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Assessing the risk of booster biocides for the marine environment: a case study at the Belgian Part of the North Sea

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Abstract

The biofouling of submerged surfaces such as ship hulls is often prevented by using anti-fouling components in combination with booster biocides. These booster biocides enter the water column and may affect non-target organisms. Although different negative effects have been associated with the use of booster biocides, their effects on non-target organisms are often unknown. So far, the environmental risks for booster biocides have barely been quantified in the North Sea. In this work, the concentration of five commonly used booster biocides as well as tributyltin has been monitored at five dredged spoil disposal sites in the Belgian part of the North Sea and the harbour and ports of Nieuwpoort, Oostende, and Zeebrugge. Hotspots were discovered where the concentration of one or more booster biocides exceeded the predicted no-effect concentration. Tributyltin has been banned since 2008, but concentrations of 237- to 546-fold of the predicted no-effect concentration were detected in the harbours and ports. Moreover, TBT has been detected in the same order of magnitude in other sea basins, emphasizing the need to monitor the trends and impact of booster biocides and TBT in environmental monitoring programs.

Keywords: Anti-fouling**;** Booster biocides; North Sea; Risk assessment

1 Introduction

Biofouling is the accumulation of microorganisms, algae, and small animals on artificial submerged surfaces (Yebra, Kiil, & Dam-johansen, 2004). Biofouling on vessels such as ships and smaller boats can disrupt the aerodynamic flow, resulting in increased fuel consumption (Schultz et al., 2011; Telegdi, Trif, & Romaīnszki, 2015). In the 1960s, biofouling was avoided by covering vessels with paint containing lead, arsenic, or copper. Besides these metal-based paints, tributyltin (TBT) proved to be the superior anti-fouling component (Almeida, Diamantino, & de Sousa, 2007). Unfortunately, TBT has a detrimental effect on the marine environment (Yebra, Kiil, & Dam-johansen, 2004), interfering with the hormonal balance of oysters (Alzieu et al., 1981), causing imposex in gastropods (Evans, Leksono, & McKinnell, 1995; Yebra, Kiil, & Dam-johansen, 2004) and growth and reproduction failures in *Crangon crangon* populations (Parmentier et al., 2019; Verhaegen et al., 2011). Therefore, TBT has been restricted since 1990 and completely banned in 2008 (European Union, 2003; MEPC Resolution, 2011).

Alternatives have been proposed for the use of TBT (Alzieu, 2000). These anti-fouling components often contain copper and are combined with booster biocides to avoid fouling of copper-resistant microorganisms (Guardiola et al., 2012; Voulvoulis, Scrimshaw, & Lester, 1990). In past decades, booster biocides such as Irgarol, Sea-Nine 211, Diuron, and Dichlofluanid have been authorized and have been widely applied (Konstantinou & Albanis, 2004). In 2006, ECHA (2006) published a manual to harmonize the authorisation of new substances such as booster biocides on EU-level. Since booster biocides leach from boat hulls into the marine environment, they may potentially affect non-target organisms (Arai et al., 2009; Thomas & Brooks, 2010). For example, Irgarol and Diuron inhibit algae photosynthesis by impairing the electron transport in chloroplasts (Arrhenius et al., 2006). Moreover, both components are persistent in the marine environment where Irgarol can be embryotoxic to echinoderms and tunicates, and Diuron may show genotoxicity and immunotoxicity in oysters (Amara et al., 2018; Barranger et al., 2014). Due to the detrimental effects of Diuron and Irgarol, both substances were banned in 2002 by the UK Health and Safety Executive (HSE) (Advisory Committee on Pesticides, 2000). The use of Irgarol was still allowed on vessels longer than 25 m (Chesworth, Donkin, & Brown, 2004). A second ban on Diuron followed in 2008 in the Netherlands and Denmark (Price & Readman, 2013), whereas Irgarol was completely banned in 2021 (EPA US, 2021). Sea-Nine is a broad-spectrum booster biocide and an endocrine disruptor, impeding the fouling of different organisms such as bacterial slime, diatoms, tubeworms, and algae (Voulvoulis, 2006). Sea-Nine was considered to have a low impact on non-target organisms due to its rapid degradation in water. Nevertheless, different toxic effects on non-target organisms have been reported for Sea-Nine. It may disrupt the cytoskeleton in tunicates and induce toxic effects during embryogenesis and larval growth (Bellas, 2006; Cima, Bragadin, & Ballarin, 2008; Wang et al., 2011). Two other booster biocides, Dichlofluanid and Tolylfluanid, suppress the proliferative activity of cells (Holovská, Pistl, & Kovalkovičová, 2007). In water, both components rapidly degrade to dimercaptosuccinic acid (DMSA) and 1-(dimethylsulfamoylamino)-4-methylbenzene (DMST), respectively (Cai et al., 2021). These hydrolysis products are persistent and can harm the marine environment when their concentration exceeds 19 µg L^{-1} and 14 µg L^{-1}, respectively (ECHA, 2014a, 2016). Since knowledge on the toxicity of most booster biocides for non-target organisms is still limited (Maraldo & Dahllöf, 2004; Moon et al., 2019; Terlizze et al., 2001), monitoring the concentrations and effects of booster biocides remains important to acquire knowledge about their potential adverse effects on the marine environment.

Being a priority chemical (Directive 2008/105/EC), the concentration of TBT has been monitored for a long time, whereas quantified concentrations of booster biocides remain scarce. Since the ban on TBT, and considering its estimated half-life of 1.85 years (in sediment) (De Mora, King, & Miller, 1989), its concentration in the marine environment has been reduced (Verhaegen et al., 2012). The environmental quality standard (EQS) for TBT was proposed at 1.6 ng kg^{-1} dry weight (5% organic carbon) in the ICES (International Council for the Exploration of the Sea) regions (Sahlin & Ågerstrand, 2018). As the concentration of TBT decreased, often below the detection limit of routine monitoring methods, most countries have stopped monitoring this component (OSPAR, 2017). However, in 2020, the concentration of TBT still ranged between 0.2 ng g^{-1} and 3 ng g^{-1} in the Belgium part of the North Sea and between 1 ng g^{-1} and 4.6 ng g^{-1} in the Dutch part of the North Sea. The last TBT values that were reported in the UK dates back to 2016. Concentrations ranging between 5 ng g^{-1} and 6 ng g^{-1} were then reported (ICES, 2022). This indicates that even for banned substances such as TBT, monitoring should remain a priority. Especially as HELCOM (2023) reported that the TBT concentration was still above the threshold to achieve a good and safe marine environment in most parts of the HELCOM sea basin. Additionally, TBT occurrences as well as cases of imposex were still detected in marine protected areas (Castro et al., 2021). According to the Marine Strategy Framework Directive (MSFD, 2008/56/EC) report of 2018, the concentration of TBT in the Belgian part of the North Sea

(BPNS) is alarming (Tornero, Boschetti, & Hanke, 2018) and concentrations up to 12.8 ng g^{-1} and 17.8 ng g^{-1} have been detected in fish (*Platichthys flesus*) and mussels (*Mytilus edulis*), respectively (ICES, 2022). According to Thomas, Blake and Waldock (2000), the concentrations of TBT and other booster biocides in the sediment are often associated with the presence of anti-fouling paint chippings. In that study, the concentration of Diuron (<0.1 µg g^{-1}) and Irgarol (<0.001 µg g^{-1}) were often below the LOQ (Thomas, Blake, & Waldock, 2000). Nevertheless, much higher concentrations have been measured. In a study in 2016, Diuron and Irgarol 1051 were detected in Panama with a concentration of <0.3 to 5.0 ng L^{-1} and <2.7 to 70 ng L^{-1}, whereas dichlofluanid was not detected (Batista-Andrade et al., 2016). In 2021, Irgarol was detected in 75% of the samples in the ports of Brazil, with a mean concentration between 1.5 and 2.2 ng g^{-1} (Soares et al., 2021). Apart from some ad hoc studies in marine waters and sediments in the Mediterranean Sea in Spain (García et al., 2020; Köck-Schulmeyer et al., 2019), the Baltic Sea in Estonia (ICES, 2022) and the Baltic Sea in Sweden (ICES, 2022), no recent data are available about booster biocides other than TBT in European Seas. For the BPNS, no quantified results have been reported so far.

In this paper, the concentrations of five commonly used booster biocides (Dichlofluanid, Diuron, Irgarol, Sea-Nine, and Tolylfluanid) as well as Tributyltin (TBT), have been measured in sediment samples from five dredged spoil disposal sites and from six reference locations in the BPNS over a period of three consecutive years (2018-2020). In this study, we aim to link the contamination of booster biocides at dredge disposal sites with their respective port or harbour and assessing the risk on the environment by calculating the risk characterization ratio. Although it may also be of added value to measure primary degradation products of each booster biocide such as DMSA, DMST, etc., this study mainly focus on the booster biocides themselves. Only for TBT, concentrations of two degradation compounds (dibutyltin (DBT) and monobutyltin (MBT)) are reported. The concentration of booster biocides and TBT has also been measured at the harbour and ports of Nieuwpoort, Oostende and Zeebrugge in 2020. The Predicted no-effect concentration (PNEC) is established in water, but we calculated values in sediment for all booster biocides and evaluated the risk by comparing environmental concentrations with these PNEC values.

2 Material and Methods

2.1 Sampling

In the BPNS, five dredged spoil disposal sites are defined: LNP, Br&WOO, Br&WZE, Br&WS1, and Br&WS2 (Figure 1). In 2020, more than 12 million tonnes dry matter of dredged material (tonnes DM) were disposed over the five sites. LNP is located close to the harbour of Nieuwpoort and is characterised by a sandy underground and a low dumping volume of 230 000 tonnes DM. Br&WOO and Br&WZE are located close to the ports of Oostende and Zeebrugge, respectively. Both disposal sites are characterised by a muddy underground and a dumping volume of 740 000 and 3.7 million tonnes DM respectively. Br&WS1 and Br&WS2 are located a bit more offshore from the port of Zeebrugge and are characterised by a sandy underground and a dumping volume of 6 and 1.5 million tonnes DM, respectively (Lauwaert et al., 2021).

Next to the sampling locations within the five dredged spoil disposal sites, six reference locations (120, 230, 330, 140bis, ZVL, B041) are sampled to assess the overall contamination in the BPNS (Figure 1). Reference locations are characterised by a similar environment (grain size, benthic community, etc.) as observed at one or more disposal sites, allowing to assess the impact of dredging (and dredged spoil disposal) on the marine environment. Sampling locations were surveyed in September/October of 2018 to 2020. Exact sampling coordinates for each location are available in Table S2. At each location, one sediment sample was collected using a Van Veen grab with a 0.1 m^2 surface with a total

mass of about 10 to 20 kg. A random subsample of 20 g from each grab was immediately deep-frozen at -20 °C on board of the RV Belgica. Before analysis, samples were freeze-dried.

Additional sediment samples were collected at several locations in the harbour and ports of Nieuwpoort, Oostende, and Zeebrugge in March 2020, by using a small Van Veen grab (0.03 m^2 surface) with a total mass of about 3 to 6 kg from the rigid inflatable boat Zeekat. Exact sampling coordinates for each location are available in Table S3. Subsamples of 20 g from each grab were taken to the lab, freeze-dried and stored in the dark at room temperature until further analysis.

