**Evaluating Biomagnification of Organohalogenated Pollutants in Juvenile Fish from Estuarine Nurseries: A Field Methodological Comparison**

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# Abstract

This study examines the **trophodynamics of organohalogenated pollutants** from three families in **juvenile *Solea solea*** from a temperate estuary (Seine, France). A total of **31 perfluoroalkyl substances (PFASs)** and **19 polychlorinated biphenyl (PCB) congeners** were measured in juvenile soles and their benthic prey.

Among the **16 Young-of-the-Year juvenile *Solea solea* (G0) samples** and **50 prey samples** collected across **three taxonomic groups** (Bivalvia, Arthropoda, Polychaeta), **PCBs were by far the predominant contaminant family** in both fish and prey (ΣPCBs = 465.20 ng/g dry weight in soles and 292.50 ng/g in prey). **PFAS contamination was approximately 10 times lower.** Among PFAS compounds, **L-PFOS** was the most abundant, representing ~50% of total PFAS contamination.

A high **inter-species variability** in contamination levels was observed, with no clear explanation related to taxonomic group or feeding mode. However, **bivalves and soles appeared to be more contaminated with PCBs than polychaetes and crustaceans**. Conversely, **bivalves were less contaminated with PFASs than the other taxa**, suggesting that **bivalves have a particular capacity for bioaccumulation** compared to other organisms.

**Biomagnification factors (BMFs)** were assessed using two approaches: (1) **comparing contaminant concentrations in individual prey species and soles**, (2) **comparing contaminant concentrations in the overall diet and soles.**

The first method indicated that **PCBs exhibited limited biomagnification** (mean BMF for ΣPCBs = 1.8), with **BMFs ranging from 1 to 2 across nearly all prey species**. **PFASs showed stronger bioaccumulation (mean BMF = 4.0)**. Across all contaminant families, **BMFs varied significantly between species**, and higher-than-average BMFs were observed in a limited number of species, differing among contaminant families.

The second method, which incorporates diet composition, **yielded slightly lower BMFs** for PCBs (by ~+0.1 to +1), except for **CB149**. However, for PFASs, **BMFs based on diet composition were significantly higher (+1 to +7) than those calculated for individual prey species**, indicating **strong biomagnification for all PFASs**—particularly for **L-PFOS (BMF = 7 instead of 1.5) and PFNA (BMF = 7.5 instead of 1.3)**.

These findings suggest that **disregarding diet composition when assessing BMFs may result in an underestimation of biomagnification potential, particularly for the emerging contaminant family studied, PFASs**.

# Keywords

Seine estuary; important nursery ground ; flatfish juvenile; *Solea solea*; high organic chemical contamination; PCBs; PFASs; BMF, TMF, assimilation efficiency

# Highlights

# Revues

* Integrated Environmental Assessment and Management
* Science of the Total Environment

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# Introduction

Exploited marine ecosystems are inherently complex, characterized by significant variability in biophysical conditions, species-specific biology and behaviour, and intricate trophic interactions. Beyond these ecological dynamics, anthropogenic pressures — such as fishing — can further disrupt habitats and ecosystem functioning, especially in densely populated coastal regions (Halpern *et al.*, 2008, 2012). Coastal and estuarine zones serve as critical nursery habitats for numerous fish species, offering unique environmental conditions essential for feeding, growth and survival during early life stages (Le Pape *et al.*, 2003; Rochette *et al.*, 2010). These nurseries are fundamental to the completion of life cycles and the renewal of fish populations, and are therefore recognized as essential fish habitats (Beck *et al.*, 2001). This is particularly true for many flatfish species, including the common sole (*Solea solea*).

However, nursery habitats do not always provide optimal conditions for juvenile fish development, and their degradation can markedly affect key biological functions such as survival, growth performance, and the capacity of individuals to contribute to stock replenishment. Both controlled experimental studies and *in situ* comparisons between estuaries have shown that environmental quality — particularly levels of chemical contamination — has direct influence on nursery function, as reflected in juvenile growth and survival rates (Gilliers *et al.*, 2006; Amara *et al.*, 2007). Due to their hydrodynamic and physicochemical properties, estuarine nurseries are especially vulnerable to contamination. These environments often act as long-term sinks for a wide range of micropollutants, with sediments serving as the main reservoir for persistent contaminants (Ridgway and Shimmield, 2002).

Over time, a scientific consensus has developed regarding the environmental risk assessment of chemicals that meet the PBT criteria — persistence (P), bioaccumulation (B), and toxicity (T) (Matthies *et al.*, 2016). Environmental persistence is linked to the structural stability of compounds, both in the environment and within organisms. Bioaccumulation, the process by which chemicals accumulate in living organisms from their surroundings, depends on several chemical, biological and ecological factors, including bioaccessibility (i.e., the fraction of a compound that can be released from its matrix), bioavailability (i.e., the fraction that is readily available for uptake), biotransformation into other compounds, and affinity for biological molecules (e.g., lipids), all of which affect uptake and accumulation at the cellular level (Gobas *et al.*, 2009). Toxicity refers to adverse biological effects of accumulated chemicals and can vary widely depending on the organism’s developmental stage, with early life stages being particularly sensitive to endocrine-disrupting effects (Matthies *et al.*, 2016). Therefore, evaluating the bioaccumulation of persistent organic pollutants (POPs) in early-life stages juvenile fish — during growth and maturation stages — feeding on benthic organisms in contact with contaminated estuarine sediments is essential for understanding environmental risks in these critical nursery areas.

In the context of chemical registration and regulation, bioaccumulation assessments rely on a range of methods and metrics, each associated with inherent uncertainties that complicate decision-making processes (Gobas *et al.*, 2009). Among the most commonly used indicators are the bioconcentration factor (BCF), which reflects chemical uptake from water, and the biomagnification factor (BMF), which indicates uptake through dietary exposure (Arnot et al., 2023). For hydrophobic organic chemicals (HOCs), such as POPs, diet is considered the primary route of bioaccumulation. However, experimentally derived BMFs—typically based on controlled feeding experiments using spiked pellets—often fail to accurately reflect field conditions, where organisms are exposed to contaminants through both dietary and aqueous pathways, as well as from complex and variable sources (Schäfer et al., 2015). Furthermore, bioaccumulation is influenced by a multitude of abiotic factors (e.g., site-specific water and sediment quality parameters such as pH or clay content), biotic factors (e.g., organism growth rate, lipid content, age, and sex), and ecological factors (e.g., trophic level, feeding habits) (Liu, Haffner and Drouillard, 2010; Deribe *et al.*, 2011; Van Ael *et al.*, 2013; Li *et al.*, 2018; Masset *et al.*, 2019). These variables underscore the complexity of reliably estimating bioaccumulation in natural systems (van der Oost, Beyer and Vermeulen, 2003). Consequently, there is a growing consensus that field-derived data should play a more prominent role in evaluating the bioaccumulation potential of chemicals and in assessing the chemical status of aquatic ecosystems (Schäfer et al., 2015). In this context, this study aimed to evaluate and compare field-based approaches for assessing biomagnification potential of several POPs in juvenile fish under natural exposure conditions.

To achieve this, common sole and their prey were sampled from the Seine Estuary, a key nursery ground for Eastern English Channel sole stocks (Riou, Le Pape and Rogers, 2001; Rochette et al., 2010). This estuary is subject to nursery habitat degradation due to substantial contaminant inputs from both local intra-estuarine sources and upstream watershed discharges (Gilliers et al., 2006; Le Pape et al., 2007; Rochette et al., 2010). We first focused on legacy POPs, specifically polychlorinated biphenyls (PCBs), for which contamination levels (Dargnat and Fisson, 2010) and bioaccumulation properties have been extensively documented (van der Oost, Beyer and Vermeulen, 2003; Borgå et al., 2005; Arnot and Gobas, 2006). However, for many other regulated or emerging contaminants, available data remain sparse or entirely lacking (Daughton, 2005; Covaci et al., 2011), despite their potential contribution to the ecotoxicological effects observed in estuarine organisms chronically exposed to complex and dynamic contaminant mixtures (Poisson et al., 2011). Among these compounds, we also included (i) substances recently listed under the Stockholm Convention on POPs, such as perfluorooctane sulfonate (PFOS, since 2009); (ii) substances currently under evaluation for inclusion in the same convention, including certain per- and polyfluoroalkyl substances (PFASs). These contaminants are known to be particularly prevalent in the Seine Estuary, notably PFOS (Munschy et al., 2013), and PFASs more broadly (Labadie and Chevreuil, 2011; Munoz et al., 2018). Due to their strong bioaccumulative properties and widespread detection in aquatic organisms, including fish, Environmental Quality Standards (EQS) have been established for these compounds in wild fish tissues (Directive 2013/39/EU, 2013). Improving our understanding of their environmental fate and bioaccumulation in natural ecosystems is therefore essential for both scientific advancement and the effective management of aquatic environmental quality.

The present study aims improve our understanding of bioamplification — the process by which the concentration of a chemical compound in a predator exceeds that found in its prey, primarily due to dietary accumulation (Gobas and Morrison, 2000). This mechanism is hypothesized to be predominant for these contaminant families studied here, and it explains why these chemicals are bioaccumulative despite their relatively low bioconcentration factors (i.e. accumulation from water) (Schäfer *et al.*, 2015; Fremlin, Elliott and Gobas, 2025). To assess dietary exposure and bioaccumulation potential of these compounds, two commonly used indicators are the BioMagnification Factor (BMF) and Trophic Magnification Factor (TMF) (Fremlin, Elliott and Gobas, 2025). The simplest method to estimate BMFs involves calculating concentration ratios between predator (in this case, juvenile common sole) and each of its prey species within the nursery area. Although this approach is rapid, it does not account for the relative dietary contribution of each prey item. A more refined method incorporates dietary weighting, whereby contaminant concentrations are adjusted according to the relative abundance of each prey item in the predator’s diet (Mackay and Fraser, 2000), offering a more accurate estimate of dietary exposure. Additionally, a third method employed in this study involves estimating TMFs based on nitrogen stable isotope levels (δ¹⁵N), which provide ecologically relevant information on trophic position and thus offer an integrated perspective on dietary exposure. For all three approaches, special care was taken to align with recent methodological recommendations for BMF and TMF calculation (Fremlin, Elliott and Gobas, 2025). Comparisons between these biomagnification estimates were conducted using PCB congeners with a 100% detection frequency to avoid bias due to left-censored data. In the case of PFAS compounds, for which non-detects were common, several innovative strategies were applied to address left-censored data and improve the robustness of the analysis.

Materials and methods

## Dataset

### Sampling

The dataset used in this study originates from the CHOPIN and CAPES research projects and is based on sampling of first-year juvenile common sole (*Solea solea*) and their main prey species in the Seine estuary (northern France) (see Table 1). A detailed description of the sampling methodology is provided in Supplementary Information, Section 1.1.

Sixteen composite samples of first-year sole were prepared by pooling individuals of similar size to obtain sufficient material for chemical analysis. A total of 50 samples were collected from 16 targeted prey species, based on their presence in sole’s diet (see next section) and sufficient biomass for chemical analysis. These encompassed three different taxonomic groups and three distinct feeding modes. Bivalves were the most represented group, with nine targeted species represented across 27 samples. The crustacean group included only two species, totaling 7 samples. Five annelid species were included, represented by 16 samples, and exhibited the widest range of trophic strategies: including suspension feeding, surface deposit feeding, and omnivory.

Table 1 - List of sampled benthic prey species and *Solea solea* analyzed for contaminants, along with their taxon, feeding mode, corresponding sample size (N = number of sampled pools) and proportion analyzed for each chemical family, and percentage in stomach contents.

| Species | Taxon | Feeding mode | Sample size | %N PCB | %N PFAS | % stomach |
| --- | --- | --- | --- | --- | --- | --- |
| *Abra alba* | Bivalve | Suspension feeder | 3 | 100% | 100% | 0.59 |
| *Cerastoderma edule* | Bivalve | Suspension feeder | 5 | 60% | 100% | 1.51 |
| *Corbula gibba* | Bivalve | Suspension feeder | 1 | 100% | 0% | - |
| *Donax vittatus* | Bivalve | Suspension feeder | 1 | 100% | 0% | - |
| *Ensis directus* | Bivalve | Suspension feeder | 1 | 0% | 100% | - |
| *Limecola balthica* | Bivalve | Deposit feeder | 5 | 100% | 80% | 0.06 |
| *Nucula nitidosa* | Bivalve | Deposit feeder | 1 | 100% | 0% | 0.03 |
| *Scrobicularia plana* | Bivalve | Deposit feeder | 6 | 100% | 100% | - |
| *Spisula subtruncata* | Bivalve | Suspension feeder | 4 | 100% | 75% | 0.06 |
| *Corophium volutator* | Crustacean | Deposit feeder | 1 | 100% | 100% | 11.46 |
| *Crangon crangon* | Crustacean | Omnivore | 6 | 100% | 100% | 1.59 |
| *Hediste diversicolor* | Polychaete | Omnivore | 2 | 100% | 100% | - |
| *Lagis koreni* | Polychaete | Deposit feeder | 7 | 57% | 100% | 0.53 |
| *Lanice conchilega* | Polychaete | Deposit feeder | 1 | 100% | 100% | 0.18 |
| *Nephtys sp.* | Polychaete | Omnivore | 4 | 100% | 100% | - |
| *Owenia fusiformis* | Polychaete | Suspension feeder | 2 | 100% | 100% | 0.44 |
| *Solea solea* | Actinopterygii | Omnivore | 16 | 100% | 94% | NA |

### Diet composition

Due to the limited knowledge available in the literature regarding the diet of early-stage G0 sole, stomach content analyses were carried out on 255 individuals to identify the main prey species and assess their relative abundance. The prey items found in the stomachs were used to reconstruct the typical diet composition of first-year juvenile sole. Dietary proportions were calculated as the number of prey items per taxonomic group, divided by the total number of identified prey individuals. The resulting diet composition consisted of 51% arthropods, 33% annelids, and 16% molluscs. The diet composition by taxonomic class is represented in Figure 1 and the detail by species can be found in the SI section 2.1.Haut du formulaire

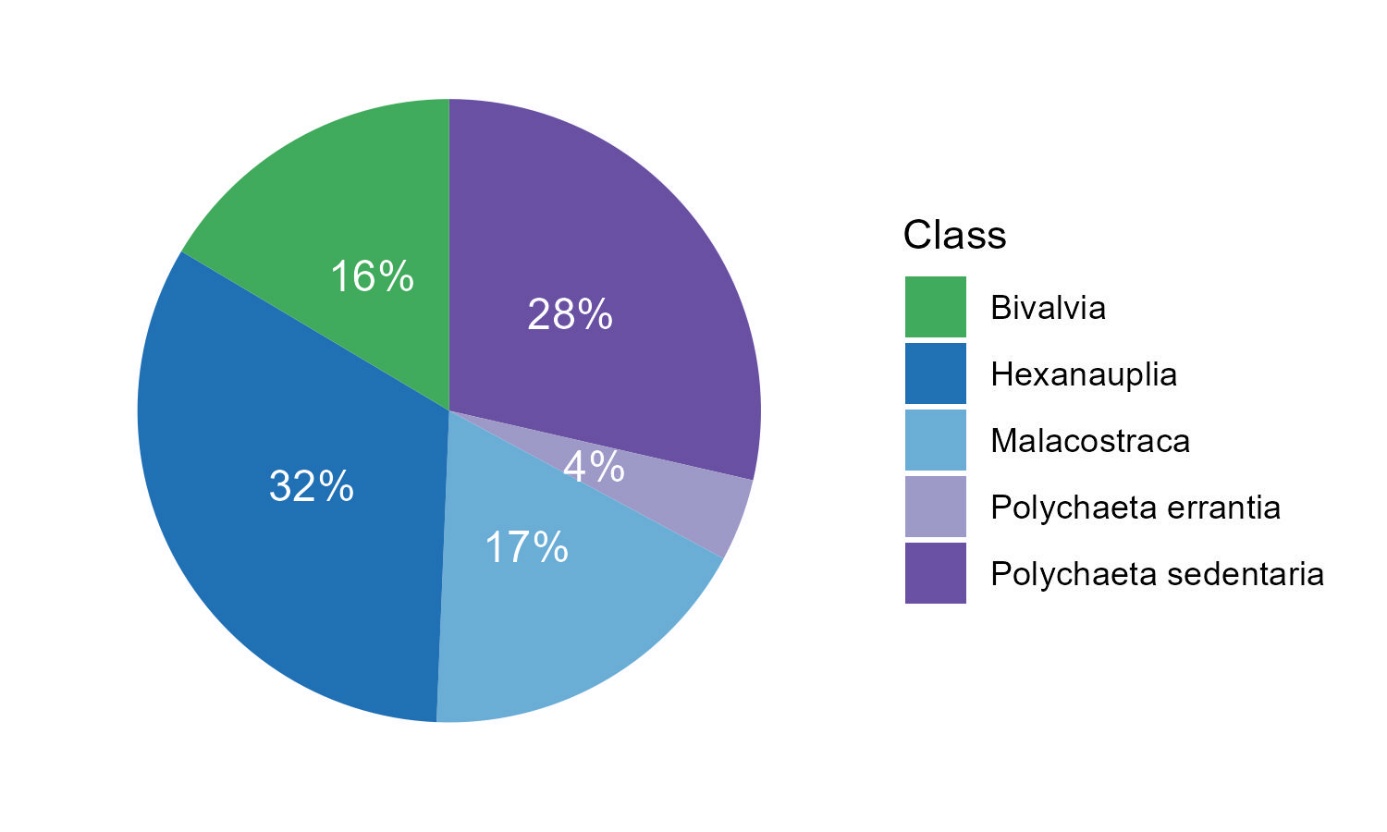


Figure 1 – Diet composition in taxonomic classes for those above 2% in the diet.

### Chemical

This study focused on two families of organohalogenated compounds: PCBs and PFASs (see SI section 1.2.1 for the presentation of use and reglementation). More precisely, concentrations of 19 PCB congeners and 26 PFAS components were determined in sole and prey samples (see SI section 1.2.2 for chemical acronyms, full names and CAS numbers). Technical details on sample preparation, chemical analysis, as well as quality assurance and control for contaminants, are provided in the SI section 1.2.3. Despite a significant sampling effort, the three contaminant families could not be systematically analysed across all samples due to occasionally insufficient material quantities. Therefore, the sample sizes for each species and chemical family are reported in Table 1 (above).

As the objective of this study was to investigate trophic transfer mechanisms, subsequent analyses were restricted to chemical compounds that were both detected and sufficiently prevalent in soles and their prey. Only PFAS compounds quantified simultaneously in more than 20% of the samples in both groups were retained for subsequent analyses. Based on this criterion, 12 out of the 26 PFAS compounds were selected for further analysis (Table 2, the list of the 26 compounds can be found in SI section 1.2.2). Additionally, the 19 PCB congeners were included (Table 3).

For each analysed sample, moisture content was measured to standardize contamination levels in ng/g dry weight (dw), ensuring comparability across species despite differences in moisture content. Additionally, lipid content was quantified as it is preferable to express contamination levels on a lipid weight basis (ng/g lipid weight lw), given the lipophilic properties of PCB (Conder *et al.*, 2012). Indeed, lipid content of test organisms can have major impact on bioaccumulation of these compounds (Schäfer *et al.*, 2015). Other biochemical markers—such as polar and neutral lipids, total proteins, or albumin—which could further assist in normalizing PCB and PFAS concentrations or refining trophic positioning (Fremlin, Elliott, and Gobas, 2025), were not analyzed due to limited sample material. The total concentrations (in ∑ ng/g for each normalisation ww, dw and lw) for each contaminant family were calculated to express their relative proportions, enabling the determination of contamination profiles within each sample.

We also studied the relationship between structural and chemical properties and bioaccumulation. For PFASs, the end-group nature and the number of carbon atoms in perfluoroalkyl chain were considered (Table 2) as they could influence bioaccumulation (Labadie and Chevreuil, 2011; Munoz *et al.*, 2017). For PCBs, we were interested in their partitioning octanol-water coefficients (log KOW) (Mackay *et al.*, 2016) and number of chlorine substitutions (Loizeau, Abarnou and Ménesguen, 2001).

Table 2 – List of PFASs searched in benthic preys and sole juveniles’ samples along with their end-group nature, chemical formula, perfluoroalkyl chain length (n carbon atoms) and percent of detection in prey and sole samples.

|  |  |  |  | Detection % | |
| --- | --- | --- | --- | --- | --- |
| Group | Compound | Formula | nC | preys | sole |
| PFCA | PFOA | F3C(CF2)6CO2H | 8 | 20 | 27 |
|  | PFNA | F3C(CF2)7CO2H | 9 | 91 | 100 |
|  | PFDA | F3C(CF2)8CO2H | 10 | 64 | 93 |
|  | PFUnDA | F3C(CF2)9CO2H | 11 | 69 | 27 |
|  | PFDoDA | F3C(CF2)10CO2H | 12 | 98 | 100 |
|  | PFTrDA | F3C(CF2)11CO2H | 13 | 62 | 87 |
|  | PFTeDA | F3C(CF2)12CO2H | 14 | 53 | 40 |
| PFSA | L-PFOS | F3C(CF2)6SO3H | 8 | 98 | 100 |
|  | Br-PFOS | F3C(CF2)7SO3H | 8 | 27 | 87 |
| FOSA | FOSA | F3C(CF2)7SO2NH2 | 8 | 91 | 73 |
| N-Alkyl FOSAA | EtFOSAA | F3C(CF2)7SO2N(C2H5)CH2CO2H | 8 | 60 | 73 |
| FTS | 8:2-FTSA | F3C(CF2)7(C2H4)SO3Na | 8 | 71 | 40 |

Table 3 - List of PCB congeners searched in benthic preys and sole juveniles’ samples. Group reflects the number of chlorine substitutions for PCBs. Octanol-water partition coefficients (logKow) are from (Hawker & Connell, 1988).

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
|  |  |  | Detection % | |
| Compound | **Group** | **log Kow** | preys | sole |
| CB 28 | Tri-CB | 5.67 | **100** | **100** |
| CB 31 | 5.67 | **100** | **100** |
| CB 44 | Tetra-CB | 5.75 | **100** | **100** |
| CB 49 | 5.63 | **100** | **100** |
| CB 52 | 5.84 | **100** | **100** |
| CB 101 | Penta-CB | 6.38 | **100** | **100** |
| CB 105 | 6.65 | **100** | **100** |
| CB 110 | 6.48 | **100** | **100** |
| CB 118 | 6.74 | **100** | **100** |
| CB 128 | Hexa-CB | 6.74 | **100** | **100** |
| CB 132 | 6.58 | **100** | **100** |
| CB 138 | 6.83 | **100** | **100** |
| CB 149 | 6.67 | **100** | **100** |
| CB 153 | 6.92 | **100** | **100** |
| CB 156 | 7.18 | **100** | **100** |
| CB 170 | Hepta-CB | 7.11 | **100** | **100** |
| CB 180 | 7.36 | **100** | **100** |
| CB 187 | 7.17 | **100** | **100** |
| CB 194 | 7.8 | **100** | **100** |

## Magnification factors calculations

Recent workshops have highlighted the importance of using field-derived trophic magnification factors (TMFs) and biomagniﬁcation factors (BMFs) to assess bioaccumulation potential of chemicals (Conder *et al.*, 2012). These approaches rely on the assumption that dietary intake is the primary route of contaminant exposure, as chemical concentrations may increase in the gastrointestinal tract during digestion—a valid assumption for the chemical families investigated in this study (Gobas, Zhang and Wells, 1993).

The bioaccumulation potential of chemicals can be estimated using several field-based approaches, which we aimed to compare in the present study. In each case, bioaccumulation was assessed using appropriately normalized concentrations: lipid-normalized values (in ng/g lw) for PCBs, and dry weight-normalized values (in ng/g dw) for PFASs (Conder *et al.*, 2012).

### BMF

Estimating BMFs requires prior confirmation of trophic relationships between predator and prey species (Fremlin, Elliott, and Gobas, 2025). In our study, such relationships were established through stomach content analysis, which provided direct evidence of feeding links.

One empirical method for estimating BMFs involves calculating the ratio of contaminant concentrations in predators to those of their food (Conder *et al.*, 2012). The simplest commonly used approach applies this calculation individually for each prey species consumed by the predator—referred to here as the predator-prey BMF (BMFPP). In this approach, a separate BMF value is computed for each prey type (Schäfer *et al.*, 2015). Although straightforward and easy to implement, this method does not consider the relative contribution of each prey item to the predator’s diet.

A more accurate reflection of the theoretical definition of BMF can be achieved by incorporating the relative abundance of prey in the predator’s diet — referred to in this study as diet-based BMF (BMFD). This method requires knowledge of both contaminant concentrations in each prey item and the proportional composition of the predator's diet. In our study, these dietary proportions were determined through stomach content analysis (see Section 2.1.2). As taxonomic resolution was limited to higher-level groupings, the analysis focused on three main prey categories: bivalves, polychaetes, and crustaceans.

To ensure a robust comparison between BMFPP and BMFD, we limited the analysis to PCB congeners with a 100% detection frequency, thereby avoiding bias due to non-detects. Additionally, CB153 was used as a benchmark compound for evaluating method performance, as it is known to consistently biomagnify across most food chains.

To explore patterns in BMF values across different PCB congeners (CB), we examined their relationships with physicochemical properties such as logKow (Hawker & Connell, 1988) using a second-order polynomial regression. This was performed with geom\_smooth() function from the ggplot2 package, employing the “lm” method). We also considered the effect of the number of chlorine substitutions.

The predator-prey BMF (BMFPP) for each CB in the was calculated as:

where is the lipid-normalized concentration of the CB in the sole, and is the corresponding concentration in a given prey species (Conder *et al.*, 2012).

The diet-based BMF (BMFD) was calculated as:

where is the concentration of the PCB congener in prey group *p* (bivalves, polychaetes, or crustaceans), and is the relative proportion of that prey group in the sole's diet (Conder *et al.*, 2012).

To explore variability in BMF estimates, we considered three scenarios: (1) Median BMF: using median concentrations for each prey group and for sole; (2) Minimum BMF: using third-quartile concentrations for prey and first quartile for sole; (3) Maximum BMF: using first-quartile concentrations for prey and third quartile for sole.

To enable a direct comparison between BMFPP with BMFD, we computed the median values of the median, minimum, and maximum BMF estimates across all prey species.

### TMF or TL-BMF

While the BMF approach is straightforward to implement, its predictive power can be limited by its dependency on the specific prey species considered, with varying sensitivity depending on the calculation method used. In this context, the trophic magnification factor (TMF) approach offers a more integrative and holistic perspective. TMFs represent the average biomagnification potential of a chemical across an entire food web by accounting for multiple trophic interactions. This method helps mitigate confounding effects related to species-specific differences biotransformation and elimination capacities, which may otherwise obscure the chemical’s overall biomagnification behaviour within the ecosystem (Conder *et al.*, 2012).

However, the estimation of a robust TMF requires comprehensive sampling across the full spectrum of trophic levels within the food web (Conder *et al.*, 2012)—an approach not feasible in our case due to incomplete food web characterization and limited sample size (Borgå *et al.*, 2012). Therefore, as an alternative, we calculated trophic level–normalized BMFs (TL-BMF), which are methodologically analogous to TMFs and allow direct comparison (Conder *et al.*, 2012). The TL-BMF for each predator-prey pair was computed using the following equation:

Where denotes the concentration of the PCB congener in the predator (*sole*) and in prey , and is the corresponding trophic level.

Trophic levels for all sampled species were derived from stable nitrogen isotope ratios (), measured in sole individuals (n = 67; CAPES project) and benthic prey (n = 102 from CAPES and n = 8 from CHOPIN). Trophic levels were calculated using the following equation:

where λ is the trophic position of the baseline organism, is the nitrogen isotopic value of the baseline species (set to the minimal measured value of in our dataset), is the trophic enrichment factor. Constants are based on values from (Munoz *et al.*, 2017), who studied comparable ecosystems (i.e., and ). Median δ¹⁵N values per species were used for TL estimation.

In addition to the predator-prey TL-BMF (TL-BMFPP), an analogous diet-based TL-BMF (TL-BMFD) was also computed. This approach integrates prey proportions in the predator’s diet and considers both the weighted mean prey concentration () and their trophic levels () (Conder *et al.*, 2012). The corresponding equations are:

and

Where denotes the relative contribution of prey taxon to the predator’s diet, based on stomach content analysis

## Accounting for left-censored concentrations

It is essential to report whether contaminant concentrations in samples fall below the limit of quantification (LOQ) and to appropriately address these nondetect (left-censored) values when estimating biomagnification factors. A commonly used but simplistic method for handling such data involves substituting nondetects with arbitrary values — typically half the LOQ (Borgå *et al.*, 2012). However, this approach can introduce bias and uncertainty. Therefore, current recommendations advocate for the evaluation of uncertainty associated with substitution methods and encourage the use of statistically robust alternatives (Helsel, 2006; Fremlin, Elliott and Gobas, 2025). Tools such as the NADA package (Julian and Helsel, 2021) in R (R core team, 2025) are specifically designed for the analysis of censored environmental data are supposed to offer more reliable solutions.

In this study, several methods for handling nondetects in PFAS concentration data were compared in the calculation of diet-based BMFs (i.e., using the same methodology as previously described for PCBs). Summary statistics for contamination levels within each dietary taxon (i.e., first quartile, median, and third quartile) were estimated under the following scenarios: (1) “Uncensored” - using only quantified values; “Half-LOQ substitution” – replacing with one-half the LOQ; (3) “Log-normal model” - fitting a log-normal distribution to the detected values and predicting censored values; (4) “Kaplan-Meier method” – using the *cenfit* function in the NADA package; (5) “Regression Order Statistic (ROS)” – using the *ros* function in the NADA package). These last two methods directly estimate summary statistics for contaminant levels while explicitly accounting for censored data. They were particularly well-suited for the characteristics of our dataset, as they offer greater robustness in the context of small sample sizes and low detection frequencies. The Kaplan-Meier method is a non-parametric approach that doesn’t require assumptions about the underlying data distribution, whereas the ROS method assumes a log-normal distribution – a common characteristic of environmental contaminant concentrations.

# Results

## Contamination data

### Total levels of contamination

Figure 2 displays contaminant concentrations (ng/g dw), expressed as the sum per chemical family, in soles and the three prey taxa. Results are summarized hereafter and detailed results by contaminant family, taxonomic group, feeding mode, and species are presented in Supplementary Information section 2.1.

Among soles and their prey, PCBs are by far the most predominant contaminant family, followed by PFASs. Prey exhibit significantly lower PCB contamination than soles, approximately by a factor of two, while PFAS levels are similar between prey and soles.

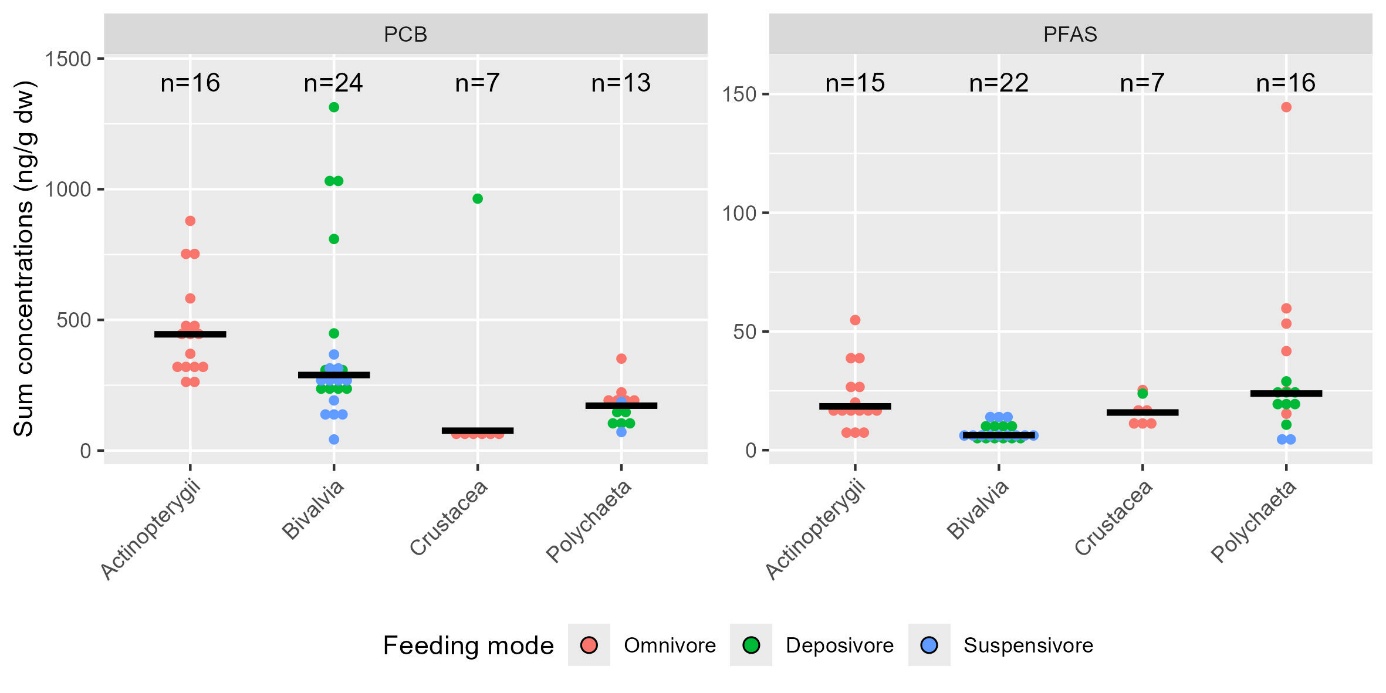


Figure 2 - Sum of contaminant families in soles and three benthic prey taxa (ng/g dw). Group means are shown in black. Colors indicate feeding modes.

Overall, contaminant concentrations displayed considerable variability within species, taxonomic groups, and feeding modes. Among prey, the effects of taxon and feeding mode differed depending on the contaminant family. For the PCB family, bivalves tend to have distinct contamination levels, largely driven by *Limecola balthica*, which exhibits elevated PCB concentrations without any obvious explanatory factor. Interestingly, contamination levels in *Crangon crangon* are lower than in other omnivores and crustaceans. Regarding PFASs, both bivalves and suspension feeders are significantly less contaminated than other taxa and feeding modes.

### Relationship with lipid content for PCB

Examining the relationship between lipid content and total concentrations for PCB family, a significant positive correlation was observed (Spearman’s test: p-value = 0.01, ρ = 0.43). See SI section 2.2 for graphs and more detailed results.

While the positive relationship with lipids for PCBs is expected (Conder et al., 2012), some species deviate from this pattern. For instance, *Crangon crangon* exhibits highly variable lipid levels without corresponding increases in PCB contamination, whereas *Limecola balthica* maintains relatively stable lipid content despite a wide range of PCB concentrations.

### Average contamination profiles

The average contamination profiles of soles and prey are shown in Figure 3.

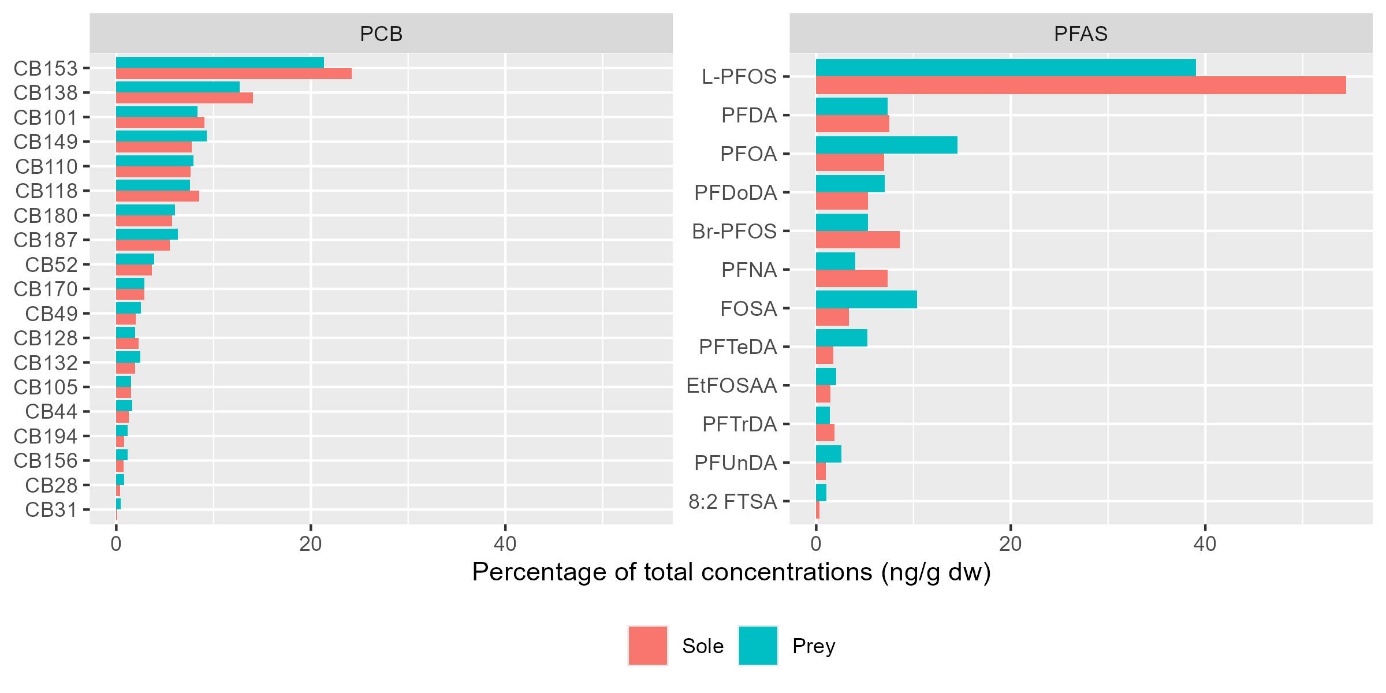


Figure 3 - Average contamination profiles of soles and prey for PCBs (left) and PFASs (right).

For the PCB family, soles and prey exhibit similar profiles, both dominated by CB153, which accounts for 24% of the total in soles and 21% in prey. The second most abundant congener, CB138, is present in slightly lower proportions — 14% in soles and 13% in prey.

In contrast, for PFAS, L-PFOS is by far the most prevalent compound, representing 55% of the total PFAS concentration in soles and 39% in prey. The subsequent most abundant compounds have much lower proportions and differ between the two groups: in prey, PFOA (15%) and FOSA (10%) are most prominent, while in soles, Br-PFOS (9 %), PFDA (7 %), and PFNA (7%) follow L-PFOS.

## BMFs for PCBs

### Predator-prey BMF (BMFPP)

Predator-prey BMF estimates for each prey are presented for the congener CB153 in Figure 4 (see SI section 3.1 for detailed results by PCB congener). The bioaccumulative potential of PCBs varies markedly among prey taxa. For instance, within the crustacean group, the BMFPP estimate based on *C. volutator* suggests strong biodilution (0.44[0.35-0.49]), whereas the estimate based on *C. crangon* indicates pronounced bioaccumulation (6.41[4.99-9.25]). Moreover, for the majority of species (8 out of 15), the variability in BMFPP estimates spans both sides of the threshold value of 1, encompassing both biodilution and biomagnification. As a result, it is difficult to draw definitive conclusions regarding the overall bioaccumulation potential of PCBs based solely on these estimates.

The relationship between logKow, the number of chlorine substitutions, and BMF estimates under the BMFPP scenario is shown in Figure 5. A non-significant polynomial relationship is observed between logKow and BMFPP values (R² = 0.23, p-value = 0.125). For the vast majority of congeners (16 out of 19), as well as for the sum of PCBs (1.05[0.77-1.15]), the range of BMFPP estimates spans both below and above 1, reflecting substantial variability and precluding firm conclusions regarding their biomagnification behavior. However, BMFPP values for CB28 and CB31 (tri-chlorinated biphenyls), and for CB132 and CB149 (hexa-chlorinated biphenyls), clearly indicate biodilution (BMFPP max < 1). Conversely, the sum of PCBs and 8 out of the 19 congeners show median BMFPP values exceeding 1, indicating biomagnification. The median BMFPP estimate for CB153 — a benchmark compound for evaluating method performance due to its consistent biomagnification across food chains — also indicates biomagnification (1.1 [0.83–1.28]).

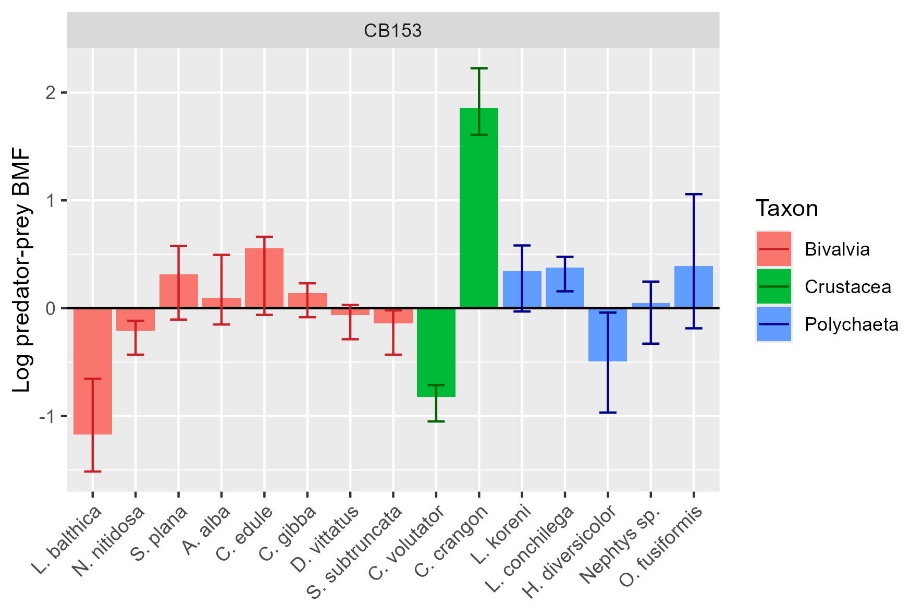


Figure 4 – Log-transformed minimum, median, and maximum BMF estimates (BMFPP scenario) for the CB153 congener across species. Taxa are color-coded; values > 0 indicate biomagnification.

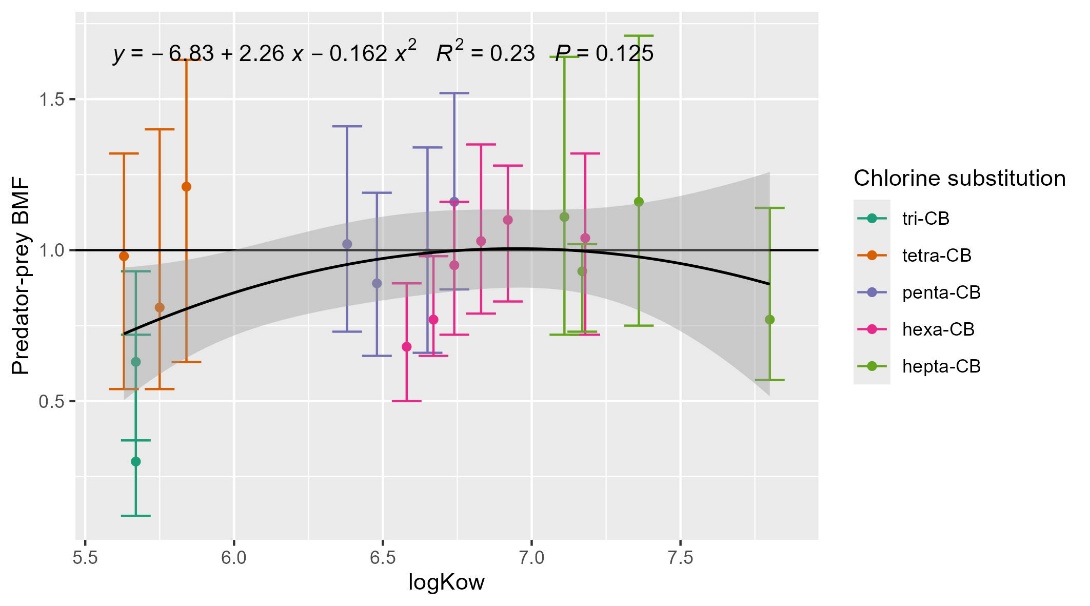


Figure 5 – Median BMF values of min, median, and max estimates by species, under BMFPP scenario in relation to logKow and chlorine substitution. The black curve represents a global quadratic regression (second-order polynomial) fitted across all chlorination classes.

### Diet-based BMF (BMFD)

The relationship between logKow, the number of chlorine substitutions, and BMF estimates under the BMFD scenario is shown in Figure 6. A significant polynomial relationship is observed between logKow and BMF values (R² = 0.45, p-value = 0.009). For most congeners, median BMFD values exceed 1, indicating biomagnification, with the exception of those containing only three chlorine atoms (CB28 and CB31). BMFD estimates for CB153 clearly indicate biomagnification (2.01 [1.24–2.68]). Similar conclusions were drawn for the sum of PCBs (1.72[1.07-2.41]) and 6 of the 18 other congeners: CB101, CB110, CB118, CB128, CB138, and CB149, which are penta- and hexa-chlorinated biphenyls). However, for the remaining congeners — except for CB31 which indicates biodilution (0.31 [0.11-0.93]) — the range of BMFD estimates spans both below and above 1, reflecting a wide variability that prevents firm conclusions regarding their specific biomagnification behavior.

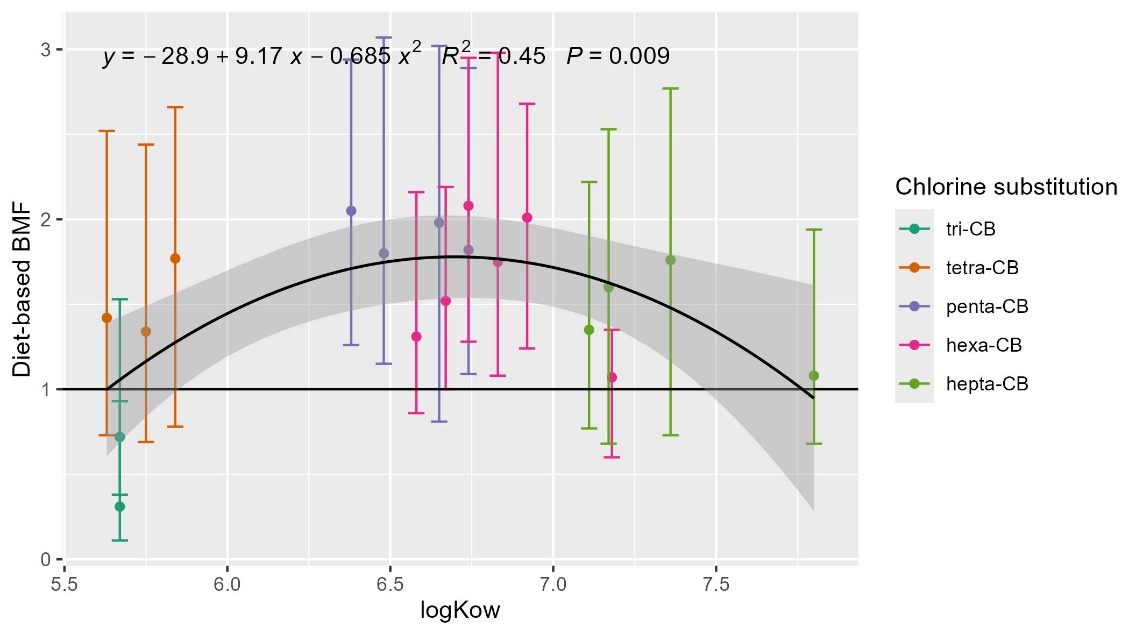


Figure 6 - BMF estimates (min, median, max) under BMFD scenario in relation to logKow and chlorine substitution. The black curve represents a global quadratic regression (second-order polynomial) fitted across all chlorination classes.

### Predator-prey trophic level–normalized BMF (TL-BMFPP)

The trophic positions used in this scenario, inferred from species-specific δ¹⁵N isotopic signatures, are presented in SI section 3.2. TL-BMFPP estimates for each prey species, based on the CB153, are shown in Figure 7 (see SI section 3.3 for congener-specific results). Due to insufficient material, isotopic measurements could not be obtained for *D. vittatus*, and consequently, no TL-BMFPP could be derived for this species. For *C. crangon*, TL-BMFPP was equal to zero, as its median trophic level exceeded that of the sole. Among the remaining 13 species, 8 exhibited median TL-BMFPP values above 1, suggesting potential biomagnification. However, the variability of these estimates spans both below and above the threshold of 1 (except for *Lanice conchilega*), preventing definitive conclusion. Five species clearly showed evidence of biodilution.

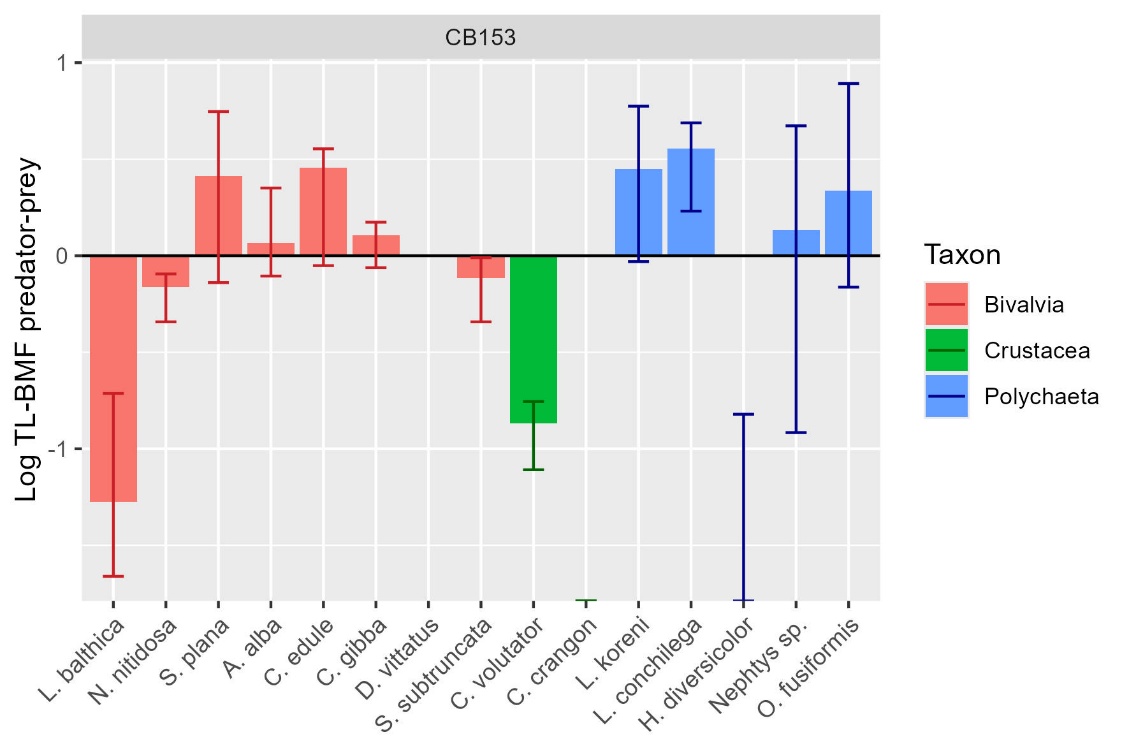


Figure 7 - Log-transformed minimum, median, and maximum BMF estimates (TL-BMFPP scenario) for the sum of PCB congeners across species. Taxa are color-coded; values > 0 indicate biomagnification.

The relationship between logKow, the number of chlorine substitutions, and BMF estimates under the TL-BMFPP scenario is shown in Figure 8. A non-significant polynomial relationship was observed between logKow and TL-BMFPP values (R² = 0.27, p-value = 0.079). For 13 out of 19 congeners, median TL-BMFPP values were below 1. However, except for five congeners whose variability clearly indicated biodilution — CB28, CB31, CB132, CB149 and CB187 — the TL-BMFPP ranges for the other congeners, including CB153 (1.09[0.78-1.3]) and the sum of PCBs (1.05[0.71-1.31]) extended both below and above 1, reflecting high variability and uncertainty in biomagnification patterns.

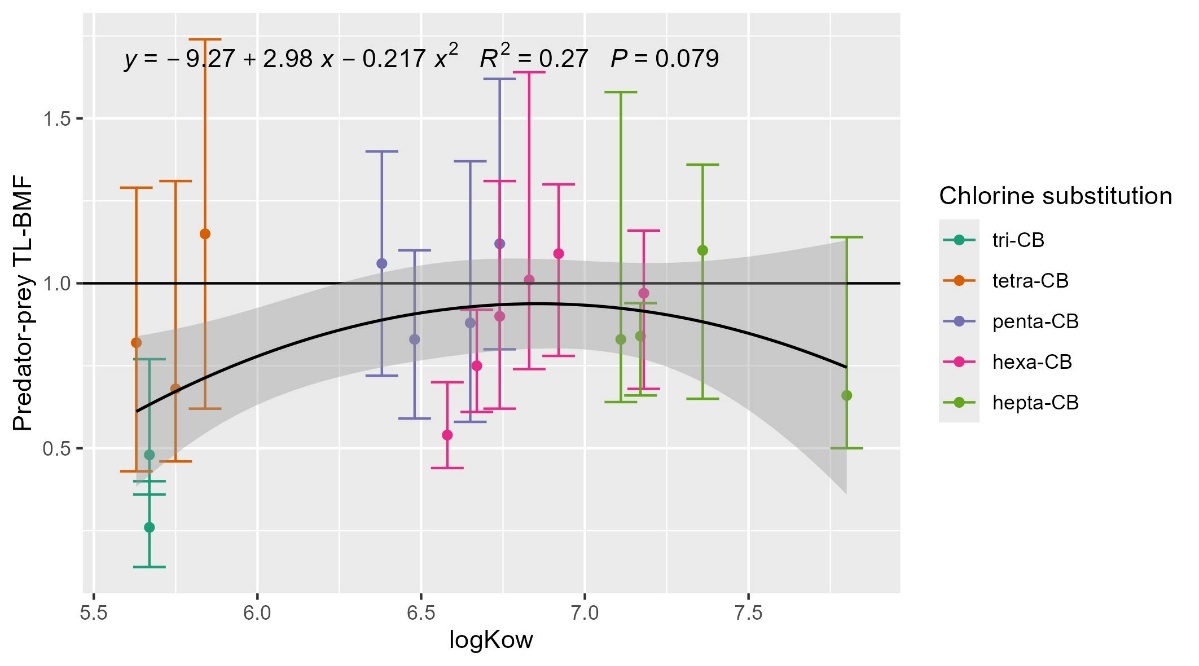


Figure 8 - Median BMF values of min, median, and max estimates by species, under TL-BMFPP scenario in relation to logKow and chlorine substitution. The black curve represents a global quadratic regression (second-order polynomial) fitted across all chlorination classes.

### Diet-based trophic level–normalized BMF (TL-BMFD)

Diet-based TL-BMFs were estimated using the same dietary composition as in the BMFD scenario. The relationship between logKow, the number of chlorine substitutions, and BMF estimates under the TL-BMFD scenario is shown in Figure 9. A significant polynomial relationship was observed between logKow and BMF values (R² = 0.42, p-value = 0.014). For 18 out of 19 congeners median TL-BMFD values exceeded 1, indicating biomagnification, except for one containing only three chlorine atoms (CB28 and CB31). Strong evidence of biomagnification was found for 7 out of 19 congeners, including CB153 (7.78 [1.90–17.95]), as well as for the sum of PCBs (4.94 [1.23-13.22]). For the other 12 congeners, however, TL-BMFD estimates ranged both below and above 1, reflecting high variability and limiting the certainty of their biomagnification potential.

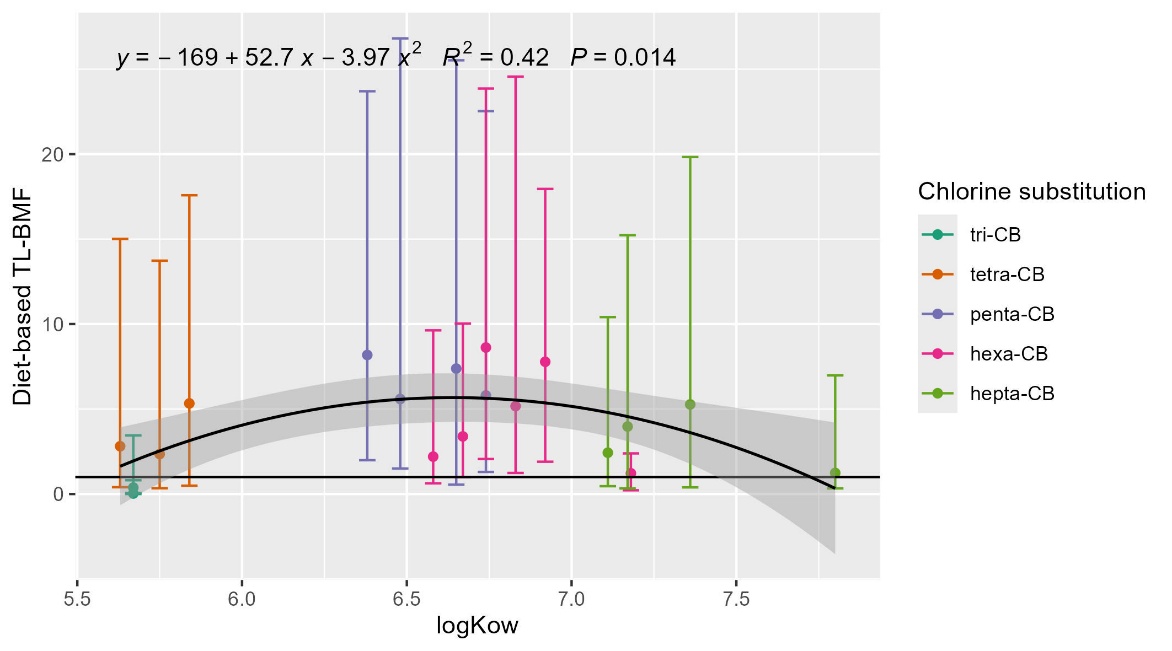


Figure 9 - BMF estimates (min, median, max) under BMFD scenario in relation to logKow and chlorine substitution. The black curve represents a global quadratic regression (second-order polynomial) fitted across all chlorination classes.

### BMF methodological comparison

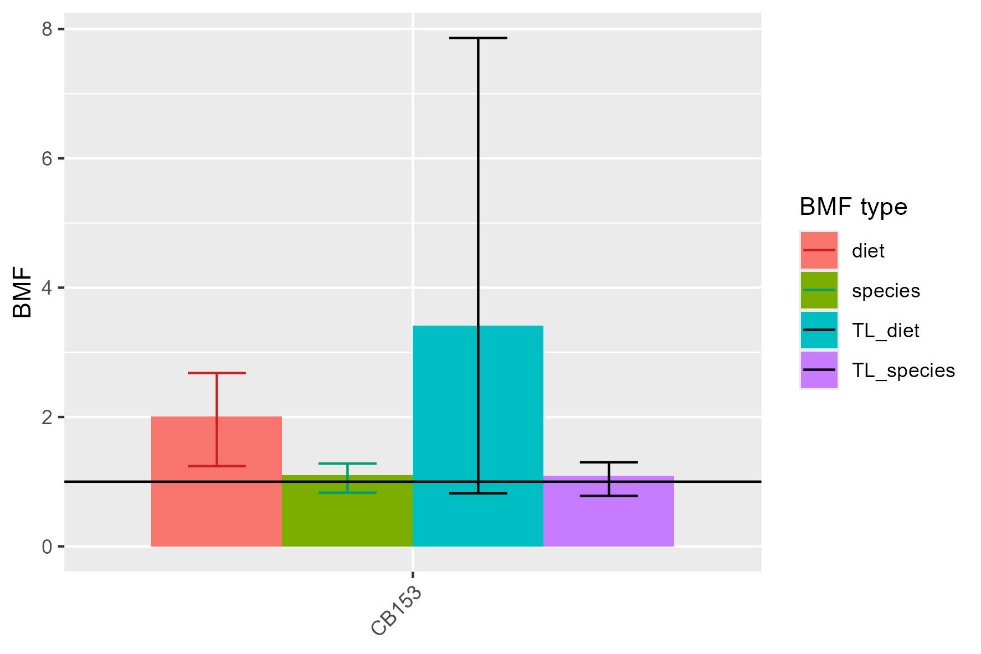


Figure 10 – Influence of BMF computation scenario on BMF estimates for CB153.

Summary statistics of BMF estimates for each computation method are presented in Figure 10, with detailed values by PCB congener and for the sum of PCBs available in SI section 3.4. Results indicate BMF estimates show greater variability in scenarios based on diet composition. Notably, only the diet-based scenario clearly supports bioaccumulation of CB153, with an estimated BMF of 2.01 [1.24-2.68], while estimates for the other methods span both above and below the bioaccumulation threshold of 1. Diet-based scenarios also yield higher median BMFs values compared to predator-prey scenarios, suggesting that the dierary contamination level (6666 [5224-8058] ng/g lw) is lower than the median contamination level observed in preys species (10 754 [922-59 074] ng/g lw).

## Diet-based BMFs for PFASs

### Influence of nondetect handling methods on BMF estimates

Regardless of the method used to estimate contamination levels in dietary taxa, the ranges of diet-based BMF estimates overlapped across all PFAS compounds. However, for certain compounds, the “uncensored” scenario (based solely on detected values) yielded median BMF estimates that differed from those obtained with the other approaches. Slight differences were observed for PFDA, PFTrDA, FOSA, EtFOSAA, and Br-PFOS. More substantial discrepancies occurred for PFUnDA, PFTeDA, and 8:2 FTSA — compounds with detection frequencies below 50% in the sole group — where the uncensored scenario indicated biomagnification, whereas the other methods consistently suggested strong biodilution.

The two substitution-based scenarios (i.e., half-LOQ and log-normal imputation) produced comparable results. Similarly, the Kaplan-Meier method yielded results consistent with the substitution approaches, except for eight compounds with detection frequencies below 70% in either sole or prey groups, for which summary statistics could not be estimated. Finally, the ROS method produced results that were generally consistent with other approaches, except for PFUnDA, PFTeDA, Br-PFOS, and 2:2 FTSA — compounds with detection frequency below 50% in at least one group — where median BMF estimates indicated a reduced effect of either biodilution or biomagnification.

Le L-PFOS, utilisé comme référence de composé biomagnifié parmi les PFAS, a bien une valeur médiane au dessus de 1 pour la méthode diet (1.48 [0.89-3.16]). Celle-ci est plus élevée pour la méthode species (4.94 [0.47-23.90]). Dans les deux cas la variabilité rend difficile de conclure sur des propriétés de bioaccumulation.

Les BMF moyens calculés sur la base de couples sole-benthos varient entre 1,3 et 2,3 pour les PCB et 1,5 et 7,6 pour les PFAS ; ils sont très variables pour l’ensemble des composés considérés. Néanmoins, pour la plupart des contaminants considérés dans le Tableau 4, les BMF moyens ou médians sont supérieurs à 1, ce qui suggère la bioamplification de ces COH en bon accord avec des résultats antérieurs obtenus sur la Gironde pour les PCB et les PFAS (Munoz et al., 2017 ; Lauzent, 2017).

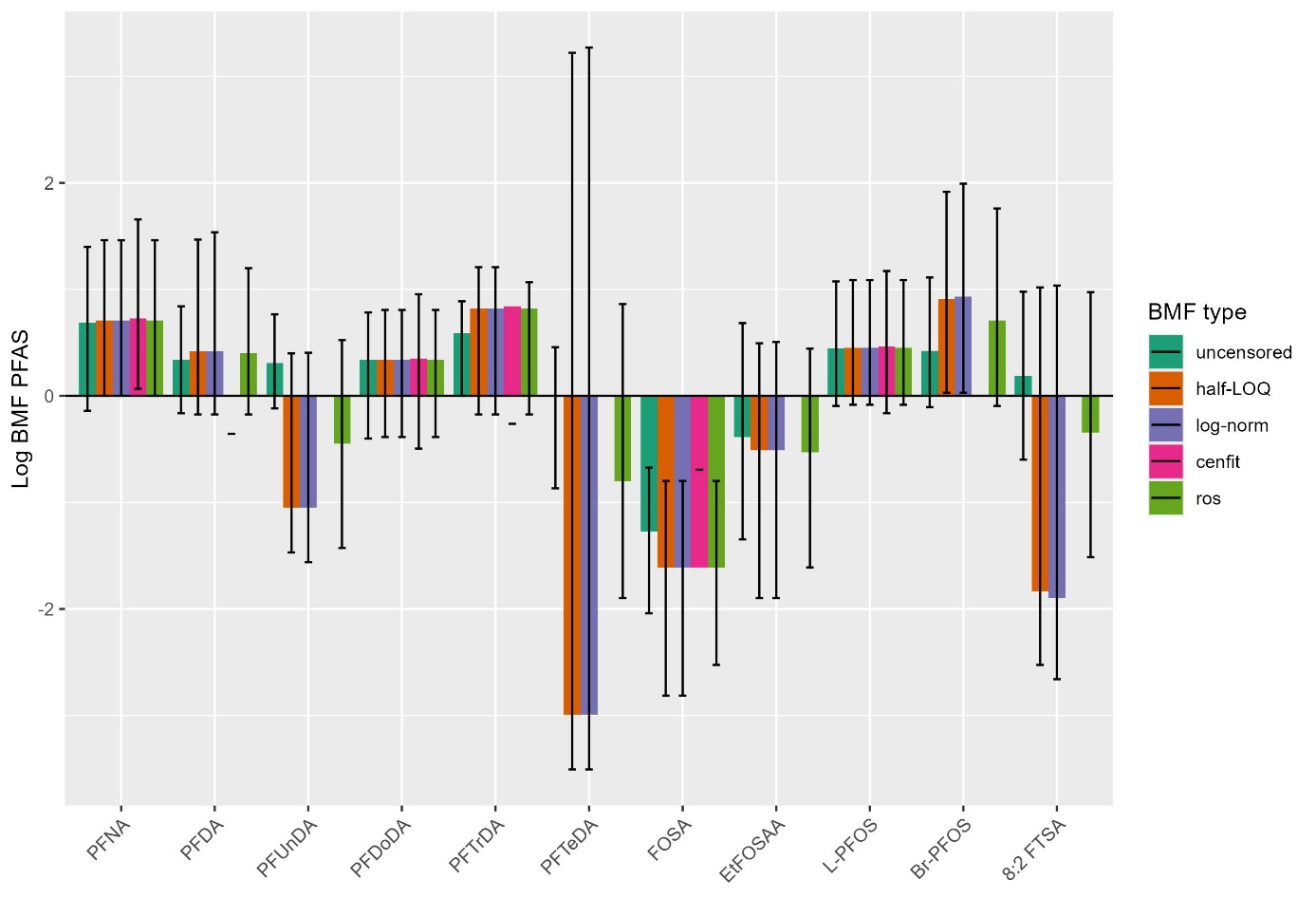


Figure 11 - Comparison of diet-based BMF estimates for PFASs using five methods for addressing nondetects.

### Influence of structural properties on BMF estimates with ROS method

For a given perfluorinated chain length of eight carbon atoms, perfluoroalkyl sulfonates (PFSAs) exhibit higher median BMF estimates, indicating biomagnification, whereas the other sub-families show patterns consistent with biodilution. In contrast, within the perfluoroalkyl carboxylic acids (PFCAs) sub-family, BMF estimates show no clear trend with increasing chain length.

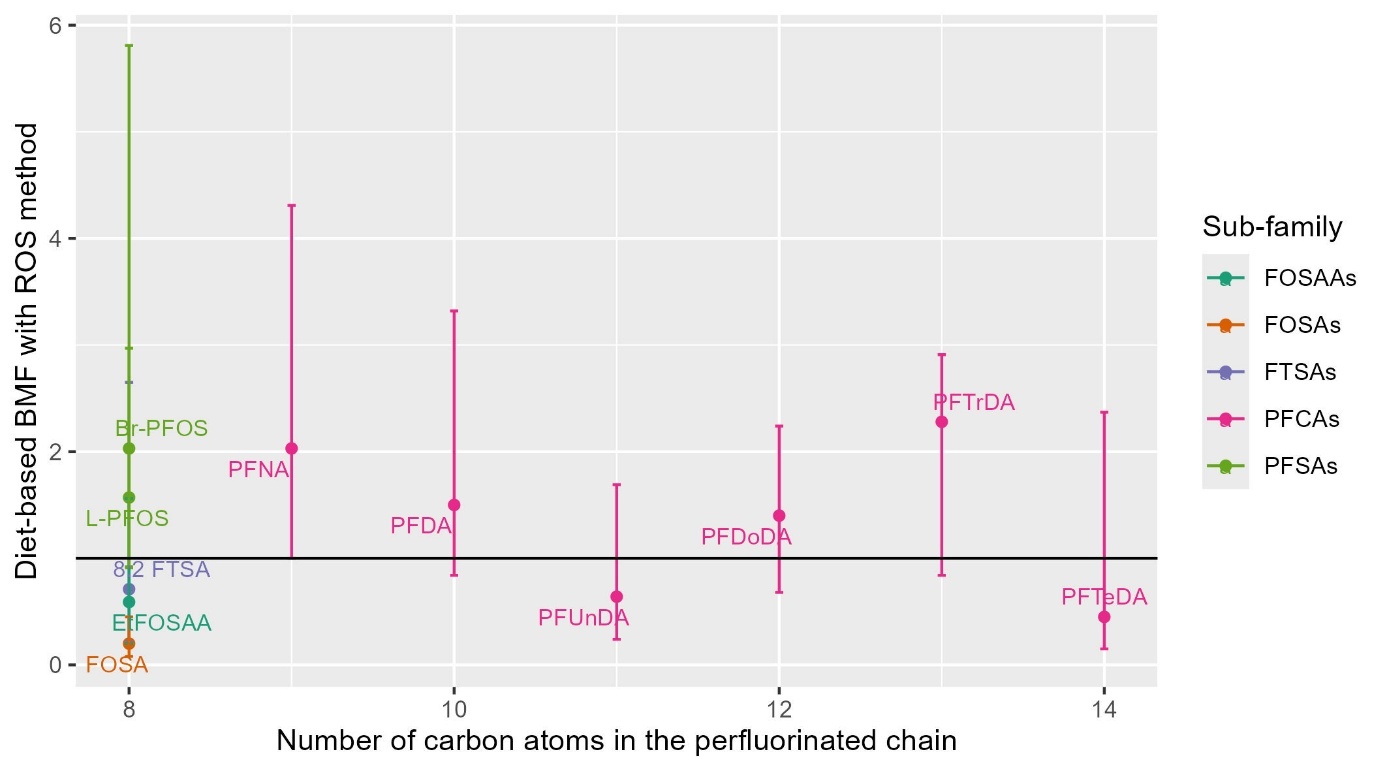


Figure 12 – Comparison of diet-based BMFs for PFASs estimated using the ROS method, by perfluorinated chain length and sub-family.

# Discussion

## Composition du régime alimentaire utilisée

* Groupe taxonomique des hexanauplia représente 32% du régime alimentaire (dont 31.5% d’harpacticoida) mais n’a pas du tout été dosé chimiquement.
* Toutefois, les proportions estimées ici étant basées sur un nombre d’individus présents dans les estomacs, cette forte proportion est surestimée car les copépodes du groupe hexanauplia ont une masse individuelle très faible en comparaison de celles des espèces du groupe des Malocostraca (espèces majoritairement retrouvées : 11.5% de Corophium volutator et 1.6% de Crangon crangon). En effet les proportions recherchées pour estimer la composition du régime alimentaire devraient être estimées en masse pour étudier la bioaccumulation.
* Etant donné la forte variabilité de contamination entre espèces au sein d’un groupe taxonomique, la composition du régime alimentaire par espèces dosées en contaminant serait importante pour estimer la composition du régime alimentaire.
  + Toutefois, les contenus stomacaux permettent difficilement de différencier les espèces de bivalves car la plupart du temps seuls les siphons sont retrouvés (11.5% de bivalves non identifiables dans les contenus stomacaux, 4 espèces dosées en contaminants sur 6 n’ont pas été identifiables).
  + Dans le cas des arthropodes malacostraca de très nombreuses espèces ont été identifiées mais seules deux d’entre elles ont été dosées en contaminant alors que certaines étaient en proportions équivalentes à celle de Crangon crangon (1.03% pour Diastylis bradyi)
  + Dans le cas des polychètes, l’espèce majoritaire dans les contenus stomacaux 24.0% de Polydora ciliata, n’a pas été dosées en contaminants. Celles dosées ont des proportions inférieures à 1% dans le régime alimentaire (0.4% pour Owenia fusiformis, 0.53% pour Lagis koreni, 0.18% pour Lanice conchilega) voir non identifiées (Hediste diversicolor et Nephtys sp.)
* Les BMF PCB plus élevés pour les scenario diet-based sont due à la concentration du diet est inférieure à la concentration médiane des proies. En effet le régime alimentaire contient une majorité de crustacés alors que ces derniers sont le groupe taxonomique le moins contaminé et qu’ils ne représentent que 7 points de mesure dans le calcul de la médiane des espèces calculées sur 44 mesures.

## Niveaux et profiles de contamination

La comparaison avec d’autres études est souvent critiquée pour la grande variabilité entre études, dues aux différences dans les caractéristiques des écosystèmes, la biologie et l’écologie des organismes ainsi qu’aux variations de conception des études (Fremlin, Elliott and Gobas, 2025). Dans la mesure du possible, les comparaisons avec la littérature ont donc été faites sur des espèces identiques, en privilégiant les milieux estuariens du continent Européen.

Globalement, on observe donc une forte variabilité des niveaux de contamination au sein des trois familles et le taux de lipides, le taxon, le mode trophique et l’espèce semblent être des facteurs peu explicatifs. Toutefois, les bivalves semblent avoir une bioaccumulation des PFAS et des isomères d’HBCDD différente des autres taxons. Le bivalve *Limecola balthica* semble également avoir une bioaccumulation des PCBs différente des autres bivalves, sans facteur explicatif.

### Niveaux généraux entre les familles

* Comparaison PCB et PFASs même zone + TMF Olifants River Basin, subtropical, South Africa (Verhaert *et al.*, 2017)

Les concentrations mesurées dans les juvéniles de *Solea solea* et leurs proies révèlent une prédominance des PCBs par rapport aux PFASs et HBCDDs. Ces résultats sont cohérents avec des études antérieures menées en milieux estuariens où les PCBs restent les contaminants organohalogénés les plus abondants, notamment en raison de leur persistance et de leur historique d’usage intensif (GIP Seine-aval, 2024). La contamination en PFASs et HBCDDs, bien que faible, est néanmoins préoccupante, car ces composés sont considérés comme des contaminants émergents ayant des propriétés de bioaccumulation encore mal comprises (Loi *et al.*, 2011, p. 201). Des recherches récentes ont mis en évidence la présence de PFAS dans les écosystèmes aquatiques, soulignant leur persistance et leur potentiel de bioaccumulation (Cara *et al.*, 2022).

L’abondance relative des contaminants au sein des proies suit des tendances qui pourraient être expliquées par leurs propriétés physico-chimiques. En effet, les bivalves ont montré une bioaccumulation plus marquée des HBCDDs, ce qui pourrait s’expliquer par leur mode de filtration favorisant l’adsorption de ces composés lipophiles (REF). Inversement, les polychètes et crustacés semblent moins affectés par l’accumulation des PCBs et PFASs, ce qui pourrait être lié à des différences de capacité de métabolisation (REF) ou de biodisponibilité des contaminants dans le sédiment (fonction de la conductivité de l’eau et clay content) (Van Ael *et al.*, 2012).

### Niveaux et profils pour les PCB

* Estuaire de la Gironde, France 2005 (Bodin *et al.*, 2014) et 2012 (Lauzent, 2018)
* Estuaire de Scheldt, Belgique (Van Ael *et al.*, 2012) (check : intro, results – reste : 2 derniers paragraphes discussion)
* Belgian north sea western Scheldt estuary (Voorspoels *et al.*, 2004)

***Une corrélation au taux de lipides peu marquée***

(Van Ael *et al.*, 2012, 2013) : “It is previously stated that the lipid content of an organism has an inﬂuence on the accumulation of pollutants (Gewurtz *et al.*, 2006; Deribe *et al.*, 2011) Dans notre étude, les concentrations en somme de PCB sont significativement (lm p-value = 0.007) mais faiblement corrélées (R²= 0.37) aux taux de lipides uniquement pour les soles.

***Un profil classique avec cependant un CB180 plus faible***

* Les profils de contamination en PCBs étaient similaires entre soles et benthos, comme observé dans le Scheldt estuary in Belgium (Van Ael *et al.*, 2012) et l’estuaire de la Gironde en France (Lauzent, 2018)
* Au sein des PCBs, les 2 composés majoritaires sont le CB153 et le CB138 avec une proportion de 24% et 14% dans les soles, ce qui est cohérent avec la littérature :
  + pour l’estuaire de la Gironde en 2014 en France (23% et 16%) (Bodin *et al.*, 2014)
  + pour l’estuaire de la Gironde en 2017 en France (37% et 23%) (Lauzent, 2018)
  + pour l’estuaire du Scheldt en Belgique (17% et 10%)(Van Ael *et al.*, 2012).
  + Le 3ème composé est le CB101 (8%) et non le CB180 (6%) qui était le plus abondant pour l’estuaire de la Gironde en 2017 en France (4% et 30%)(Lauzent, 2018). Cependant, les niveaux sont sensiblement identiques pour l’estuaire du Scheldt en Belgique (6.3% et 5.6%) (Van Ael *et al.*, 2012)

Pourquoi un CB180 faible ?

***Des concentrations dans le benthos et les soles plus fortes que dans d’autres estuaires Européens***

* Comparaison PCB poisson (European sea bass) en estuaires Européens (Schnitzler *et al.*, 2011) : « The Scheldt and the Seine are still among the most contaminated estuaries in Europe. Each region presented their speciﬁc contamination patterns reﬂecting different sources due to the input of the respective rivers.”

Compared to the results from other studies and regions, POP concentrations in biota from the present study were relatively high.

***Benthos :***

On retrouve le pattern Bivalves > Polychaete > Crustacés dans la littérature(Voorspoels *et al.*, 2004; Van Ael *et al.*, 2012; Lauzent, 2018)

* Bivalves are known for their ability to concentrate lipophilic substances. They exhibit a low bio-transformation capacity and as ﬁlter feeders, they achieve high ﬁltration rates and accumulate chemicals from water, as well as from particulate material (Dörr and Liebezeit, 2009).
* ces auteurs suggèrent que les plus faibles contaminations des crustacés viendraient d’une nature plus pélagique que les autres espèces, vivants plus au contact du sédiment (Oh, Hartnoll and Nash, 2001).

La comparaison de la contamination en PCB du benthos avec la littérature montre des niveaux similaires ou plus forts dans notre étude. Seule la comparaison avec les bristle worms et

la moule bleue (Mytilus edulis) montre des niveaux plus faibles mais, dans cette étude, ces niveaux étaient anormalement élevés en comparaison de la littérature.

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| Present study | ∑PCB ng/gww  *Lipid %* | Other studies | ∑PCB ng/gww  *Lipid %* | Zone | Ref | Comparaison  Contamination et lipides |
| Benthos | 30[8-175] | Benthos | 9[1-27] | Gironde 2012 | (Lauzent, 2018) | 4-5x plus fort |
| 53[3-1690] | Scheldt | (Van Ael *et al.*, 2012) | 1.8x plus faible (10x pour les max) |
| *Crangon crangon* | 13[8-26]  *2.9%[2.2-4.5]* | Crangon crangon | 3[1-5] | Gironde 2012 | (Lauzent, 2018) | 5x plus fort |
| Crangon crangon | 11[3-40]  *0.7[0.5-0.9] %* | Scheldt | (Van Ael *et al.*, 2012) | [] similaire vs taux de lipides 4x supérieurs |
| crevette blanche | 15[13-17] | Gironde 2012 | (Lauzent, 2018) | similaire |
| Polychaete | 32[15-49]  *1.5%[1.0-6.1]* | Nereis | 3.4[3.2-3.6] | Gironde 2012 | (Lauzent, 2018) | 10x plus fort |
| bristle worms | 53[12-106]  *1.3[1.0 -1.6]%* | Scheldt | (Van Ael *et al.*, 2012) | Niveau similaire mais max [] 2 fois plus faible VS *max % 4 fois plus forts* |
| *Scrobicularia plana* | 30[24-44] | *Scrobicularia plana* | 10[4-16] | Gironde 2012 | (Lauzent, 2018) | 3x plus fort |
| Bivalves | 30[10-175]  *2.3%[0.8-3.7]* | Huitre | 18[10-26] | Gironde 2012 | (Lauzent, 2018) | 1.7x plus fort (max 6.7x) |
| *Mytilus edulis* | 782[287-1690]  *2.7[2.0-3.7] %* | Scheldt | (Van Ael *et al.*, 2012) | [] 26x plus faible VS *% similaires* |

***Des concentrations plus fortes dans nos G0 que dans les adultes de la littérature***

Tableau 6 - (6 PCB indicateurs de l’ICES)

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| Present study | ∑PCB  *%lipid* | Other studies | ∑PCB  *%lipid* | Zone | Ref | Comparaison |
| 19 PCB | 101[55-211] ng/gww  *4%[2-7]* | 33 PCB | 39[9-213] ng/gww  *0.7% [0.4-1.3]* | Scheldt | (Van Ael *et al.*, 2012) | [] 3x plus fort  *% 5-6 fois plus fort* |
| Individu entier | 13 ng/gww | Gironde 2012 | (Lauzent, 2018) | [] 10x plus fort |
| filet | 513 ng/gww | [] 5x plus faible |
| 19 PCB | 445[250-879] ng/g dw  *4%[2-7]* | filet | 75 ng/gdw  *3%* | Gironde 2005 | (Bodin *et al.*, 2014) | [] 6x plus forte vs % similaires |
| foie | 1177 ng/gdw  *26%* | [] 2.5 fois plus faible mais % 6.5 fois plus faible |
| individu entier | 57[31-123] ng/gww |  | 17[4-97] ng/gww | Scheldt | (Van Ael *et al.*, 2012) | [] 4 fois plus forte |
| filet | 75 ng/gww | (European Commission, 2022) | | Plus élevé pour 4 poissons /!\ filet |
|  |  |  |  |  |  |  |

Les niveaux de contamination des soles apparaissent plus élevés en PCB dans notre étude en Seine, par rapport à d’autres estuaires européens : 3 à 4 fois plus élevés qu’en estuaire de Scheldt (Van Ael *et al.*, 2012), bien connu pour être l’un des plus contaminés en OHC en Europe depuis 40 ans ; 6 à 10 fois plus élevés qu’en estuaire de la Gironde (Bodin *et al.*, 2014; Lauzent, 2018). Cette contamination plus forte de la Seine a été communément considérée depuis les années 80 cependant l’étude de (Bodin *et al.*, 2014) mettait en évidence des contaminations du flet 2 à 5 fois plus faibles en Seine (1986-1988) qu’en Gironde (2005). Cette tendance ne semble donc pas être observée sur la sole mais cela pourrait également refléter un changement de niveau de contamination de la Seine depuis la fin des années 80.

Les **concentrations en poids frais dans les soles** (107[55-211 ng/gww pour 19 CB, =61[31-123] ng/gww pour les) sont:

et de la Gironde (=2100-7700ng/g dw)

Aussi, la valeur moyenne des sommes de PCBs de 465 ng/g dw de notre étude, est encadrée par les valeurs dans le muscle (75 ng/gdw) et dans le foie (1177 ng/gdw) en estuaire de la Gironde, ce qui est attendu étant donné les différences de taux de lipides entre ces tissus (muscle : 3%, foie : 26%) et l’individu entier dans notre étude (4[2-7]%). Cette concentration plus élevée dans le foie que dans le muscle est bien connue pour les HOC (Schäfer *et al.*, 2015). Toutefois, si on compare les concentrations en poids de lipides, la valeur moyenne de 11334 ng/g lw de notre étude est deux fois plus élevée que la contamination du foie en estuaire de la Gironde (5201 +/- 1922 ng/g lw) indiquant des niveaux de contamination plus élevés en estuaire de Seine une fois la différence de taux de lipides prise en compte. En effet, les taux de lipides varient en fonction du stade de vie et il est donc recommandé d’étudier les concentrations normalisées par les lipides.

Il convient de préciser que notre étude porte sur les G0, premiers stades de vie alors qu’en Seine et en Scheldt des adultes ont été mesurés (32.9 et 15.4 cm, respectivement). Cette différence de contamination entre les stades de vie et taille de poisson est bien connue et peut supplanter les effets spatio-temporels (Schäfer *et al.*, 2015). Elle a par exemple été observée chez les poissons pour les PFAS (Lescord *et al.*, 2015; Munoz *et al.*, 2017). De plus faibles concentrations chez les jeune adultes peut possiblement s’expliquer par une modification des taux métaboliques, un changement de régime alimentaire ou un phénomène de dilution par la croissance. Cette différence de contamination entre les stades de vie a été observée chez les poissons pour les PFAS (Lescord *et al.*, 2015; Munoz *et al.*, 2017).

The PCB family is by far the most predominant in soles, with an average total PCBs concentration of 465.20 ng/g dw, compared to 22.34 ng/g dw for PFASs and 0.15 ng/g dw for HBCDD isomers. This hierarchy is also observed in prey; however, PCB levels are lower at 292.50 ng/g dw, PFAS levels are similar at 23.55 ng/g dw, and HBCDD isomer concentrations are higher at 9.57 ng/g dw.

## Biomagnification des contaminants

### Des différences entre familles

L’examen des BMF suggère un potentiel de bioamplification significatif pour la plupart des HOC étudiés ici, notamment PCB et PFAS.

Les PCBs, bien qu’accumulés par les soles, présentent des BMFs relativement faibles (~1.8), ce qui est conforme à d'autres études sur des poissons benthiques démontrant une biomagnification limitée de ces composés en raison de leur biotransformation dans les tissus des prédateurs (Gobas *et al.*, 1999; Borgå *et al.*, 2004, 2005).

En revanche, les PFASs présentent des BMFs plus élevés (jusqu’à 7.5 pour certains composés), indiquant une forte capacité de transfert trophique. Ce résultat est en accord avec les observations récentes sur la bioaccumulation des PFASs dans les écosystèmes aquatiques (Houde *et al.*, 2006; Conder *et al.*, 2008; Ng and Hungerbuhler, 2014; Cara *et al.*, 2022). Pour une meilleure estimation des BMFs, la normalisation des concentrations par le taux de protéine aurait pû être mesurée, les PFAS étant connus pour se lier aux protéines (Conder *et al.*, 2012; Schäfer *et al.*, 2015). Cette normalisation par les lipides polaires est aujourd’hui recommandée pour ces substances dites « ionisantes » dans l’estimation des BMFs et TMFs (Kelly *et al.*, 2009; Fremlin, Elliott and Gobas, 2025).

L’analyse des isomères d’HBCDD met en évidence une différence marquée entre les proies et les soles, avec une dominance de l’α-HBCDD chez les poissons et du γ-HBCDD dans les proies. Ce phénomène est bien documenté et résulte de la sélectivité métabolique des organismes aquatiques, qui tendent à convertir les isomères γ et β en α, ce dernier étant plus stable et bioaccumulable (Ruan *et al.*, 2018).

### Une variabilité inter-spécifique marquée des BMFs prédateur-proie

Nos résultats montrent des variations significatives des facteurs de biomagnification (BMFs) en fonction des groupes taxonomiques et des familles de contaminants. La variabilité des BMF est cependant forte, en lien avec la variabilité de la contamination du benthos. Le choix des espèces étudiées pour le calcul de ces BMFs a donc une influence sur le résultat des études (Schäfer *et al.*, 2015).

Cette variabilité n’a pu être expliquée par le taxon (hypothèse de différences métaboliques) ou le régime alimentaire (hypothèse de différentes voies de contamination – importance de l’apport par l’eau pour les filtreurs ou du sédiment pour les espèces enfouies), bien que ces facteurs soient connus pour influencer les niveaux de contamination (REF).

Les différences physiologiques entre les espèces ont été approchées via leur groupe taxonomique mais des variations pourraient exister au sein de ces taxons. L’âge et les conditions physiologiques peuvent également faire varier la bioaccumulation et expliquer une partie de la variabilité intra-spécifique (Schäfer *et al.*, 2015).

Dans le cas de notre étude, les contaminations en PCB sont deux à trois fois plus fortes pour *Limecola balthica* en comparaison des autres bivalves déposivores. Toutefois, le fait que ces variations soient dû à la variabilité spatio-temporelle ne peut pas être écarté ici. En effet, la contamination est également connue pour varier dans l’espace et dans le temps dans les estuaires (Van Ael *et al.*, 2012; Olisah, Adams and Rubidge, 2021; Pitacco *et al.*, 2021; Fremlin, Elliott and Gobas, 2025) mais n’a pas pu être étudiée ici, faute d’échantillonnage suffisant pour toutes les espèces. Le transfert du sédiment vers le benthos peut également être lié à des facteurs locaux du sédiment, comme le taux d’argile, ou de l’eau, comme la conductivité dépendante de la position dans l’estuaire (Van Ael *et al.*, 2012).

Failing to account for all these factors could affect the monitoring of POPs and the determination of BMFs. In field studies, where local variations can be significant, variability may pose challenges and raise concerns about the accuracy of field-based BMFs. Pour étudier l’effet spatio-temporel, une étude complémentaire, affranchie des facteurs biotiques, pourrait porter sur l’utilisation d’échantillonneurs passifs standardisés artificiels (Schäfer *et al.*, 2015) ou biologiques, comme l’utilisation du vers endobenthic Nereis diversicolor, présent dans les estuaires et proie de la sole (Durou *et al.*, 2007). Toutefois, seule, cette méthodologie ne représenterait pas forcément un bon indicateur du facteur de BMF dans la mesure où des variabilités inter-spécifiques sont grandes entre les proies. L’utilisation des contenus stomacaux pour estimer le régime alimentaire des soles dans l’estimation des BMFs semble donc nécessaire.

### Influence de l’intégration du régime alimentaire dans l’estimation des BMFs

L’une des contributions majeures de cette étude est la mise en évidence de l’impact de la méthodologie sur l’estimation des BMFs. En effet, la prise en compte de la composition du régime alimentaire des soles, la méthode la plus proche de la définition théorique du BMF (Gobas *et al.*, 1999), a conduit à des BMFs plus élevés pour les PFASs et HBCDDs. Ceci suggère que l’approche traditionnelle basée uniquement sur les concentrations des proies individuelles peut sous-estimer la biomagnification réelle, surtout dans le cas d’un régime alimentaire varié (Mounier *et al.*, 2020). Ce biais méthodologique pourrait expliquer certaines incohérences dans la littérature concernant les tendances de transfert trophique des PFASs (Houde *et al.*, 2006; Conder *et al.*, 2008; Loi *et al.*, 2011; Munoz *et al.*, 2017). L’influence de la composition de la nourriture a par ailleurs été montrée en expérimentation (Liu, Haffner and Drouillard, 2010). Celle-ci serait également un facteur clé en milieu naturel, en raison des différences de digestibilité et de biodisponibilité dans les proies, pouvant être notamment liées à leur teneur en lipides dans le cas des PCB, et amener à des concentrations encore plus élevées du bol alimentaire durant la digestion (Gobas *et al.*, 1999).

Ces résultats soulignent la nécessité d’une approche intégrative prenant en compte les préférences alimentaires et les proportions relatives des proies consommées, voire de leur digestibilité, pour affiner l’évaluation des risques écotoxicologiques. De plus, ils confirment que les PFASs, en particulier L-PFOS et PFNA, présentent un fort potentiel de biomagnification, ce qui doit être pris en compte dans les stratégies de surveillance environnementale et de gestion des polluants émergents.

Several reviews explore the use of monitoring-derived trophic biomagnification factors to assess the bioaccumulation potential of chemicals and have provided guidelines for improving experimental design, data treatment, and statistical analyses when conducting biomagnification and trophic magnification studies (Borgå *et al.*, 2012; Conder *et al.*, 2012; Kim *et al.*, 2016; Arnot *et al.*, 2023; Fremlin, Elliott and Gobas, 2025). Furthermore, information on prey/predator trophic interaction and exact trophic positions of the studied species have to be confirmed (e.g. by stomach content analyses or carbon and nitrogen stable isotope ratio shifts (δ13C, δ15N) ; (Borgå *et al.*, 2012)

### BMF vs TMF

Field- and laboratory-derived BAFs and bioconcentration factors are generally less accurate in predicting biomagniﬁcation (Conder *et al.*, 2012).

Although BMFs explicitly describe enrichment of chemicals between predator and prey, BMFs represent only a single trophic transfer. A BMF > 1 indicates that the substance has biomagnified in the predator, compared to the given prey (Fremlin, Elliott and Gobas, 2025). Variation in the ability of organisms to biotransform and eliminate chemicals can produce variation in BMFs among predator-prey relationships in a food web, and this can obscure the chemical’s overall food web biomagniﬁcation behaviour. TMFs provide a characterization of the average degree of biomagniﬁcation that occurs with increasing trophic position within organisms of an entire food web (Fremlin, Elliott and Gobas, 2025). Thus, it needs to be incorporating multiple food web interactions (Conder *et al.*, 2012). As in our case the number of food web interactions is the same for BMF and TMF, they are representing the same thing, and thus can be directly compared to each over. On the contrary, TMF from this article should not be compared with other TMFs.

The scientific consensus is that chemicals are considered bioaccumulative if TMF > 1. However, TMF couldn’t be estimated from our limited part of trophic web, coupled with small sample size for prey species (Borgå *et al.*, 2012). Moreover, with a small sample size as ours, a TMF below 2 or 3 is unlikely to show a statistical difference from 1, even if the true TMF is above 1 (Conder *et al.*, 2012). Indeed, we preferred considering trophic level-normalized biomagniﬁcation factors (BMFTL). BMFTL is still the second preferred metric of bioaccumulation potential recommended by (Conder *et al.*, 2012). Indeed, although BMFTL does not reflect bioaccumulation across the entire food web, the studied predator-prey relationship (i.e., soles feeding on invertebrates) remains relevant for stakeholders. It can aid in risk assessments, as sole is a commercially consumed fish (with muscle contamination posing potential human health risks), and early-life contamination may negatively impact fish stocks. Moreover, BMFTL values vary but they generally include the TMF range. This is expected since TMF can be viewed as the average BMF in the studied food web. When TMF data are unavailable, BMFTL can be a useful substitute, though it may overestimate TMFs in cases where no biomagnification occurs (Conder *et al.*, 2012).

* TMF prédit par un modèle pour les PCB - Fan Y. 2008. Development and testing of a new bioenergetic/bioaccumulation model for persistent organic pollutants in aquatic and terrestrial food webs [Master thesis]. Burnaby (BC): Department of Biological Sciences, Simon Fraser Univ.
* Supp data word (Conder *et al.*, 2012) : biblio TMF et BMFTL PFOS (Houde et al., 2006 Sarasota Bay + Tomy et al., 2004 Arctic) PCB 52(Houde et al., 2008 Wollaston, Thunder, Superior, Simcoe, Seneca, Sandybeach, Reindeer, Paguchi, Opeongo, Namur, La Ronge, Kingsmere, Grist, Eva, Cold, Champlain, Athabasca) ; PCB 153 (Hoekstra et al., 2003 Beaufort-Chukchi Sea Arctic + Fisk et al., 2001 Baffin Bay Arctic + Hop et al., 2002 Barents Sea + Houde et al., 2008 same locations as PCB 52), 209 (Mackintosh et al., 2004 Vancover, BC)

Several studies indicate that chemicals with a log KOW > 4 have the potential to biomagnify in the food web and can exhibit a TMF>1 (Conder et al., 2012)

Le calcul du TMF est basé sur l’examen de la relation statistique entre le niveau moyen de contamination des espèces et leur niveau trophique moyen le long d’une chaine trophique (Borgå *et al.*, 2012). Récemment, (Munoz *et al.*, 2017) ont listé et évalués différentes méthodes statistiques pour estimer de façon pertinente les TMF compte-tenu des différents biais statistiques potentiels (Borgå *et al.*, 2012; Mackay *et al.*, 2016). Parmi ces méthodes, le package R « LMEC » (Vaida and Liu, 2009) permet de tenir compte des données censurées à gauche (valeurs < LOD ou LOQ suivant le seuil retenu), de l’incertitude de mesures et de la variabilité intra et interspécifique via l’introduction d’un effet aléatoire « espèce » dans le modèle linéaire décrivant la relation statistique entre le log de la contamination mesurée à l’échelle individuelle et le niveau trophique. Ce dernier est estimé au moyen des concentrations en δ15N, en utilisant un ou deux organismes pour la définition de la ligne de base.

### Influence de la prise en compte des valeurs manquantes

(Schäfer *et al.*, 2015) indique que bien souvent, les résultats sont transférés depuis les analyses chimiques vers la modélisation, dans que les incertitudes associées au niveau précédent ne soient prises en compte. Cela conduit fréquemment à des précisions et fiabilités **apparentes**.

Conclusions et perspectives

Cette étude met en évidence des différences marquées dans la bioaccumulation et la biomagnification des organohalogénés chez *Solea solea*, avec des implications pour la compréhension des dynamiques trophiques des contaminants. Les résultats confirment que les PCBs, bien que persistants, ne présentent qu’une biomagnification limitée par rapport aux PFASs, et que l’HBCDD suit des schémas d’accumulation spécifiques aux isomères.

À l’avenir, des travaux complémentaires pourraient explorer :

* L’influence des facteurs physiologiques des soles sur la métabolisation des contaminants.
* L’analyse des métabolites de PFASs et PCBs pour mieux comprendre leur devenir dans l’organisme.
* L’impact de la variabilité saisonnière et spatiale sur la contamination des proies et des prédateurs.

En intégrant ces dimensions, il sera possible d’améliorer les modèles de prédiction de la bioaccumulation et de mieux évaluer les risques écotoxicologiques associés aux contaminants émergents dans les écosystèmes estuariens.

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Résultats : Trophic magnification transfers

## TMFs

Les résultats obtenus sur l’ensemble du jeu de données sont regroupés dans le Tableau 5. D’après les estimations obtenues sur l’ensemble du jeu de données (i.e. tous secteurs et périodes confondus, soles G0 à G2), l’ensemble des PFAS sélectionnés sur la base de leur fréquence de détection dans le biote apparait bioamplifiable, excepté le PFOA, tandis que parmi les composés des autres familles, seul le CB‑153 présente une valeur de TMF significativement supérieure à 1. Les valeurs obtenues ici pour les PFAS dans l’estuaire de la Seine sont globalement cohérentes avec celles obtenues pour l’estuaire de la Gironde par Munoz et al. (2017) et avec celles obtenues dans différents écosystèmes marins (Houde et al., 2004 ; Kelly et al., 2009).

En revanche, les résultats sont plus surprenants pour les PCB puisque seul le CB-153 apparait bioamplifié avec une valeur de TMF tout juste égale à 1. Les résultats sont en partie « biaisés » par le fait que la chaine trophique prise en compte est finalement assez courte : les plus grands prédateurs sont les soles G2 (dont le niveau trophique est voisin de 3) par rapport à la plupart des études de la littérature. Or, un écart de deux niveaux trophiques entre la base du réseau et les prédateurs supérieurs considérés est idéalement souhaitable pour une détermination robuste du TMF (Borgå et al., 2012). Ici cet écart est de l’ordre de 1 et l’échantillonnage couvre ainsi deux niveaux trophiques. Par ailleurs, la forte contamination de certains bivalves (situés en bas de chaine trophique) et la sous-estimation potentielle des niveaux trophiques des G2 pourraient également contribuer à la sous-estimation des TMF pour les PCB. Les valeurs de TMF obtenues pour les PCB, bien que plus faibles que celles observées dans d’autres études centrées sur le benthos et une ou plusieurs espèces de poissons (ex : Goutte et al., 2020), sont néanmoins cohérentes avec celles déterminées pour l’estuaire de la Gironde (Lauzent, 2017).

Les valeurs de TMF déterminées pour les isomères α et γ du HBCDD sont inférieures à 1, ce qui suggère l’absence de bioamplification de ces composés dans le réseau trophique de la sole dans l’estuaire de la Seine. En milieu dulçaquicole, la bioamplification est hétérogène entre sites à l’échelle régionale et ces deux isomères du HBCD ne sont pas systématiquement bioamplifiés (Lauzent, 2017). Néanmoins, les TMF déterminés ici sont largement inférieurs à ceux estimés récemment en milieu côtier pour des chaines trophiques beaucoup plus longues (Zhang et al., 2018, Ruan et al., 2018).

Tableau 7 - TMF déterminés pour les principaux composés de chaque famille (estimations et intervalle de confiance à 95%). En gras les composés pour lesquels le TMF est statistiquement supérieur à 1.

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| Famille | Composé | TMF (IC 95%) | Famille | Composé | TMF (IC 95%) |
| PFAS | PFOA | nc | **PCB** | CB52 | 0.9 (0.9-1.0) |
| **PFNA** | **1.9 (1.7-2.0)** | CB118 | 0.9 (0.9-0.9) |
| **PFDA** | **2.2 (2.0-2.4)** | CB138 | 0.8 (0.8-0.8) |
| **PFUnA** | **1.3 (1.2-1.5)** | CB149 | 0.9 (0.8-0.9) |
| **PFDoA** | **1.3 (1.3-1.4)** | **CB-153** | **1.0 (1.0-1.0)** |
| **PFTrDA** | **2.5 (2.0-3.2)** | CB180 | 0.9 (0.9-1.0) |
| **PFTeDA** | **2.1 (1.4-3.2)** |  |  |  |
| **PFOS** | **1.3 (1.3-1.4)** |  |  |
| **FOSA** | **2.4 (2.1-2.7)** |  |  |  |
| **EtFOSAA** | **3.9 (3.1-5.1)** |  |  |  |

Globalement, les valeurs de TMF ne sont donc pas tout à fait convergentes avec les diagnostics précédemment établis au regard des BMF. Quel que soit le mode de calcul du BMF, tous les congénères de PCB apparaissent bioamplifiés entre proies benthiques et soles alors que seul le CB-153 ressort si l’on considère les TMF. Les approches sont plus cohérentes entre elles pour PFAS et HBCDD. Tous les PFAS apparaissent bioamplifiés au regard des valeurs de TMF et de BMF classiques obtenues même si la conclusion est plus nuancée lorsque l’on tient compte du bol alimentaire dans le calcul du BMF.

En réalité, BMF (tels que calculés ici) et TMF reposent sur des conceptions légèrement différentes. En particulier, le calcul du TMF tient compte de l’ensemble des compartiments et des niveaux trophiques. Or, dans le cas présent, les concentrations en PCB dans les bivalves sont très fortes et leur niveau trophique faible ce qui tire la régression linéaire et influence l’estimation du TMF.

**Discussion**

Méthode de calcul des TMF : Plus récemment encore, Ballutaud et al. (2019) ont développé une méthode originale, implémentée dans un cadre bayésien, afin de tenir compte de la propagation des incertitudes dans le calcul des TMF. Ce modèle, baptisé ESCROC, repose sur le principe des modèles de mélange isotopiques classiques : il postule que la signature chimique (contaminant ou isotope) d’un prédateur est la moyenne pondérée de la signature chimique de ses proies plus un facteur d’enrichissement. Dans le cas des isotopes, ce facteur est appelé facteur d’enrichissement isotopique ; dans le cas des contaminants, il correspond au TMF. Initialement, il était prévu d’utiliser ESCROC sur les données acquises dans ce projet. Malheureusement, malgré un important effort d’échantillonnage et d’acquisition de données, les contraintes du projet n’ont pas permis de construire un jeu de données regroupant (1) suffisamment de données isotopiques acquises sur les mêmes individus que pour les données de contamination pour (2) assez d’organismes le long de la chaîne trophique (3) depuis les consommateurs primaires jusqu’à différents prédateurs. