranean Sea. Because most members of the Tetraodontidae are contain Tetrodotoxin (TTX). However, in the Far East, this fish is considered a delicate fish, especially in Japan where it is prepared by experts. In this study, changes of sexual and seasonal of proximate composition, mineral composition and fatty acid profile of puffer fish (*Lagocephalus sceleratus*) were investigated.

The samples used seasonally were caught by bottom trawl, longline and fishing line from Mersin Bay during the period of December 2012 and November 2013.

Nutritional composition analysis showed that the average protein, lipid, moisture and ash content for males were 20.44%, 0.65%, 76.98%, 1.43% and for female individuals were 20.58%, 0.87%, 77.12%, 1.4%, respectively. In terms of fatty acid analysis, 21 different fatty acids have been identified in muscle of puffer fish. Palmitic acid (C16:0), stearic acid (C18:0), linoleic acid (C18:2n6), linolenic acid (C18:3n3), vassenic acid (C18:1 ω 7), arachidonic acid (C20:4ω6), docosadienoic acid (C22:2 cis), eicosapentaenoic acid (C20:5 n3) and docosahexaenoic acid (C20:6n3) were the predominant fatty acids in all samples. The differences in Na, Mg, P, K, Ca, Co, Ni, Cu, Zn, Se and Mo levels of muscle tissue in all seasons were determined as 2729.9-4884.64 µg/g, 1040.0-1477.5 µg/g, 10978.0-20037.0 µg/g, 14593.0-20577.0 µg/g, 196.7-1104.5 µg/g, 0.440.69 µg/g, 1.32-2.44 µg/g, 1.69-2.65 µg/g, 28.55-54.22 µg/g, 2.81-3.98 µg/g and 1.00-2.13 µg/g, respectively. The quantity relationships generally found in the metal levels of muscle in all the species were: K > P > Na > Mg > Ca > Zn > Se > Cu > Ni > Mo > Co.

Our results showed that pufferfish is highly nutritious seafood in terms of nutritional composition and fatty acid levels. However, consumption of pufferfish is dangerous that due to TTX.

Key words: puffer fish, *Lagocephalus sceleratus*, proximate composition, mineral composition, fatty acids

The effects of nanoemulsions based on commercial oils (sunflower, canola, corn, olive, soybean, and hazelnut oils) on the fatty acid compositions of farmed sea bass stored at 2±2 °C

Presenting author: Deniz Ayas[1]

Co-authors: Yesim Ozogul[2], Mustafa Durmus[2], Yılmaz Ucar[2], Ali Rıza Köşker[2], Deniz Ayas[1], Hatice Yazgan[2]

[1]Mersin University, Faculty of Fisheries, Department of Seafood Processing Technology, Mersin, Turkey

[2]Cukurova University, Faculty of Fisheries, Department of Seafood Processing Technology

The term “nanoemulsion” refers to an almost thermodynamically stable isotropically clear dispersion of two immiscible liquids, such as oil and water, stabilized by an interfacial film of surfactant molecules. Nanoemulsions can act as carriers or delivery systems for lipophilic compounds, such as nutraceuticals, drugs, flavors, antioxidants, and antimicrobial agents. The advantages of nanoemulsions involve high physical stability, high bioavailability, and low turbidity, making them attractive systems for application in the food, cosmetics, and pharmaceutical industries. Little work has been done using nanoemulsions in seafood. Therefore, the aim of this work is to investigate the effects of oil-in-water nanoemulsions using different commercial oils (sunflower, canola, corn, olive, soybean, and hazelnut oils) on the fatty acid profile of sea bass (*Dicentrarchus labrax*) fillets stored at 2±2 °C. This project was supported by Scientific and Technological Research Council of Turkey (TÜBİTAK) (TOVAG-113O379).

Nanoemulsions were prepared according to Hamouda *et al.* (1999). Physical properties of nanoemulsions were analysed in terms of viscosity, particle size of droplets, thermodynamic stability, refractive index, and surface tension. Sea bass were obtained from a local fish farm in İzmir, Turkey. Fish were killed by dipping in ice-cold water (hypothermia). After death, the fish were transported to the laboratory in ice within 24 to 25 h from harvesting. They were immediately gutted and divided into seven lots. One lot was stored on plates wrapped with permeable stretch film. The other samples were treated with nanoemulsions. All samples were stored in a chill room (2±2 °C) and analyses were carried out seven times (d 0, 2, 4, 6, 8, 10 and 12). Lipid content was measured by the method of Bligh and Dyer (1959). Lipid samples were converted to their constituent fatty acid methyl esters by the method of Ichihara *et al.* (1996).

Myristic acid (C14:0), palmitic acid (C16:0), palmitoleic acid (C16:1), stearic acid (C18:0), linoleic acid (C18:2n6), linolenic acid (C18:3n3), eicosapentaenoic acid (C20:5 n3) and docosahexaenoic acid (C20:6n3) were the predominant fatty acids in all samples. The results showed that the level of SFA increased whereas MUFA and PUFA decreased with storage time. Generally, the treated samples showed slower loss of fatty acids than those of the untreated samples.

The use of nanoemulsions regardless of oil type reduced the oxidation of fatty acids and all oils used can be recommended for nanoemulsions as a preservative for fish.

Key words: nanoemulsions, *Dicentrarchus labrax*, sea bass, fatty acids, PUFA

The effects of nanoemulsions based on commercial oils on biogenic amine concentration of refrigerated farmed sea bass

Presenting author: Deniz Ayas[1]

Co-authors: Mustafa Durmus[2], Yesim Ozogul[2], Esmeray Kuley Boga [2], Deniz Ayas[1], Fatih Ozogul [2], Yılmaz Ucar[2]

[1]Mersin University, Faculty of Fisheries, Department of Seafood Processing Technology, Mersin, Turkey

[2]Cukurova University, Faculty of Fisheries, Department of Seafood Processing Technology, Adana, Turkey

Biogenic amines are important as agents of food intoxication and indicators of fish spoilage. Biogenic amine formation can be controlled by inhibiting microbial growth or inhibiting the decarboxylase activity of microbes. Nanoemulsions are regarded as self-preserving antimicrobials since the water present in them is effectively bound by its structure and access to the water by microorganisms is restricted. Nanoemulsions have adverse effects on the structure and function on bacteria by destabilizing the organism's lipid envelope. Therefore, the aim of this work is to investigate the effects of oil-in-water nanoemulsions using different commercial oils (sunflower, canola, corn, olive, soybean, and hazelnut oils) on the biogenic amine contents of sea bass (*Dicentrarchus labrax*) fillets stored at 2±2 ^{o}C. This project was supported by Scientific and Technological Research Council of Turkey (TÜBİTAK) (TOVAG-113O379).

Nanoemulsions were prepared according to the method of Hamouda *et al.* (1999). Physical properties of nanoemulsions were analyzed in terms of viscosity, particle size of droplets, thermodynamic stability, refractive index, and surface tension. Sea bass were obtained from a local fish farm in İzmir, Turkey. Fish were killed by dipping in ice-cold water (hypothermia) and transported to the laboratory in ice within 24 to 25 h from harvesting. They were immediately gutted and divided into seven lots. One lot was stored on plates wrapped with permeable stretch film. The other samples were treated with nanoemulsions. All samples were stored at 2±2 °C. Biogenic amines were analyzed using a HPLC method (Ozogul et al., 2002). Benzoyl chloride as a derivatization reagent was used and the derivatization procedure was based on that of Redmond and Tseng (1979).

In this study, histamine was not detected in any sample analyzed until 10 days of storage. As storage time progressed, putresine, cadaverine, spermidine and spermine, seratonine, tyramine, dopamine and agmatine became the dominant amines. The levels of biogenic amines fluctuated during the storage period. Generally, biogenic amine accumulation in the control is higher than the treated samples. There are many factors affecting the formation of biogenic amines such as aquaculture conditions, food, fish species, body composition, and storage and processing conditions and the presence of decarboxylase-active microorganisms and the availability of free amino acids.

The use of nanoemulsions regardless of oil type was found to be effective in inhibiting bacterial growth, hence reducing the biogenic amine formation.

Key words: nanoemulsions, *Dicentrarchus labrax*, sea bass, fatty acids, PUFA

Growth of red halophilic bacteria and quality assessment of heavily salted cod fillets (*Gadus morhua*) – when produced with used salt

Presenting author: Ann Helen Hellevik

Co-authors: Kristine Kvangarsnes, Trygg Barnung

Møreforsking AS, Ålesund, Norway.

When producing salted fish the use of salt reduces water activity and inhibits enzymatic spoilage. Salt also reduce bacterial growth in fish muscle, with the exception of halophilic bacteria. These bacteria come from marine sources (solar salt, sea salt) and survive in salted and dried salted fish. The most likely spoilage organism found in solar salt is the red halophilic bacteria. Even though they attack the muscle, brakes down proteins and make an unpleasant smell. Halophilic bacteria do not create toxins. Red discoloration caused by the bacteria is not accepted and is considered commercially unsuitable.

Large amounts of salt are used in industrial production of salted cod. If salt are reused the result may be lower production costs as well as lower environmental impact. One of the arguments for not reuse salt in the salt fish industry, have been incidence of red halophile bacteria growth. However, studies made in recent years, has shown that this is not the case. The aim of this study was to examine whether growth of red halophilic bacteria are higher in cod fillets salted with reused salt compared to fillets salted with unused salt in pilot scale trials under controlled conditions.

Fillets without skin of thawed cod (*Gadus morhua*) produced on board (400g ±100) were used in all experiments. Three experiments were performed; where unused salt, salt used once and salt used twice of both rock salt and sea salt was used for salting. The experiments have been carried out in controlled conditions in small scale. In order to evaluate quality changes in cod fillets, sensory analysis, determination of instrumental colour, content of water and NaCl, microbiological analysis and statistical calculations was conducted.

Results showed that salting procedures when storing in low temperatures (4-6 °C) did not result in growth of red halophilic bacteria. In addition, salting with reused salt did not lead to lower quality of the final salted product. The study also showed that fillets salted with used salt had higher yield than fillets salted with unused salt. This may be caused by less salt uptake and thereby fillets loose less water and may also be related to the low pressure on fillets during salting, compared to commercial salting procedure.

Results indicate that reuse of salt is suitable in production of salted cod fillets.

Key words: heavily salted, fillets, halophilic bacteria

Spectroscopic differentiation between rehydrated fully salted cod and lightly salted cod

Presenting author: Karsten Heia[1]

Co-authors: Martin Hansen Skjelvareid[1], Rodrigo González Reboredo[2]

[1] Nofima, Tromsø, Norway

[2] Anfaco, Spain

Rehydrated heavily salted cod products (bachalao) are more expensive to produce than lightly salted cod products. False labelling is a problem and there is a need of objective and fast methodology that can differentiate between these products. The scope for the presented work was to test if VIS/NIR spectroscopy can be a reliable candidate for this task.

Two Norwegian commercial companies delivered “industry-produced“-samples. Company 1 provided two groups with heavily salted cod in June 2014, while company 2 delivered two batches with lightly salted cod in November 2014. For both heavily and lightly salted cod, one group was produced from fresh caught cod and the other from frozen/thawed cod. The lightly salted cod fillets were frozen after production and arrived at Nofima as frozen products.

ANFACO-CECOPESCA collected market samples in Spain in February 2015 from five producers of rehydrated heavily salted cod and six producers of lightly salted cod. No other information was available about these samples. DHL transported the frozen samples from Spain to Nofima (Tromsø, Norway).

The only sample preparation performed prior to VIS/NIR measurements was rehydration of the fully salted products and thawing of the frozen products. Spectral data from the samples was extracted using hyperspectral imaging in interactance mode. As spectral pre-processing, Standard Normal Variate transform and first and second derivative was tested. To classify samples, based on spectral data, as heavily salted rehydrated or lightly salted products we applied partial least square regression. 60% of the samples was used for modelling, while the remaining 40% was allocated for testing of models (test set).

Modelling and testing with industrial samples only, the product type was 100% correctly identified (rehydrated heavy salted or lightly salted cod). Including market samples, for training and testing, lower performance occurred. Market samples achieved 94% correct identification while the industrial samples still had 100% correct classification. The reason for this was the low number of market samples available. To improve the model performance a higher number of marked samples needs to be included in the modelling and testing.

Experiments shows that spectroscopy is a good measurement method for differentiation of lightly salted and rehydrated heavily salted cod. 94% of market samples and 100% of industrial samples are correctly identified. Training a robust classifier for market samples will require a wider span of market samples in the training set.

Key words: spectroscopy, differentiate, heavily/lightly salted cod

Biotechnological production of omega-3 fatty acids from microalgae

Presenting author: Osman Kadir Topuz

Co-authors: Tuğçe Aygün

Department of Seafood Processing Technology, Faculty of Fisheries, Akdeniz University, Antalya, Turkey

Consumption of fish oil containing Omega-3 fatty acids (docosahexaenoic acid, DHA; eicosapentaenoic acid, EPA; and α-linolenic acid, ALA) has been steadily increasing for decades due to their health benefits, such as reducing the risk of cardiovascular diseases, type II diabetes, ocular diseases, arthritis, etc. Microalgal oil might be the most promising alternative to fish oil, since they are the primary producers of Omega-3 fatty acids. They can be cultured either photoautotrophically or heterotrophically, with each system having its advantages and disadvantages. Bacillariophyceae and Chrysophyceae species are rich sources of EPA and DHA fatty acids; Cryptophyceae, Prasinophyceae, Rhodophyceae, Xanthophyceae, Glaucophyceae and Eustigmatophyceae can represent EPA sources, whereas DHA is found in significant amounts mostly in Dinophyceae, Prymnesiophyceae, and Euglenophyceae. The development of biotechnological processes for microalgal oil production, particularly oils rich in DHA and EPA, has benefited from the fact that a number of organisms can accumulate high lipid contents in biomass up to 50% biomass dry weight, including 30%–70% of this fatty acid.

Nannochloropsis oculata Hibberd is a phototrophic algal species and present elevated levels of EPA in total fatty acids, although relatively low cell lipid contents tend to result in small EPA amounts in the biomass. *Schizochytrium* sp. and *Crypthecodinium cohnii* have been used in the commercial production of DHA-rich oils, particularly for inclusion in infant formulas due to its low EPA content, which in high levels can induce bleeding in both infants and nursing mothers. *Ulkenia* sp. another thraustochytrid, is currently used as a commercial source of DHA-rich oil, which may contain up to 50% DHA. *P. lutheri*, a marine microalga commonly used in aquaculture, is a well-known source of EPA and DHA, reaching up to 53% of its total fatty acid under specific conditions. Overall, uses of microalgae for the commercial production of Omega-3 fatty acids rich algal oils requires advances in scientific studies to enhance growth performance and lipid deposition in addition to lowering of costs involved in biomass production and harvesting.

Key words: omega-3 fatty acids, microalgae, DHA, EPA, algal oil

Effect of frozen storage temperature prior to different heat treatments on physicochemical properties of Atlantic mackerel *(Scomber scombrus)*

Presenting author: Magnea G. Karlsdottir[1]

Co-authors: Paulina Elzbieta Wasik[1,2], Telma B. Kristinsdóttir[1,2], Sigurjón Arason[1,2], Ásbjörn Jónsson[1], Hordur G. Kristinsson[1,3]

[1]Matis ohf, Icelandic Food and Biotech R&D, Vinlandsleid 12, IS-113 Reykjavík, Iceland

[2]University of Iceland, Department of Food Science, Vinlandsleid 12, IS-113 Reykjavík, Iceland

[3]Department of Food Science and Human Nutrition, University of Florida, 359 FSHN Building, Newell Drive, Gainesville, FL 32611, USA

Atlantic Mackerel is relatively novel specie in Iceland. Since 2007, mackerel has been caught in Icelandic waters and its abundance increases steadily along with its importance. For instance, exportation values of frozen mackerel from Iceland reached 21 billion ISK in 2013. Mackerel is a fatty fish and a great source of omega-3 polyunsaturated fatty acids (PUFA) which makes it an excellent choice of food for human consumption. However, due to the high amount of PUFA the mackerel has short shelf-life and therefore requires appropriate storage and processing conditions.

Physicochemical changes of mackerel during frozen storage (6, 9, 12 months) at -18 °C vs. -25 °C were analysed. Different heat treatments (75 °C, 90 °C) and brining were applied. Measurements of heating yield, texture, liquid holding capacity (LHC), lipid and water content, primary and secondary oxidation products as well as lipid hydrolysis were analysed. Prolonged storage showed negative effects on the raw material prior to processing due to increased level of lipid oxidation, where fish stored at -18 °C was of significantly poorer quality than fish stored at -25 °C. Moreover, the results indicated that heat treatment at 75 °C, in contrary to heat treatment at 90 °C showed higher water content, LHC, heating yield as well as lower maximum shear force of texture. In conclusion, it is important to apply proper heat treatment in order to meet consumers and market demands, as well as proper time of exposure to heat treatment. Furthermore, mackerel caught in Icelandic water has potential to be utilized for the production of value-added products.

Key words: pelagic fish, heat treatment, frozen storage temperature, lipid oxidation, physicochemical properties

Physicochemical changes of frozen herring – Effect of holding time before freezing and storage temperature

Presenting author: Magnea Karlsdottir[1]

Co-authors: Dandan Ren[3], Sigurjón Arason[1,2], Dang Thi Thu Huong[2]

[1]Matis ohf, Icelandic Food and Biotech R&D, Vinlandsleid 12, IS-113 Reykjavík, Iceland

[2]University of Iceland, Department of Food Science, Vinlandsleid 12, IS-113 Reykjavík, Iceland

[3] Dalian Ocean University, Heishijiao Street 52, Dalian, 116023, China

Freezing is one of the most common procedures applied to preserve physicochemical properties and prolong shelf life of fish. The main purpose of freezing is to prevent or slow down bacterial spoilage, enzyme activity and oxidation reactions. Sensory, chemical and physical changes continue to some extent during frozen storage. Undesirable changes associated with lipids and proteins are the main reasons for reduced fish quality during frozen storage. Quality changes of frozen fish during storage can be influenced by many factors including fish species, handling on board, temperature and storage time before freezing, freezing rate, frozen storage temperature and temperature fluctuations. Optimal handling and transport conditions can be used to ensure high quality of fish products on the market. However, temperature fluctuations through the production and distribution chain could affect the fish quality and safety. These fluctuations mainly occur during handover from one party/function to the next in the logistics.

The objective of this study was to investigate the effects of different frozen storage temperature (-25 °C and fluctuant -12 °C) on lipid degradation and protein conformation changes of frozen herring. Moreover, physicochemical changes of herring frozen at different post-mortem period (frozen on board and frozen on land) were compared. The effect of the physicochemical parameters were studied by measuring liquid holding capacity (LHC), drip loss, peroxide value (PV), thiobarbituric reactive substances (TBARS), free fatty acids (FFA), lipid content, water content, pH value, disulfide bonds content, available and total SH groups.

The results showed that lower and stable storage temperature (-25 °C) can effectively reduce drip loss, especially inhibit lipid degradation, including PV, TBARS and FFA, compared with fluctuant storage conditions at -12 °C. Furthermore, freezing herring on-board proved to be more beneficial compared to when frozen at land. Herring frozen on board was frozen in pre-rigor state, resulting in more moderate pH change than of herring frozen at land. Freezing herring during pre-rigor period can slow down lipid oxidation including PV and TBARS, and reduce conformational changes of protein caused by freezing process compared with post-rigor frozen herring.

Key words: herring; freezing; temperature fluctuation; physicochemical properties; fatty fish

Improved storage stability of fresh redfish fillets

Presenting author: Magnea Karlsdottir[1]

Co-authors: Steinunn Jónsdottir[2], Sigurjón Arason[1,2]

[1]Matis ohf, Icelandic Food and Biotech R&D, Vinlandsleid 12, IS-113 Reykjavík, Iceland

[2]University of Iceland, Department of Food Science, Vinlandsleid 12, IS-113 Reykjavík, Iceland

Today, the export of fresh redfish fillets from Iceland to European markets is mainly by airfreight. Due to the relative short shelf life of fresh fish, exporting the fish by airfreight ensures a fast delivery to the consumer. Exporting the fresh redfish fillets by sea freight rather than airfreight is both environmentally and economically beneficial. However, sea freight has not been found feasible due to the short shelf life of fresh fish. Therefore, it is of great importance to the Icelandic fishing industry to find ways to improve the shelf life of fresh redfish fillets.

The overall objective of present project was to study how different packaging methods post filleting affect the rate of lipid oxidation, microbiological spoilage, and therefore the storage life of fresh redfish fillets. Three different packaging methods were compared, including the traditional method of air packaging using EPS boxes with the addition of ice, modified atmosphere packaging (MAP) using plastic boxes with and without addition of CO_2 emitting pads. MAP was effective in reducing lipid oxidation to some degree. Microbiological observations revealed no difference between the Air group and the two MAP groups in terms of total viable count, and the growth of *Photobacterium phosphoreum*. However, MAP was beneficial in reducing the number of *Pseudomonads*. The benefits of adding CO_2 pads to the gas packed fish were not significant with respect to lipid oxidation and microbial growth. Moreover, the colour of the fish was not affected by the MAP packaging. MAP fish had, as expected, lower pH than air packed fish but increased water loss during storage was observed for the MAP fish. The MAP fish maintained the freshness period of the redfish fillets for one additional day compared to the air packed fish, but the shelf life of the two MAP groups was determined to be about 12 days.

Key words: fresh fish; *Sebastes marinus*; modified atmosphere packaging (MAP); storage life; sensory

Improved microbial quality and safety of fish

Presenting author: Françoise Leroi [1]

Co-authors: Bruno Le Fur[2], Hélène L. Lauzon[3], Eyjólfur Reynisson[4], Turid Rustad[5], Jens Rohloff[5], Morten Sivertsvik6, Evelyne Fenart[7], Marie-France Pilet[8], Ómar Grétarsson[9], Hannes Garðarsson[10],Þorvaldur Þóroddsson[11]

[1]Laboratory EM3B, Ifremer, BP 21105, 44311 Nantes Cedex 03, France, [2]PFI Nouvelles Vagues, [3]Primex ehf, [4]Matis, [5]NTNU, [6]Nofima, [7]CITPPM, [8]Oniris, [9]Fjardalax ehf, [10]Rafeyri ehf,[11]Samherji hf

The aim of this poster is to present the objective of a European project SAFEFISHDISH (2015-2018) funded by the ERANET COFASP.

SAFEFISHDISH is a 3-year project coordinated by Dr Françoise Leroi, (Ifremer, France). Eleven partners from 3 countries are involved (France, Iceland and Norway):

- 5 Academics (NTNU, NOFIMA, MATIS, Ifremer, ONIRIS)
- 5 Industrials (Primex, Rafeyri, Fjardalax, Samherji, PFI Nouvelles Vagues)
- 1 Confederation of industry (CITPPM)

Norway and Iceland are among the world's leading seafood nations and this has been achieved by increased value adding and knowledge-based management of resources. France is an important producer but also the major European fish importing country. The main challenge remains though in successfully maintaining freshness, quality and value as well as safety of seafood through handling, processing and distribution. Seafood deterioration is mostly governed by microbial and biochemical activities which are influenced by temperature and storage conditions. The main cause of bacterial contamination of fish processing line is due to rapid bacterial proliferation on the skin during early storage which spreads during filleting and by post-contamination during processing. Reducing the microbiota before process and preventing its development during storage will extend shelf life.

The main objective of the SAFEFISHDISH project is to improve the microbial and sensory quality and safety of fish from harvest to consumer. The project will focus on farmed salmon and wild cod, which are the major species traded in Europe. Novel handling techniques and combination of innovative preservation technologies involving biopreservatives (protective cultures and chitosan), superchilling and modified atmosphere will be evaluated. Treatment well ahead of the food chain (on the skin upon harvest and on flesh just after filleting) may maximize its efficiency and will be explored. Combination of these preservation techniques is innovative and needs to be tested. Bacterial ecosystem and their metabolism profile will be explored via modern tools such as new generation sequencing (NGS) and various chromatographic methods.

Developed innovative handling and processing technologies will better control safety and deterioration of valuable seafood and, simultaneously, contribute to nutritional quality and consumer health as well as increased sales return and competitiveness of European seafood.

Key words: hurdle technology, cod, salmon, chitosan, biopreservation

Fatty acid profile in ready-to-eat white meat of edible crab (*Cancer pagurus*) as affected by cooking method and refrigerated storage

Presenting author: Federico Lian

Co-authors: Sabine Harrison, Santiago Condón-Abanto, Paul Whyte, James G. Lyng, Nigel P. Brunton

UCD Institute of Food and Health, University College Dublin, Belfield, Dublin 4, Ireland

Edible crab (*Cancer pagurus*) is a crustacean species distributed along the west coast of Europe. The white meat, picked from the claws of this decapod, is widely consumed worldwide given its outstanding sensory and nutritional properties. To add value to the product, the crabs can be processed and marketed under refrigeration in a ready-to-eat form. Because of microbial safety concerns, it is common practice in the Irish crab industry to conduct a two-step heat treatment which is composed of a cooking process in boiling water and a severe in-pack pasteurisation. These thermal treatments may affect thermolabile chemical compounds present in the meat, such as fatty acids, leading to changes in nutritional value and sensory attributes. Conducting the cooking process in a single-step and at milder temperatures could help retain higher product quality. The aim of this study was to evaluate the effect of different thermal treatments, including ultrasound-aided cooking, on the fatty acid profile of ready-to-eat white crab meat both after processing and during refrigerated storage.

Raw and cooked male exemplars of edible crab caught in the Irish Sea were used in this experiment. Cooked crabs were processed according to three different thermal treatments, that is (i) at 97°C for 25min, (ii) at 75°C for 45min, and (iii) at 75°C for 45min but in an ultrasound-aided process (ratio power/crab weight of approximately 180 $W \cdot kg^{-1}$). The white meat, picked from the claws, was analysed for fatty acid profile by gas-chromatography at the beginning and after 30 days of refrigerated storage in vacuum pouches.

Preliminary results indicated that, in all samples, palmitic, oleic and eicosapentaenoic acid (EPA) were, respectively, the most abundant saturated (S), monounsaturated (MU) and polyunsaturated (PU) fatty acids. The thermal processing affected the fatty acid profile of the meat, with higher MU/S and PU/S ratios present in samples cooked in mild conditions and with the aid of ultrasound. Similar results were observed for the total amount of omega-6 fatty acids whereas the ratio between EPA and docosahexaenoic (DHA) acid did not vary with respect to the cooking temperature. Little change in the fatty acid profile was observed during the shelf-life.

Cooking process had an impact on the fatty acid profile with possible implications on the nutritional value of the product. This study could represent the starting point to develop specific models to better evaluate the effect of ultrasound and cooking parameters on the fatty acid profile of crab meat.

Key words: edible crab, fatty acid profile, omega-3, ultrasound, shelf-life

Characterization of whiting (*Merlangius merlangus*) fillets stored under modified atmosphere packaging by instrumental and front face fluorescence spectroscopy techniques

Presenting author: Abdo Hassoun

Co-authors: Romdhane Karoui

Univ. Artois, EA 7394, Institut Charles Viollette, Lens, F-62307, France

In recent years, there has been an increasing demand for human consumption of fish and other fishery products, thanks to their richness in protein, long chain polyunsaturated *n-3* fatty acids, vitamins, and minerals. However such products are highly fragile, exceptionally perishable and susceptible to spoilage. Therefore, care must be taken to minimize these negative impacts during handling, processing, transporting, packaging, and storage. Modified atmosphere packaging (MAP) has been demonstrated to be beneficial in preventing deterioration and extending the shelf life of fish and seafoods. The objective of the present study was to determine the impact of two MAP conditions on the shelf life of whiting fillets stored at 4 °C by monitoring physico-chemical, color, texture and spectral during storage.

Fresh whiting fillets were divided into three batches: one batch was packaged in normal air and considered as control, and the two others were packaged in 50% N_2⁄50% CO_2 for MAP1 and 80% N_2⁄20% CO_2 for MAP2. Physico-chemical, texture, color and spectral measurements were determined on 1, 3, 6, 8, 13, and 15 days of cold storage.

For a considered storage time, the highest pH value was observed for control group, while the lowest pH, TBARS and TVB-N values were noted for MAP1. The average hardness value of fresh whiting fillets was of 455 g ±18 and decreased ($p<0.05$) during the whole storage time regardless of storage conditions. Similar trend was observed for gumminess and chewiness.

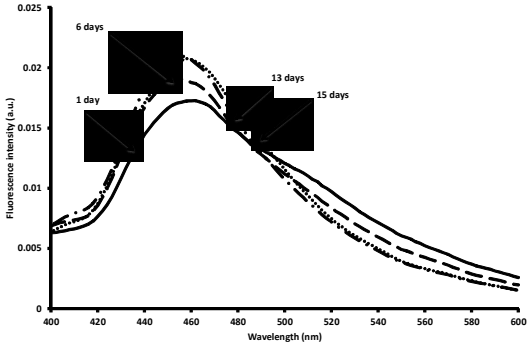


Figure 1: Normalized emission fluorescence spectra (excitation: 360 nm, emission 400–600 nm) recorded on fresh fillets (1 day) (▬) and those stored at 4 °C for 6 (----), 13 (−·−), and 15 (····) days for MAP1 samples

The emission spectra acquired at 360 nm exhibited a maximum located around 460 nm, which could be attributed to the formation of Schiff base originated from reactions between carbonyls, generated by lipid oxidation, and the amino groups of amino acids (Figure 1). A high correlation (R=0.89) was obtained between fluorescence intensity at 460 nm and TVB-N content during the whole storage period.

MAP is widely used for extending the shelf life of fish and seafoods. In the present study, the use of modified atmosphere composed of 50% CO_2 and 50% N_2 (MAP1) induced a positive impact on fish quality. Indeed, better texture of treated samples was observed compared to the control one.

Key words: Fish freshness, modified atmosphere packaging, fluorescence, texture

Comparison of validated methods for the determination of ester bound 2,-3-MCPD and esterified glycidol in fishery products

Presenting author: Sybille Merkle[1]

Co-authors: Horst Karl[2], Jan, Kuhlmann[3], Jan, Fritsche[4]

[1]University of Applied Sciences Hamburg, Hamburg, Germany

[2]Max Rubner-Institut, Department of Safety and Quality of Milk and Fish products, Hamburg, Germany

[3]SGS Germany GmbH – Weidenbaumsweg 137 – 21035 Hamburg, Germany

[4]University of Applied Sciences Hamburg, Hamburg, Germany

2- and 3-Monochloropropanediol (2-/3-MCPD) are food processing contaminants that might be formed by heating foods containing a chloride source. One opportunity is the formation during the wood smoking of fish and meat [1]. Investigations have shown that 3-MCPD in refined oils and fats also occur as mono- or di-esters of fatty acids [2,3]. Further identified compounds in the same matrices are glycidyl esters and 2-monochloropropane-1,3-diol (2-MCPD) esters [4]. The IARC has defined 3-MCPD as a "possible human carcinogen (group 2B)" while glycidol has been classified as "probably carcinogenic to humans (group 2A)".
By now three analytical procedures for the determination of ester bound 2-, 3-MCPD and glycidyl esters in edible oils have been validated by DGF and AOCS ("Unilever method": AOCS method Cd 29a-13, "3-in-1-method": AOCS method Cd 29b-13 and DGF method C-VI 18 (10): AOCS method Cd 29c-13). They provided true and comparable results in investigation of refined edible oils and fats.
In the presented study AOCS methods Cd 29b-13 and Cd 29c-13 were modified and validated "in house" in order to quantify ester bound 2- and 3-MCPD and glycidyl esters in fishery products [5]. A comparison with a method latest published by EFSA (2015) for determining ester bound 2-, 3-MCPD and glycidyl esters in foods is shown [6].
Results of the validation and comparison of the different methods will be presented.

[1] Kuntzer, J., Weißhaar, R., The smoking process - a potent source of 3-chloropropane-1,2-diol (3-MCPD) in meat products. *DLR*. 2006, *102*, 397-400.

[2] Zelinkova, Z., Svejkovska, B., Velisek, J., Dolezal, M., Fatty acid esters of 3-chloropropane-1,2-diol in edible oils. *Food Addit. Contam.*. 2006, *23*, 1290-1298.

[3] Weisshaar, R., Perz, R., Fatty acid esters of glycidol in refined fats and oils. *Eur. J. Lipid. Sci. Technol.* 2010, *112*, 158-165.

[4] Kuhlmann, J., Determination of bound 2,3-epoxy-1-propanol (glycidol) and bound monochloropropanediol (MCPD) in refined oils. *Eur. J. Lipid Sci. and Technol*. 2011, *113*, 335-344.

[5] Karl, H., Merkle, S., Kuhlmann, J., Fritsche, J. Development of analytical methods for the determination of free and ester bound 2-, 3-MCPD and esterified glycidol in fishery products. *Eur. J. Lipid Sci. Technol.* 2015, *117*.

[6] EFSA (European Food Safety Authority): Development and validation of analytical methods for the analysis of 3-MCPD (both in free and ester form) and glycidyl esters in various food matrices and performance of an ad-hoc survey on specific food groups in support to a scientific opinion on comprehensive risk assessment on the presence of 3-MCPD and glycidyl esters in food. 2015, EFSA supporting publication 2015: EN-779.

Key words: ester bound 2-MCPD, ester bound 3-MCPD, esterified glycidol, fishery products, validated analytical methods