2.2 Extraction and analysis of booster biocides

Chemical detection, identification, and quantification was done by extracting freeze-dried sediment samples by pressurised liquid extraction using an ASE350 (Dionex). Stainless steel extraction cells of 22 mL were filled with 4 g of sediment sample, 20 µL of recovery standard (RS) solution (2.5 µg mL^{-1} atrazine d5 and 2.5 µg mL^{-1} Diuron D6 in acetonitrile) and diatomaceous earth (Celite 545, Sigma Aldrich). For each sample, a non-spiked and spiked subsample was analysed and booster biocides were quantified by applying standard addition to correct for matrix effects. Spiked subsamples were made by adding 20 µl solution containing 1 µg mL^{-1} of Diuron, Irgarol, Sea-Nine, Tolylfluanid, and Dichlofluanid in acetonitrile to the sediment subsample. Extraction was performed at a pressure between 10.3 MPa and 11.7 Mpa using hexane:acetone (3:1 v:v) as solvent. Three extraction cycles were run at 100 °C, 5 min. static time, 5 min. heat time and 60% rinse volume (total volume of solvent added after all static cycles, expressed as a percentage of the empty extraction cell volume). The extract was purged out of the cell by a stream of nitrogen gas for 60 s and evaporation to 1 mL by a Turbovap II evaporator (Zymark), 3 mL of isopropanol were added and evaporated to 1 mL. This extract was analysed by liquid chromatography-tandem mass spectrometry (LC-MS, Nexera 8400, Shimadzu) with electrospray ionisation in multiple reaction monitoring modes for the analysis of Diuron, Irgarol, and Sea-Nine. After injection, the extract was evaporated to dryness and redissolved in 1 mL of water:acetonitrile (50:50) for injection on the LC-MS with atmospheric pressure ionisation for the analysis of Dichlofluanid and Tolylfluanid. For Diuron, Irgarol, and Sea-Nine 211, separation was done on a kinetix C18 column (1.7 µm, 10 nm, 150x2.1 mm) at 40 °C at a flow rate of 3 mL min^{-1} with methanol as solvent A and 10 mM ammonium acetate in water as solvent B. For Dichlofluanid and Tolylfluanid determination, a Kinetix EVO C18 column (1.7 µm, 100 µm, 100x2.1 mm) was applied at 40 °C at a flow rate of 0.2 mL min^{-1} with acetonitrile as solvent A and water (0.1% formic acid) as solvent B. For both methods, an identical gradient programme was run, starting with 5% solvent A for 2 min, up to 95% solvent A after 8 minutes which was held for 4 min. The mobile phase went back to starting conditions (5% A) in 6 sec, which was held for 5 min.

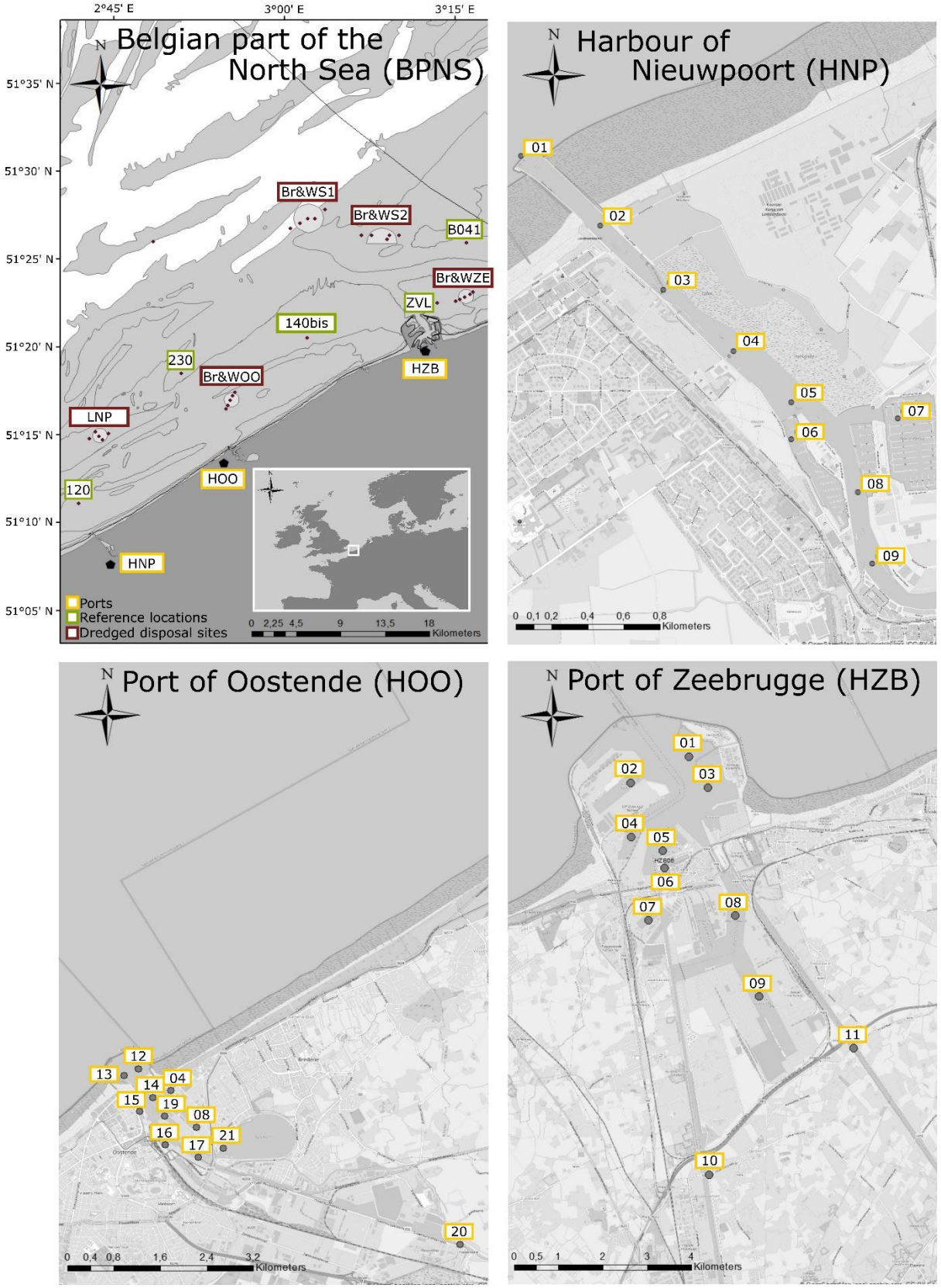

Figure 1: Overview of the sampling locations within the five dredged spoil disposal sites (LNP, Br&WOO, Br&WZE, Br&WS2, Br&WS1) and reference locations (120, 230, 140bis, ZVL, B041) in the Belgian part of the North Sea, and in the harbour and ports of Nieuwpoort (HNP), Oostende (HOO), and Zeebrugge (HZB)..

2.3 Extraction and analysis of TBT

Tributyltin in sediment is determined starting from 1 g of freeze-dried sediment to which 15 mL of methanol, Internal standards (deuterated TBT(D_{27})Et, deuterated MBT(D9)Et_3, $SnPr_4$, $SnPr_3Cl$), 4.5 mL of acetate buffer and 7 mL of hexane was added. After adjusting the pH to 4-5, the sample was ethylated with 2 mL 5% $NaBEt_4$ (dissolved in ultrapure water) under intense stirring for 5 minutes. Subsequently, 4.5 mL of 1M NaOH was added, the mixture was stirred for 5 minutes, checked if the pH >12, and centrifugated. The hexane layer was recovered, the sample was washed with an additional 3 mL of hexane, and the combined hexane layers were concentrated to 1 mL. The extract was filtered over a Pall Acrodisc 13 mm Minispike with 0.2 µm GHP membrane, the filter was rinsed with an additional 1 mL of hexane and the extract underwent a Gel Permeation Chromatography (Shimadzu combo of LC-20AT, SIL-20AHT, FRC-10A, SPD-20A and CBM-20A) clean-up where a solvent change to dichloromethane took place. 100 µl of iso-octane was added, the eluate concentrated to 1 mL, and 5 µl analysed on a Thermo Scientific Trace 1300 GC coupled to a TSQ 8000 EVO Triple Quad equipped with PTV injection using He in all gas streams. The PTV was operated in splitless mode for 1 min with a carrier gas flow of 1mL min^{-1}, at a split flow of 20 mL min^{-1}, started at 65 °C, heated at a rate of 6 °C min^{-1} to 300 °C, held for 2 min, followed by a cleaning cycle at 340 °C. Separation in the GC was achieved on a Restek 20 m RXi Sil-MS capillary column (0.18 mm ID; 0.18 µm film thickness) with an oven programme starting at 55 °C for 1 min, first heated at 20 °C min^{-1} to 120 °C, then at 7 °C min^{-1} to 150 °C, finally at 20 °C min^{-1} to 300 °C. In the QqQ MS-system, three ion conversions per determinant are checked, the quantification took place at following mother-daughter ion-conversions: 316.08 -> 187.95 for TBT(D_{27})Et, 289.09->176.95 for TBTEt, 165.00 -> 122.90 for $SnPr_4$ and 249.08 -> 164.91 for $SnPr_3Et$, 244.08 -> 150.98 for MBT(D9)Et_3, 235.08 -> 178.95 for $MBTEt_3$, 263.03 -> 207.03 for $DBTEt_2$.

2.4 Total organic carbon

The concentration of each booster biocide in the sediment depends on the total organic carbon (TOC) value (OSPAR, 2018). Therefore, the TOC value is measured and used to normalise the booster biocide concentrations (see further). To measure the TOC of a sediment sample, a mixture of dichromate-sulphuric acid was added to 0.5 g of sediment. Next, the mixture was titrated with 0.2 N Mohr's salt using N-phenylanthranilic acid as indicator.

2.5 Quality control

All analytical sequences included the analysis of positive control samples and procedure blanks. Proficiency testing was done for TBT and TOC analysis by yearly participation to the Quasimeme interlaboratory exercises (Quasimeme, Wageningen, The Netherlands), focused on marine samples. The analyses of TBT and TOC are ISO/IEC 17025 accredited. A summary of booster biocide method validation is provided in Supplementary Materials.

2.6 Data analysis

The obtained dataset contains the concentrations of each component and the TOC fraction in the sediment for each sample. To be able to compare the concentration of each booster biocide at different locations in the BPNS, the measured concentration is normalised to a TOC value of 2.5%. The normalised concentration of a component in the sediment (C_n) can thus be calculated as:

$$C_n = \frac{C_s}{f_{\mathrm{TOC}}} 2.5\% \,,$$

with C_s the measured concentration of a component in the sediment and f_{TOC} the measured TOC value (ICES, 2018).

A concentration below the quantification limit was marked with < LOQ (or – in Tables). The quantification limit for each component is given in Table 1.

Table 1: Detection limits (LOQ) for six booster biocides (including TBT) and TOC.(TBT concentration presented as ng TBT-cation g^{-1})

Component	LOQ
DBT	0.31 ng g^{-1}
Dichlofluanid	0.81 ng g^{-1}
Diuron	0.08 ng g^{-1}
Irgarol	0.08 ng g^{-1}
MBT	1.37 ng g^{-1}
Sea-Nine 211	0.11 ng g^{-1}
TBT	0.41 ng g^{-1}
Tolylfluanid	1.14 ng g^{-1}
TOC	0.21%

When the measured TOC value was below the detection limit of 0.21%, the measured data points at that location were omitted. As the concentration of different booster biocides was often below the quantification limit, each location was assessed using the median value. If the median value was below the quantification limit, the lowest quantifiable measurement was used. The minimum and maximum values for each site, port, or harbour are also reported. Additionally, the measured concentrations will (after normalisation) be compared with concentrations reported in the ICES DOME database for the same environmental matrix (ICES, 2022). The data analysis is performed in R (version 4.1.2.) (R Core Team, 2013).

2.7 Risk assessment

The risk of each individual booster biocide on the marine environment was assessed based on the measured environmental concentrations (MEC) and the predicted no-effects concentration (PNEC). In this study, the MEC represented the concentrations measured in natural water (C_w) and was calculated based on the measured concentration of the corresponding booster biocide in the sediment (C_s):

$$C_w = \frac{C_s}{f_{\mathrm{TOC}} K_{\mathrm{OC}}},$$

with f_{TOC} the fraction of organic carbon in the sediment and K_{OC} the soil adsorption coefficient that was predicted using the MCI method in Epiweb (version 4.1) (EPA US, 2012).

The PNEC represents the maximum concentration of a specific booster biocide at which no adverse effects are expected to occur. The PNEC value is calculated by constructing a species sensitivity distribution (SSD) which was developed using a lognormal model as described by Aldenberg and Jaworska (2000) and implemented by Szöcs (2015) using the fitdistrplus package in the free statistical software R (R Core Team, 2013). Effective concentrations (EC_x), based on existing peer-reviewed ecotoxicity data, were collected from US EPA (2022). For an SSD to be valid, at least ten different species representing eight different taxonomic groups need to be integrated into the meta-analysis (ECHA, 2008). Therefore, the PNEC value was only calculated for Dichlofluanid, Diuron, Irgarol, Sea-Nine and TBT. When multiple concentrations were available for the same organism, the lowest concentration was used (worst-case scenario). For Tolylfluanid, not enough data were available, and a PNEC value reported in the literature was used instead. Based on an SSD, the HC5 is defined as the concentration at which 95% of the organisms are not affected. The mean HC5 and a confidence interval around the HC5 were derived using 1000 random parameter iterations of the distribution. The PNEC is then calculated as:

$$\mathrm{PNEC} = \frac{\mathrm{HC5}}{\mathrm{AF}},$$

with AF the assessment factor that is set at 5 to correct for the measurement uncertainty. The PNEC value is different for each booster biocide. When a booster biocide exceeds the PNEC value, that component may negatively affect the marine environment. This is expressed by the risk characterization ratio (RCR) value:

$$\mathrm{RCR} = \frac{C_w}{\mathrm{PNEC}},$$

with RCR >1 when the MEC exceeds the PNEC value.

3 Results

3.1 Booster biocide and TBT occurrence

Concentrations of booster biocides and TBT were highly variable, with highest variation at the harbours and ports (Table 2; Figure 2). Similar variations were also observed by others (Viana et al., 2019). For example, at dredged spoil disposal site Br&WOO, the concentration of TBT varied between 1.36 and 13.8 ng g^{-1}, whereas the concentration of Irgarol varied between <0.08 (LOQ) and 14.8 ng g^{-1}. Hotspots were identified where a concentration was detected at least three times the median concentration for that zone or site.

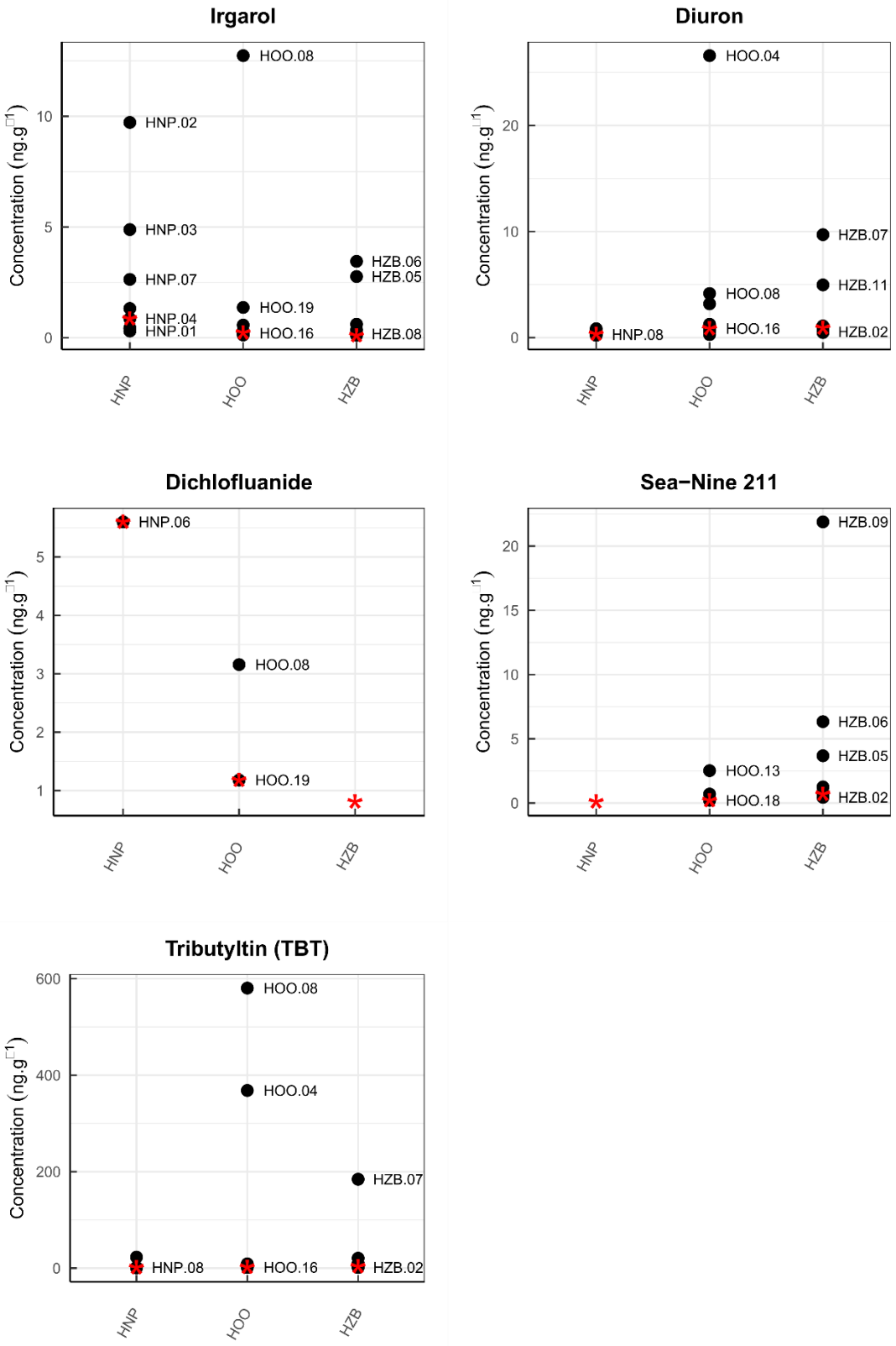

*Figure 2: The concentration (> LOQ) of Irgarol, Diuron, Dichlofluanid, Sea-Nine and TBT in sediment at each location in the harbour and ports of Nieuwpoort (HNP), Oostende (HOO) and Zeebrugge (HZB). Tolylfluanid is not shown as it was not detected in the harbour and ports (< LOQ). For each location, the median concentration is shown with as *. If the median < LOQ, the first quantifiable concentration is plotted, otherwise, the LOQ is used instead.*

Diuron and Irgarol were detected at all dredged spoil disposal sites (except for Diuron at LNP), ports and harbours. For Diuron, the highest concentrations in the dredged spoil disposal sites were detected at Br&WOO (4 ng g^{-1}), while for Irgarol high concentrations were detected at Br&WS2 (18.6 ng g^{-1}) and at Br&WOO (14.8 ng g^{-1}). For Diuron, concentrations above 4 ng g^{-1} were only measured at four locations. For example, in the port of Oostende, Diuron was detected with a maximum concentration of 26.6 ng g^{-1} in the "*Visserijdok*" (HOO 04), 31 times the median concentration (0.86 ng g^{-1}) for that site. In the port of Zeebrugge, highest concentrations of Diuron were detected in the channel that connects the port of Zeebrugge with the river Leie (HZB 11) and the "Prins Filip dock" (HZB07) with values of 4.97 ng g^{-1} and 9.70 ng g^{-1}, respectively. In the case of Irgarol, high concentrations were equally detected in the "*Visserijdok*" in the port of Oostende and in the military docks (HZB05) and the marina (HZB06) in the port of Zeebrugge, with values of 12.7 ng g^{-1}, 2.76 ng g^{-1}, and 3.45 ng g^{-1}, respectively. These concentrations are up to 55-fold the corresponding median concentration in the respective ports (Table 2).

Sea-Nine was detected in the ports of Oostende and Zeebrugge and the respective dredged spoil disposal sites Br&WOO and Br&WZE. Dichlofluanid was not detected at any dredged spoil disposal site, but was found in the harbour of Nieuwpoort and port of Oostende. Concentrations of Tolylfluanid never exceeded the detection limit (< LOQ). At the reference locations (REF), booster biocide concentrations (excluding TBT) seldomly exceeded the quantification limit (in 5 out of 50 cases), with only Diuron, or Irgarol being quantifiable in 4 reference samples (2018, 2019, 2020) from location ZVL and in 1 reference sample (2020) from location 120 (Table 2).

Although TBT has been banned since 2008, high concentrations of this component were still detected in the BPNS and the harbour and ports of Nieuwpoort, Oostende, and Zeebrugge (Table 2). The median concentration of TBT in the BPNS and the Belgian harbour and ports varied between 1.24 ng g^{-1} and 3.55 ng g^{-1}, however with a high spatial variation. In the port of Zeebrugge, high TBT concentrations were detected in HZB 06 and HZB 07 with concentrations of 21 ng g^{-1} and 184 ng g^{-1}, respectively. At the locations that are frequently dredged (e.g. HZB 02 and HZB 03) much lower TBT concentrations of respectively 1.6 ng g^{-1} and 1.7 ng g^{-1} were detected. At port Oostende, except for the "*Visserijdok*", the concentration of TBT ranged between 1.50 ng g^{-1} and 8.99 ng g^{-1}. In the "*Visserijdok*", a very high TBT concentration of 580 ng g^{-1} was detected. In the dredged spoil disposal sites, the median concentrations of TBT were more or less comparable with the median concentrations observed in the respective harbour and ports; and the maximum observed concentrations of TBT at most dredged spoil disposal sites were comparable to the reference locations (maxima between 3.4 and 4.2 ng g^{-1}), except for dredged spoil disposal sites LNP (max. 6 ng g^{-1}) and Br&WOO (max. 13.8 ng g^{-1}). By comparing the ratio between the concentration of TBT and the total concentration of tributyl compounds (MBT+DBT+TBT), a value higher than 0.5 could indicate a recent input in TBT (Mil-Homens et al., 2023). This was only the case for HOO and HNP with a ratio of 0.57 to 0.60. Overall, we can assume that the TBT concentrations measured at the BPNS as well as in the ports and harbour are mostly historical.

*Table 2: Median, minimum and maximum normalised concentrations of five booster biocides, TBT, DBT and MBT (expressed in ng g^{-1}), in the sediment from five dredged spoil disposal sites and six reference locations in the Belgian part of the North Sea (2018-2020), and from three Belgian harbours and ports (2020). When the median value was below the quantification limit (LOQ), the smallest quantifiable concentration was reported and marked with *. Values < LOQ were marked as -.*

	Dichlofluanid	Diuron	Irgarol	Sea-Nine	Tolyl-fluanid	TBT	DBT	MBT
LNP	-	-	2.21* [LOQ-2.21]	-	-	1.24 [0.79-5.97]	0.94 [0.83-3.33]	1.33 [1.05-2.64]
Br&WOO	-	0.47 [LOQ-4.0]	0.23 [LOQ-14.8]	0.16* [LOQ-0.16]	-	2.07 [1.36-13.8]	1.26 [1-7.51]	2.13 [1.79-8.92]

Br&WZE	-	0.46 [LOQ-1.47]	0.13* [LOQ-1.11]	0.39* [LOQ-0.39]	-	2.62 [1.48-3.60]	2.15 [1.18-2.92]	2.51 [1.22-4.37]
Br&WS2	-	0.87* [LOQ-0.87]	18.63* [LOQ-18.6]	-	-	1.77 [1.21-3.36]	1.45 [1.21-2.29]	2.21 [1.68-3.41]
Br&WS1	-	0.91* [LOQ-0.91]	0.45* [LOQ-0.45]	-	-	2.14 [1.34-4.24]	1.41 [1.02-1.75]	1.58 [0.92-2.63]
REF	-	0.32* [LOQ-0.6]	0.19* [LOQ-0.65]	-	-	1.68 [0.59-3.40]	1.89 [0.52-4.97]	2.54 [1.30-6.33]
HNP	5.60* [LOQ-5.60]	0.35 [LOQ-0.84]	0.86 [LOQ-9.72]	-	-	1.54 [0.59-22.9]	1.12 [0.89-2.69]	-
HOO	1.18* [LOQ-3.16]	0.86 [0.28-26.58]	0.23 [0.12-12.74]	0.21* [LOQ-2.52]	-	2.32 [1.50-580]	1.51 [1.2-258]	-
HZB	-	0.92 [LOQ-9.7]	0.10* [LOQ-3.45]	0.70 [LOQ-21.88]	-	3.55 [1.48-184]	3.54 [1.42-74.9]	-

3.2 Risk assessment of booster biocides and TBT in the BPNS

To assess the risk of each booster biocide, a PNEC value was calculated. First, an SSD was constructed for Diuron, Irgarol, Sea-Nine and for TBT and Dichlofluanid. The SSD for TBT is shown in Figure 3, whereas the SSD for Diuron, Dichlofluanid Irgarol and Sea-Nine are reported in the Supplementary Materials. The HC5, as well as the 95% confidentiality interval and the PNEC values are reported in Table 3.

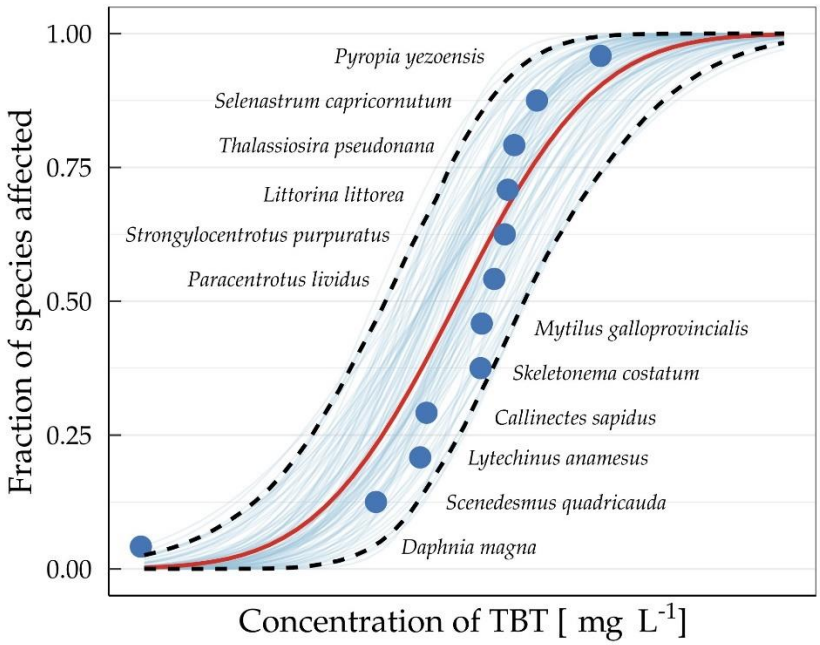

Figure 3: The SSD for TBT using the EC values of 12 different organisms with at least 8 different taxonomical classes. The data was collected from the ESA platform (EPA US, 2022).

*Table 3: The PNEC values and their 95% confidentiality interval. The Log K_{oc} was predicted using epiweb (MCI method). *The PNEC value for Tolylfluanid was reported by de Campos et al. (2022). The PNEC in sediment was calculated based on the PNEC in water and a TOC value of 2.5% (see Data analysis in Material and Methods).*

	HC5	**95% confidentiality interval**			**Log K_{oc}**	**PNEC**	
	[mg L^{-1}]	2.5%	50%	97.5%	[-]	Water [ng L^{-1}]	Sediment [ng g^{-1}]
Dichlofluanid	$6.2*10^{-3}$	$3.37*10^{-4}$	$1.63*10^{-3}$	$6.89*10^{-3}$	2.60	1242	12.4
Diuron	$1.62*10^{-4}$	$5.56*10^{-6}$	$3.84*10^{-5}$	$2.51*10^{-4}$	2.04	32.5	0.09
Irgarol	$1.07*10^{-5}$	$4.51*10^{-7}$	$2.53*10^{-6}$	$1.35*10^{-5}$	2.40	2.14	0.01

Sea-Nine 211	$3.60*10^{-4}$	$1.43*10^{-5}$	$9.52*10^{-5}$	$5.60*10^{-4}$	3.36	72.1	4.13
TBT	$1.58*10^{-7}$	$1.74*10^{-9}$	$5.66*10^{-8}$	$1.65*10^{-6}$	3.91	0.032	0.007
Tolylfluanid	-	-	-	-	2.80	2.53*	0.04

The potential environmental risk for each booster biocide and TBT is given by the RCR in Table 4. In the case of Irgarol, the median concentration at any location resulted in an RCR value higher than one, with maximum RCR values of 1386 at Br&WS2, 1098 at BR&WOO, and 948 at the "*Visserijdok*" (HOO 08) in port of Oostende. The median concentrations of Diuron also exceeded the safe threshold at each dredged spoil disposal site (except LNP) and even at some of the reference locations. In the harbour and ports, the safe threshold is exceeded in the most contaminated sampling locations, such as the "*Visserijdok*" (HOO 04 and HOO 08) in port of Oostende, the channel connecting the port of Zeebrugge with the Leie (HZB11) and the "Prins Filip dock" (HZB07) in the port of Zeebrugge.

Sea-Nine was limited to the ports of Oostende and Zeebrugge and their respective dredged spoil disposal site, but the RCR values remained below one (except for HZB06 and HZB09 in port of Zeebrugge). Both Tolylfluanid and Dichlofluanid were measured at low concentrations (or below the LOQ), resulting in RCR values below one.

Table 4: The RCR value for four booster biocides and TBT at five dredged spoil disposal sites, and six reference locations in the Belgian part of the North Sea, and at three Belgian harbour and ports. The RCR values are calculated based on the median and the maximum measured concentrations. Values above 1, indicates a risk for the marine environment and are marked in bold. Values < LOQ were marked as -. As Tolylfluanid was not detected in any samples, it was omitted from this table.

		Dichlofluanid	Diuron	Irgarol	Sea-Nine	TBT
LNP	Median	-	-	**164**	-	**190**
	Max	-	-	**164**	-	**918**
BR&WOO	Median	-	**5.26**	**17.2**	0.04	**318**
	Max	-	**44.9**	**1098**	0.04	**2115**
BR&WZE	Median	-	**5.13**	**9.52**	0.09	**403**
	Max	-	**16.5**	**82.7**	0.09	**554**
BR&WS2	Median	-	**9.77**	**1386**	-	**272**
	Max	-	**9.76**	**1386**	-	**517**
BR&WS1	Median	-	**10.2**	**33.6**	-	**330**
	Max	-	**10.2**	**33.5**	-	**652**
REF	Median	-	**3.55**	**13.7**	-	**258**
	Max	-	**6.71**	**48.1**	-	**523**
HNP	Median	0.45	**3.31**	**64.2**	-	**237**
	Max	0.45	**9.42**	**723**	-	**3524**
HOO	Median	0.10	**9.62**	**17.0**	0.05	**357**
	Max	0.26	**298**	**948**	0.61	**89275**
HZB	Median	-	**10.3**	**7.74**	0.17	**546**
	Max	-	**109**	**257**	**5.30**	**28352**

TBT was detected at each location, resulting in RCR median values ranging between 190 and 546. The highest RCR values were calculated for the "*Visserijdok*" (HOO 04 and HOO 08) in port of Oostende with maximum values of 56,632 and 89,274, and in the "Prins Filip dock" (HZB 07) in port of Zeebrugge, with a value of 28,352. For the dredged spoil disposal sites, highest RCR values were calculated for BR&WOO (2115). The median RCR values were a little higher in most dredged spoil disposal sites (ranging between 272 and 403) except for LNP (190) compared to the reference locations (258).

4 Discussion

4.1 Booster biocides and TBT at dredged spoil disposal sites

In the ports and harbour of Oostende, Zeebrugge, and to a lesser extent Nieuwpoort, excessive sludge is dredged and dumped at the disposal sites Br&WOO, Br&WZE, and LNP, respectively. Sludge is hypothesised to be contaminated with PAHs, PCBs and, metals, and as such may cause adverse environmental effects in their disposal locations, away from their original sources. The contamination at dredged spoil disposal site LNP is limited. For the measured booster biocides and TBT, only Irgarol and TBT impose a potential risk on the marine environment at LNP. LNP is sandy, limiting the adsorption of contaminants to the sediment (OSPAR, 2018). Indeed, for organics, the measured concentration in sediment is correlated to the concentration in the water fraction and the total organic carbon in the sediment. Moreover, the amount of sludge dumped at LNP is low compared to other dredged spoil disposal sites resulting in low contamination.

At dredged spoil disposal sites Br&WOO and Br&WZE, Irgarol, Diuron, and TBT were detected at quantifiable concentrations. Both Irgarol and Diuron are persistent in the marine environment, allowing their accumulation at dredged spoil disposal sites, resulting in an RCR value higher than one for both components. Especially compared to the reference locations, an increased contamination was observed at both dredged spoil disposal sites. Also for TBT, higher maximum concentrations were detected at BR&WOO compared to reference locations or reported values in the ICES Dome database. So, even in the open sea, a detrimental effect of Irgarol, Diuron and TBT can be expected on the marine environment. Due to the ban on TBT and Irgarol, it is expected that these concentrations will decrease over time.

There was no clear link between the contamination pattern at the two dredged spoil disposal sites located further offshore (Br&WS1 and Br&WS2) and the port of Zeebrugge, although these sites are extensively used for the disposal of dredged material from the port of Zeebrugge (± 55% of all material disposed here). Most probably, the dredged spoil disposal sites Br&WS1 and Br&WS2 are more sandy (comparable to LNP), resulting in a lower TOC value and lower adsorption of contaminants in the sediment. On the other hand, both sites are mainly used for the disposal of dredged material coming from the channel connecting the port of Antwerp with the North Sea (± 45% of all dredged material). It is anticipated that the mixture of dredged materials from different sources may have an impact on the chemical fingerprint found in our analysis ((Vandermarken et al., 2018), Table 2). The contamination of Irgarol at these sites could thus be related to the contamination in the main traffic channel, although more research is required to confirm this hypothesis. As the RCR values of both components exceed a value of one, a detrimental effect on the marine environment can be expected, meaning that the source of contamination is to be established.

4.2 Concentration and risk assessment of booster biocides and TBT in coastal harbours and ports

There is a clear link between the concentration of TBT and booster biocides at a port or harbour and its size and activity. The harbour of Nieuwpoort is one of the biggest marinas in Northern Europe with more dan 2000 berths and three different yachting clubs (Belgian Coast, 2021). At the harbour of Nieuwpoort, a higher concentration of Irgarol and Dichlofluanid was measured compared to the port of Zeebrugge, which might be linked to the higher traffic of small vessels and yachts (Thomas, Blake, & Waldock, 2000). Surprisingly, the highest concentrations of Irgarol were not measured in the marina Portus Novus, but in the Yser channel leading to the marina. These high concentrations could be the results of recreational boating and due to inland waterway transport. In contrast to Irgarol, TBT

concentrations at Nieuwpoort are lower than at the industrial port of Zeebrugge. Nevertheless, even the lowest measured concentration resulted in an RCR value higher than one.

The activities at the port of Oostende vary from transporting and stocking bulk products (e.g. sand and gravel), welcoming cruises, harbouring more than 65 fishery vessels, and delivering technical services like boat maintenance (Port Oostende, 2021). Because the port of Oostende is home to both recreational boating and commercial shipping, the contamination pattern of Oostende is intermediate between the port of Zeebrugge and the harbour of Nieuwpoort. Similar to the port of Zeebrugge and the harbour of Nieuwpoort, the TBT concentration exceeded the critical RCR value of one. At port Oostende, high concentrations were measured at HOO 04 and HOO 08, a region called the "*Visserijdok*", leading to high RCR values. The "*Visserijdok*" is used for harbouring fishery vessels and their maintenance. Because that area is isolated from the port by a sluice, and the water bottom is not dredged, booster biocides and TBT can accumulate. Moreover, the maintenance of these ships can result in a higher concentration of paint leachates in the sediment. Remarkably, Sea-Nine was also detected in the port of Oostende, but not in the "*Visserijdok*" as Sea-Nine has major use on large commercial vessels (Rohm and Haas, 2006).

The port of Zeebrugge is one of the major commercial ports in Europe. In 2020, more than 47 million tonnes of cargo were transhipped. The shipment varies from liquid natural gas to high and heavy cargo and cars (Port of Zeebrugge, 2021). The sediments in the industrial port of Zeebrugge are characterised by relatively high concentrations of Sea-Nine, coupled to the presence of large commercial vessels and super yachts. Although the observed concentrations of Sea-Nine are relatively high at Zeebrugge, only at 2 sampling locations (HZB 06 and HZB 09) an RCR value >1 was observed. Diuron and Irgarol were also detected in the port, however, high critical concentrations were only detected at specific hotspots, especially where smaller vessels are moored (<25 m) (Thomas, Blake, & Waldock, 2000). However, due to the high toxicity and persistence of Irgarol and Diuron, even the lowest detectable concentration resulted in an RCR >1.

Even after the ban on TBT since 2008 (15 years ago), high TBT concentrations are still measured in the industrial port of Zeebrugge. These concentrations can be linked to the amount of ship traffic, but dredging activities also affect the concentrations in the environment: lowest concentrations of TBT were found at locations where a lot of dredging takes place, whereas the highest concentrations were found at docks where excessive sludge is not (or less often) removed, leading to an accumulation of TBT. Unfortunately, because MBT was not measured in the ports, it was not possible to assess if current contamination levels were historical.

At both the "*Visserijdok*" in port of Oostende and the "*Prins Filip dock*" in port of Zeebrugge, at least two booster biocides (Irgarol and Diuron) and/or TBT were detected which resulted in an RCR >1. At these locations, a detrimental effect on the marine environment can be expected. As long as the sludge remains in place, the effect on the marine environment is contained. However, when the sludge is dredged, it should not be dumped in the open sea because of the risk to the marine environment. In the case of polycyclic aromatic hydrocarbons (PAHs), polychlorinated biphenyls (PCBs), metals, and TBT, concentrations must be below predefined thresholds, before the dredged material may be dumped at the dredged spoil disposal sites at sea (MMO, 2015). For booster biocides other than TBT, there are (yet) no disposal limits, while these are indispensable to minimise the contamination of these booster biocides, both in the open sea and at dredged spoil disposal sites, as they can affect residential organisms.

4.3 Booster biocides: overlooked contaminants?

Despite booster biocides being routinely applied after the TBT ban, only a limited amount of concentration data is available in the ICES region. The monitoring of booster biocides is limited, regardless that reported concentrations often exceed the PNEC values. Consulting the ICES Dome database (ICES, 2022), Tolylfluanid and Dichlofluanid have only been reported by Estonia in 2017, with measurements below the detection limit (10 ng g^{-1}). Unfortunately, it was not possible to normalise the concentration as the corresponding TOC values were not reported.

Sea-Nine was only reported by Sweden in 2008, reporting an averaged normalised concentration of 0.03 ng g^{-1} (ICES, 2022), comparable to our concentrations observed at Br&WOO and slightly below the concentrations recorded for BR&WZE, as the latter dredged spoil disposal site is linked with the industrial port of Zeebrugge. Diuron and Irgarol were monitored by Estonia and Sweden in 2021. Sweden reported non-normalised concentrations of 0.13 ng g^{-1} and 0.08 ng g^{-1} for Diuron and Irgarol, respectively, whereas Estonia reported a concentration of 0.09 ng g^{-1} for Diuron and concentrations below the detection limit of 0.5 ng g^{-1} for Irgarol. In the ports of Southwestern Spain, concentrations of 2.1 ng g^{-1} and 2.9 ng g^{-1} were reported for Sea-Nine and Irgarol, respectively (García et al., 2020). In Malaysia, similar concentrations were detected for Diuron (<0.1-22.9 ng g^{-1}) and Irgarol (0.1-1.4 ng g^{-1}), and even higher concentrations for Sea-Nine (9.1-170 ng g^{-1}) (Mukhtar et al., 2019).

The most recent data in the ICES DOME database reported for TBT go back to 2018. For the United Kingdom and Denmark, values are mostly below the detection limit of 2 ng g^{-1} and 1 ng g^{-1}, respectively (ICES, 2022). However, in both cases the PNEC value is exceeded and thus TBT has the potential to exhibit a negative effect on the marine environment. For Belgium, an averaged normalised concentration of 1.91 ng g^{-1} was reported in 2018 (ICES, 2022), corresponding to the present values reported in this study (Viana et al., 2019).

Although several publications have been published in the early 2000 (Harino et al., 2006; Thomas et al., 2001; Thomas, McHugh, & Waldock, 2002), the total number of publications on booster biocide occurrence stays limited. Several publications focus on the concentration of booster biocides in South America (Abreu, Martins, & Fillmann, 2021; Batista-Andrade et al., 2016; Castro et al., 2021). In Batista-Andrade et al. (2016) Irgarol was detected in 72% of the samples with a concentration up to 2.8 ng g^{-1}, whereas Diuron was detected in 33% of the cases with a concentration up to 14.1 ng g^{-1}. In another study, concentrations up to 55.2 and 45.6 µg kg^{-1} were reported for Diuron and Irgarol, respectively. In Çetintürk and Ünlü (2022) concentrations up to 11.28 and 26.53 ng g^{-1} have been reported in a port nearby the Black Sea in Turkey.

It is clear that sediment contamination by booster biocides is not limited to the BPNS, and a detrimental effect on the marine environment can be expected in many regions. sDetermination of the PNEC value, remains therefore import to assess the potential negative effects of these booster biocides on the marine environment. Abreu, Martins and Fillmann (2021) reported a PNEC value of 0.15 ng g^{-1} and 16 ng g^{-1} for Diuron and Irgarol in sediment, respectively. The PNEC value for Diuron in this study was slightly more conservative (0.09 ng g^{-1}). However for Irgarol, a much lower value was obtained (0.01 ng g^{-1}). This was however much closer to the values (0.04 and 0.005 ng g^{-1}) reported by ECHA (2014b) and Barbieri et al. (2019), respectively. de Campos et al. (2022) reported a PNEC value of 5.52 µg L^{-1} for tolylfluanid. The value obtained in this study is again slightly more conservative (1.2 µg L^{-1}). The differences in PNEC values can be explained by differences in the used method and included toxicological data. In this study, the PNEC value was calculated using an SSD which was reported as being more accurate and ecological relevant than other methods (Figueiredo, Loureiro, & Martins, 2020; Sorgog & Kamo, 2019).

As RCR values are based on species sensitivity distributions, an assessment of the environmental impact is possible. High RCR values, especially for Diuron and Irgarol, indicate the importance of measuring these compounds on a regional scale within the greater North Sea and beyond. Moreover, the concentration of booster biocides may be linked to the pollution by shipping traffic and dredged spoil disposal activities. To assess trends and impacts of booster biocide concentrations, it is advised to monitor these components within environmental monitoring programs and to define environmental quality standards (EQS).

Moreover, having an RCR value exceeding 1 is only an indication that the ambient environmental concentration of a specific chemical may provoke negative environmental effects. Synergistic effects of different booster biocides or other chemicals can equally be expected, resulting in an underestimation of their effective field toxicity (Kottuparambil, Lee, & Han, 2013). The microalgae community, for example, can already be affected at low concentration of both Diuron and Irgarol due to their synergetic effect (Koutsaftis & Aoyama, 2006). Therefore, it remains difficult to predict the overall negative effect of a mixture of booster biocides at a specific location, as the interaction depends on their relative concentration (Koutsaftis & Aoyama, 2007). Sea-Nine was, due to its rapid degradation (Jacobson & Willingham, 2000; Moon et al., 2019), only detected in a low concentration in the BPNS resulting in an RCR value below one. Nevertheless, even at these low concentrations, Sea-nine can still interact with other components such as Irgarol, increasing environmental toxicity (Arrhenius et al., 2006).

5 Conclusion

In each port and harbour, a specific contamination pattern of booster biocides was detected according to the vessel type and activities. Booster biocides such as Irgarol, Diuron, and Dichlofluanid are more strongly associated with yachts and fishery vessels, whereas Sea-Nine is more associated with cargo ships and other commercial vessels. Similar contamination patterns were also detected at the respective dredged spoil disposal sites. Sea-Nine easily degrades in coastal waters and was therefore only detected in low concentrations. However, more persistent components such as Irgarol, Diuron, and TBT do accumulate in time both in the ports and at dredged spoil disposal sites, still affecting the marine environment even long after the ban on TBT (> 15 years) and a more recent ban on Irgarol (since 2021).

In the BPNS, concentrations with an RCR value of more than 1 were observed in 97% of the samples at four out of the five dredged spoil disposal sites. This indicates that the current ambient booster biocide concentrations may pose a risk to the marine environment. Moreover, different hotspots have been detected in different parts of the main Belgian ports (*e.g.* “Visserijdok” in port of Oostende, “prins Filip dock” and “military dock” in port of Zeebrugge, “Yser channel” in harbour of Nieuwpoort), where higher concentrations of certain components were detected.

Booster biocides and TBT are detectable in the marine environment and may thus affect non-target organisms. It is therefore recommended to monitor these components such that hotspots may be discovered, and that actions can be taken in time if the concentration of a component exceeds the threshold in which unreversible damage on the marine environment may be expected. Additionally, further toxicity tests may help to understand the effects of booster biocides on non-target organisms as well as the synergistic effects between the different booster biocides.

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7 Author contributions

BDW, GE and CJ conceived and supervised the study. The data were analysed by DV. DV and BDW drafted the manuscript. KH supervised the study and adapted the manuscript. KP assisted in TBT analysis and wrote that methodological part. All authors reviewed and approved the manuscript.

8 Conflict of interest

The authors declare that they have no conflicts of interest.

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Figure3

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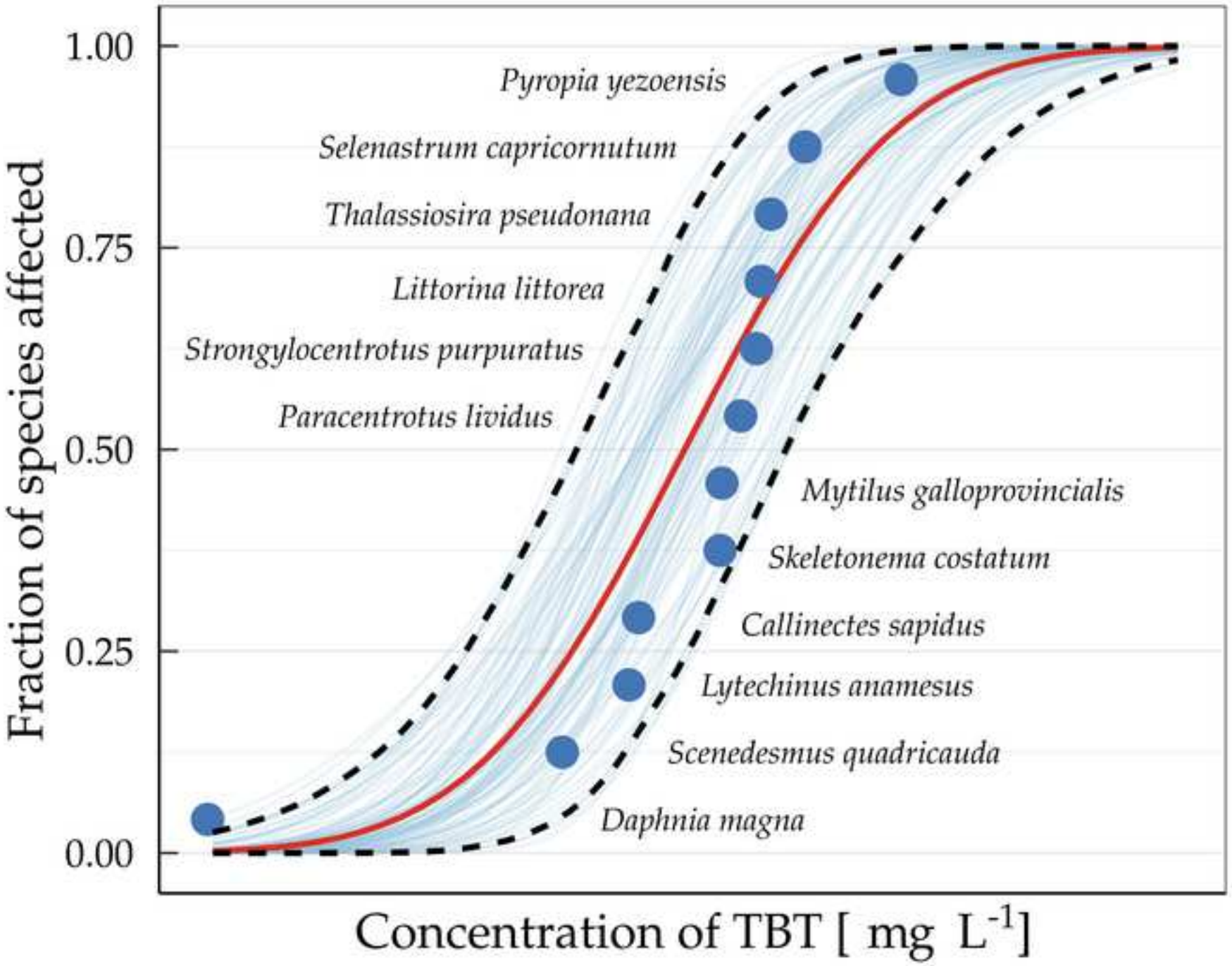

Figure 1

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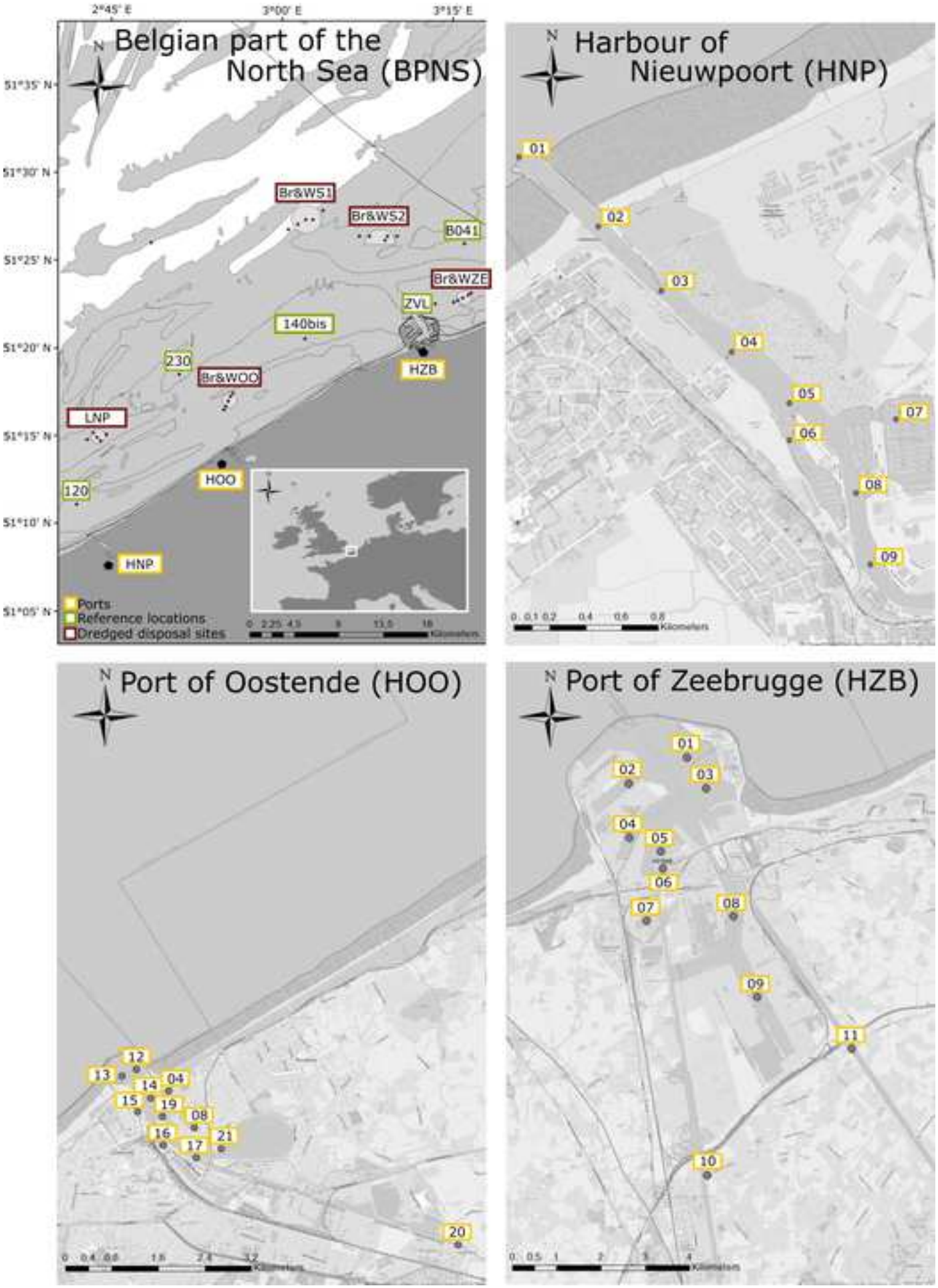

Figure 2

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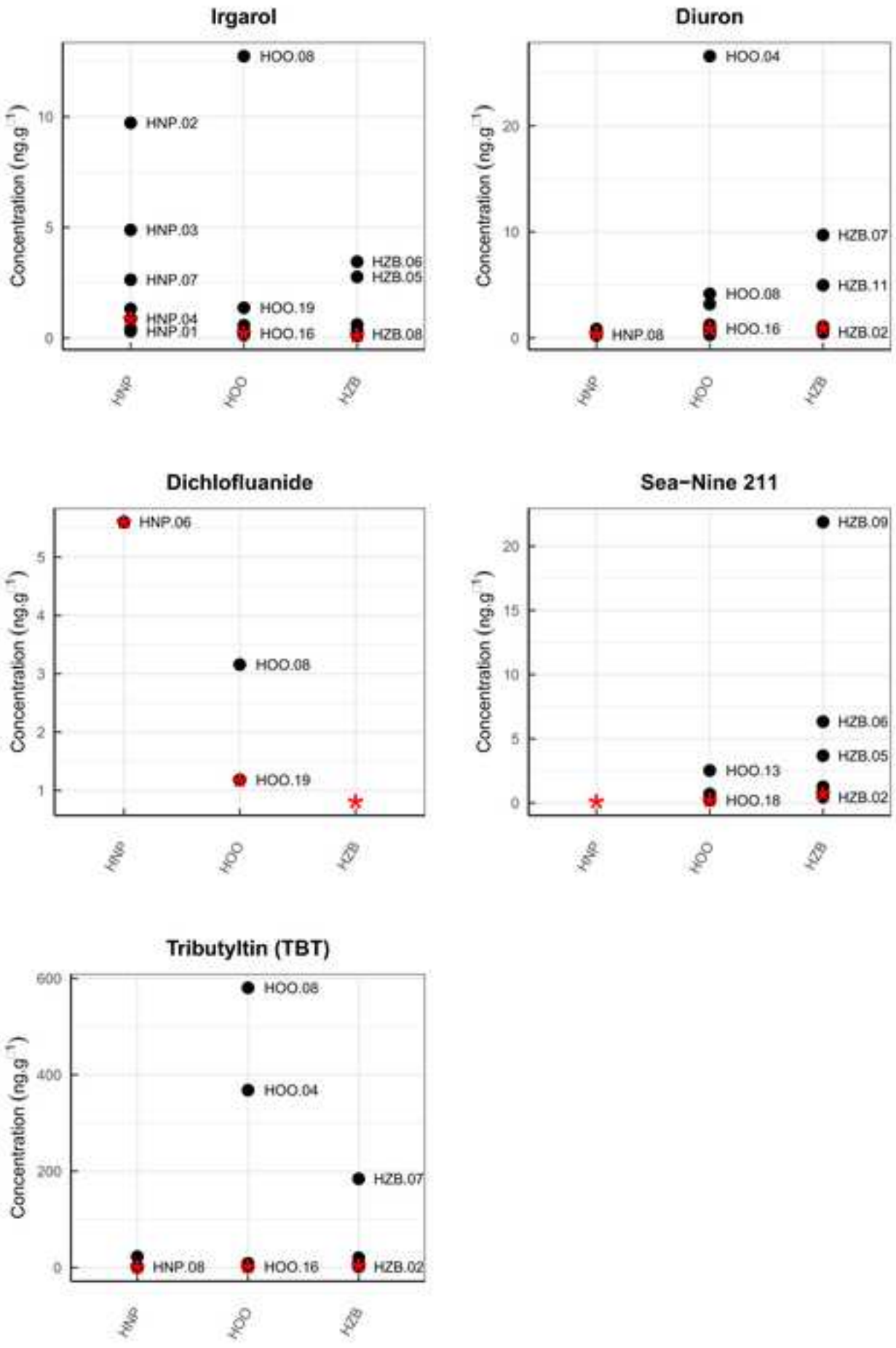

Validation of the booster biocide analytical methods

For the validation of the booster biocide analytical methods, the accuracy, reproducibility and linearity of the method are analysed and the detection and quantification limits are defined. First, a mixed sample is made by combining freeze-dried sediment from different locations in the Belgian part of the North Sea (BPNS). The mixed sample is then analysed to determine the background noise. Next, the mixed sample is spiked with each booster biocide. The accuracy of the analytical method is calculated by comparing the measured concentration with the concentration that was added to the sample. In total six samples were analysed per booster biocide over three different days within three weeks to calculate the reproducibility of the methods. Finally, the mixed sample was spiked with different concentrations for each booster biocide to calculate the linearity of the analytical method.

The detection limit (LOD) and quantification limit (LOQ) were calculated using a procedure blank. The LOD and LOQ are defined as respectively three and six times the standard deviation of the measured concentration in the procedure blank. The precision and reproducibility of the analytical methods are reported in Table S1. The linearity is beyond the scope of this publication and is thus not reported.

Table S1:Accuracy and reproducibility of the analytical methods for the detection and quantification of booster biocides.

Booster Biocide	Low spike		High Spike	
	Accuracy (%)	Reproducibility (%)	Accuracy (%)	Reproducibility (%)
Dichlofluanid	98.9	10.07	102.2	15.84
Diuron	108.4	4.33	121.5	15.92
Irgarol	100.4	5.97	106.8	15.33
Sea-Nine	105.2	8.22	123.9	30.16
Tolylfluanid	89.4	13.46	103.5	19.90

Sample Locations

Table S2: Coordinates of sampling locations on the Belgian Part of the North Sea (BPNS).

	Latitude (WGS84; °N)	**Longitude (WGS84; °E)**
Reference points		
120	51.18500	2.70118
140bis	51.34217	3.03292
230	51.30833	2.84999
330	51.43333	2.80833
B041	51.43250	3.26534
ZVL	51.37553	3.22228
BR&WS1		
BR&WS1.01	51.45548	3.03343
BR&WS1.06	51.45090	3.02262
BR&WS1.07	51.45538	3.04420
BR&WS1.19	51.44607	3.00861
BR&WS1.20	51.46437	3.05875
BR&WS2		
BR&WS2.04	51.43951	3.12733
BR&WS2.06	51.43565	3.14981

BR&WS2.07	51.43958	3.15275
BR&WS2.08	51.43945	3.11219
BR&WS2.10	51.43952	3.16707
BR&WZE		
BR&WZE.01	51.38077	3.26257
BR&WZE.04	51.37852	3.25547
BR&WZE.05	51.38365	3.26970
BR&WZE.10	51.37668	3.24963
BR&WZE.11	51.38557	3.27458
BR&WOO		
BR&WOO.01	51.28300	2.92129
BR&WOO.03	51.27835	2.91753
BR&WOO.06	51.28725	2.92487
BR&WOO.09	51.27498	2.91479
BR&WOO.12	51.29093	2.92806
LNP		
LNP.01	51.24873	2.73023
LNP.03	51.24545	2.73553
LNP.06	51.25265	2.72517
LNP.08	51.24625	2.71653
LNP.11	51.25138	2.74395

Table S3: Coordinates of sampling locations at the ports and harbour of Nieuwpoort (HNP0, Oostende (HOO), and Zeebrugge (HZB).

	Latitude (WGS84; °N)	**Longitude (WGS84; °E)**
HNP		
HNP.01	51.15650	2.71708
HNP.02	51.15305	2.72337
HNP.03	51.14985	2.72840
HNP.04	51.14680	2.73400
HNP.05	51.14425	2.73858
HNP.06	51.14240	2.73862
HNP.07	51.14345	2.74703
HNP.08	51.13977	2.74388
HNP.09	51.13620	2.74507
HOO		
HOO.04	51.23586	2.93716
HOO.08	51.22981	2.93716
HOO.12	51.23928	2.92283
HOO.13	51.23825	2.91930
HOO.14	51.23465	2.92647
HOO.15	51.23248	2.92322
HOO.16	51.22700	2.92953
HOO.17	51.22503	2.93768
HOO.19	51.23172	2.92932
HOO.20	51.21099	3.00220

HZB		
HZB.01	51.35473	3.20810
HZB.02	51.34945	3.18917
HZB.03	51.34843	3.21432
HZB.04	51.33845	3.18950
HZB.05	51.33573	3.19963
HZB.06	51.33215	3.20017
HZB.07	51.32160	3.19498
HZB.08	51.32248	3.22303
HZB.09	51.30608	3.23060
HZB.10	51.36986	3.21263
HZB.11	51.29557	3.26105

Species sensitivity distribution plots

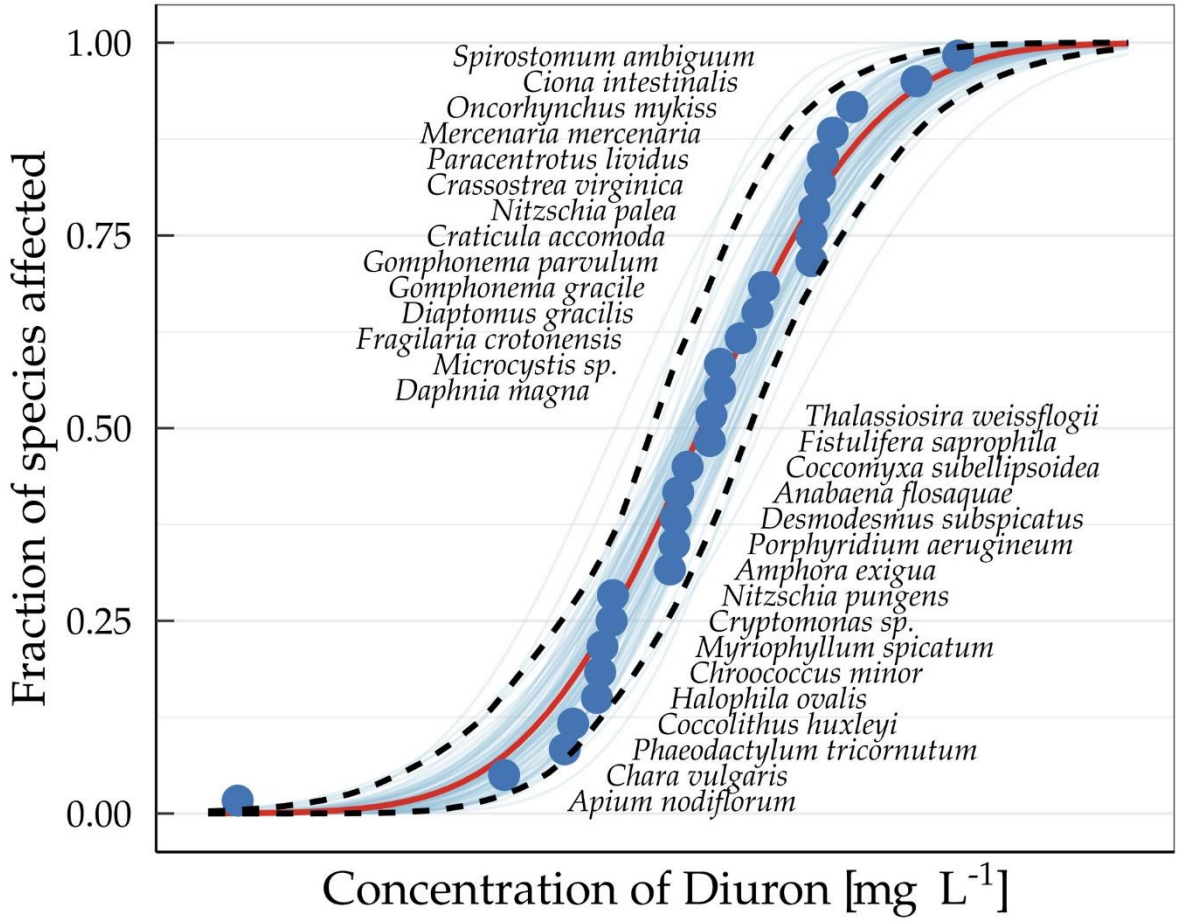

Figure S1: The SSD for Diuron using the EC values of 12 different organisms with at least 8 different taxonomical classes. The data was collected from the ESA platform (EPA US, 2022).

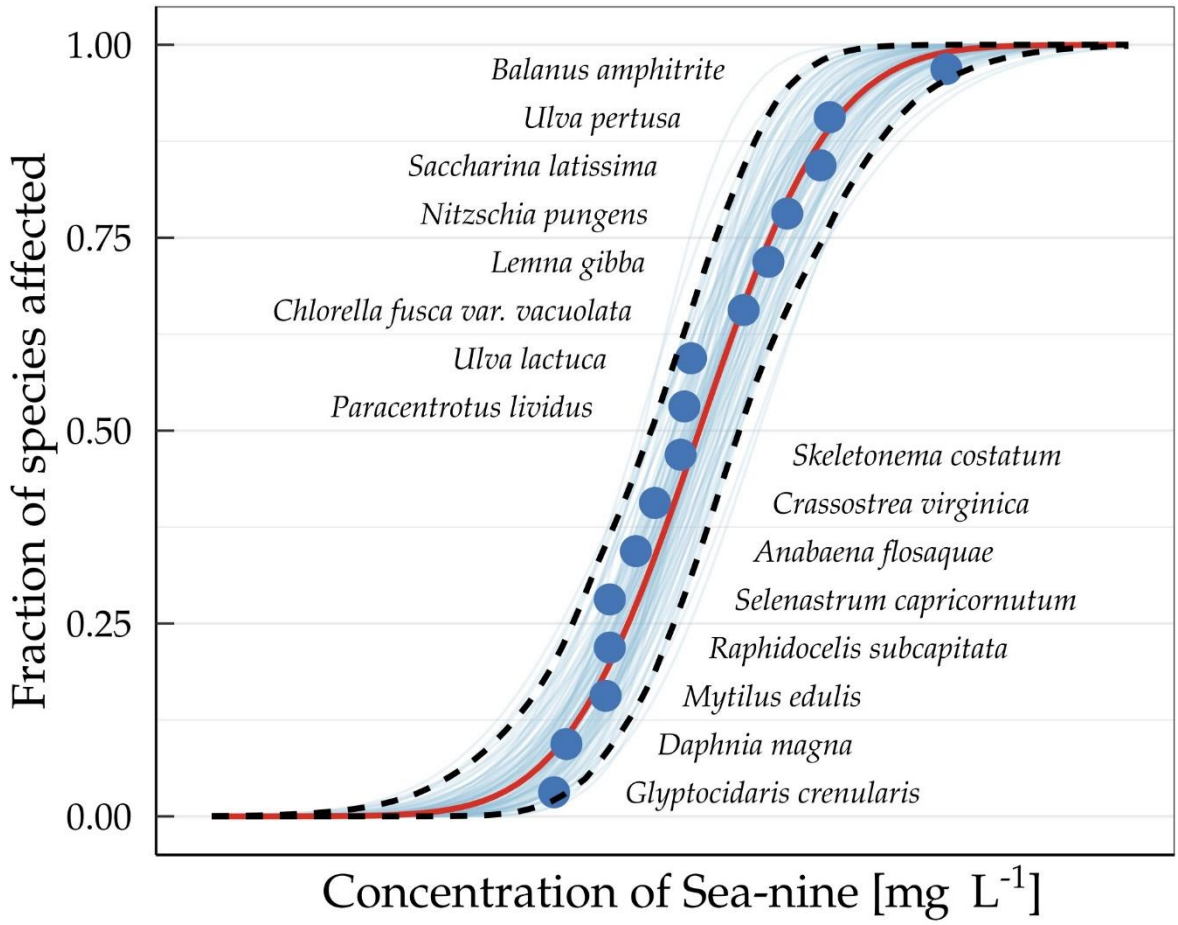

Figure S2: The SSD for Sea-Nine using the EC values of 12 different organisms with at least 8 different taxonomical classes. The data was collected from the ESA platform (EPA US, 2022).

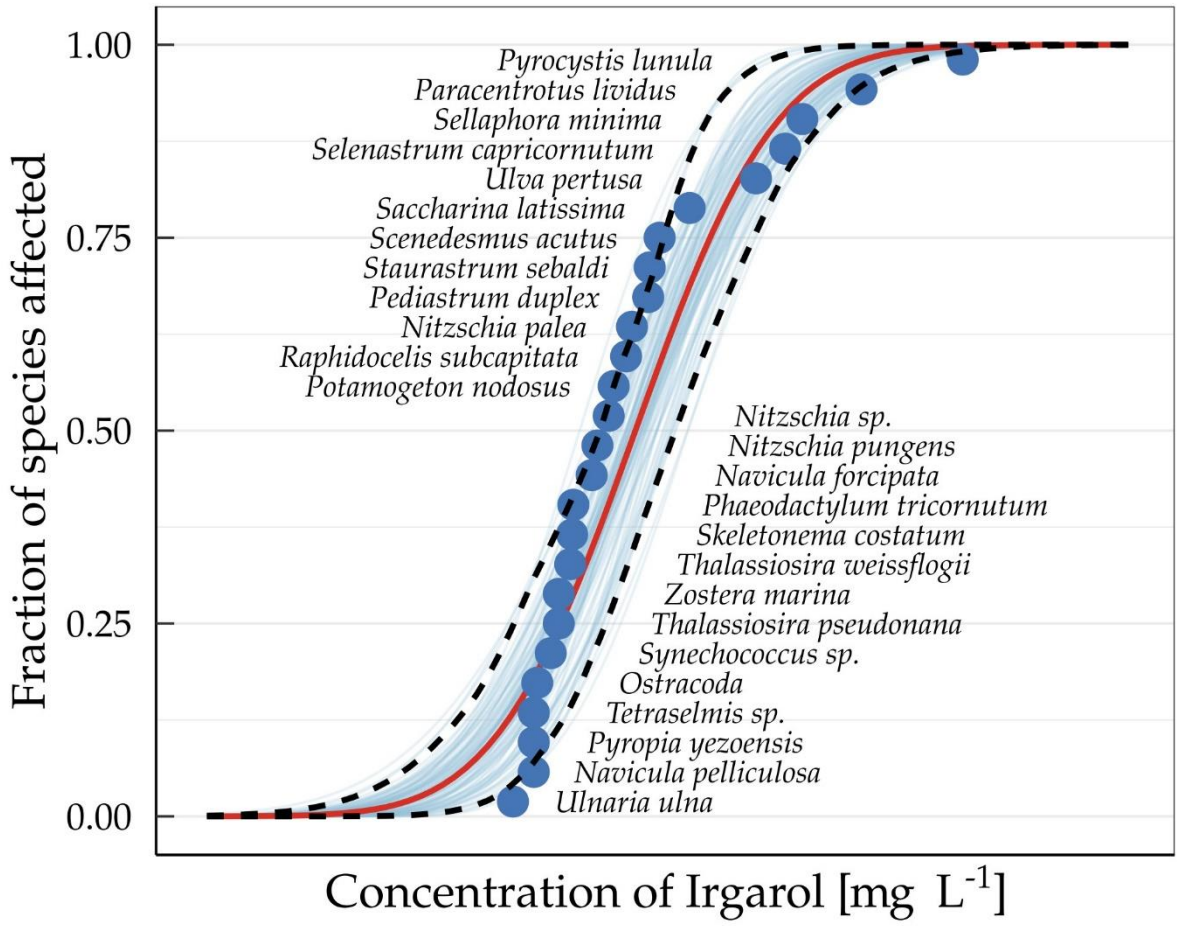

Figure S3: The SSD for Irgarol using the EC values of 12 different organisms with at least 8 different taxonomical classes. The data was collected from the ESA platform (EPA US, 2022).

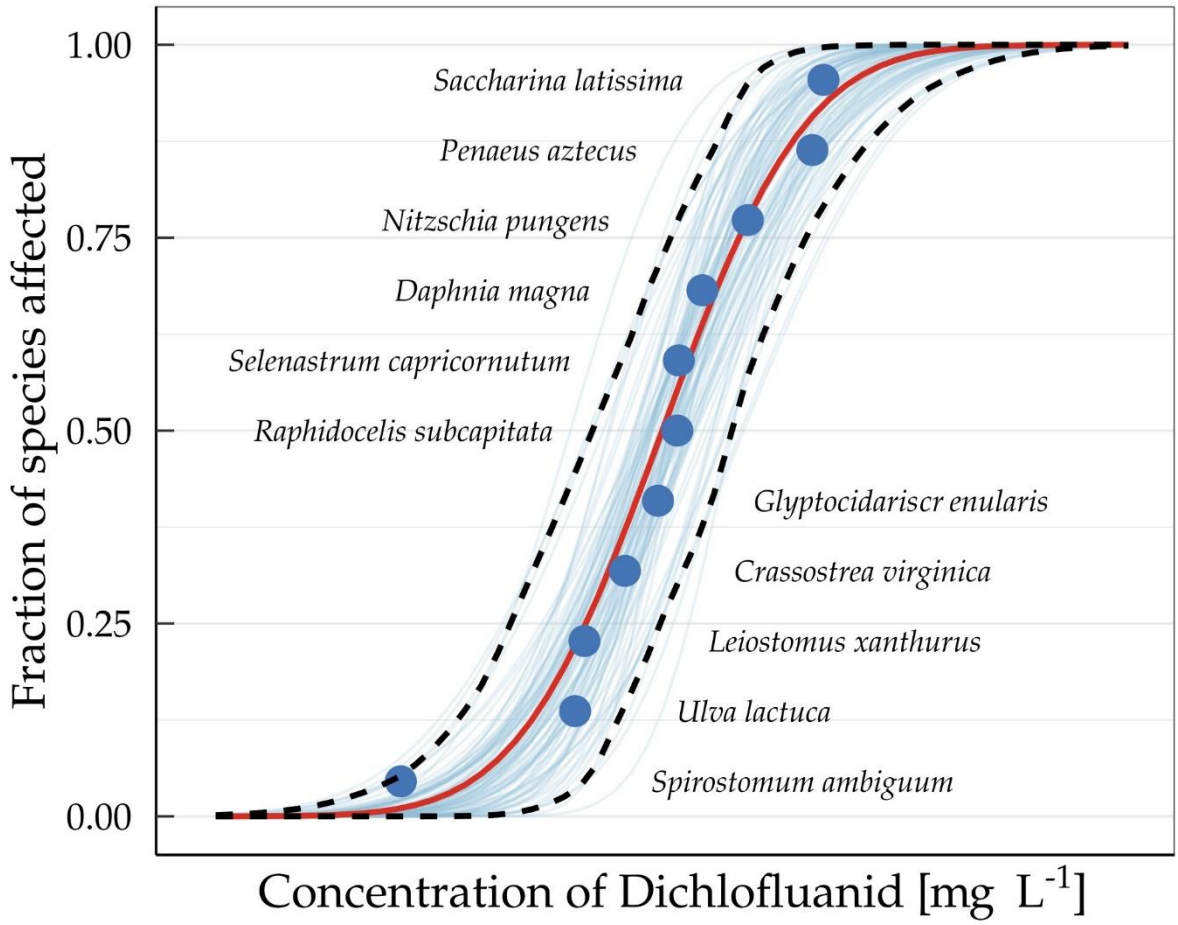

Figure S4: The SSD for Dichlofluanid using the EC values of 12 different organisms with at least 8 different taxonomical classes. The data was collected from the ESA platform (EPA US, 2022).

Reference

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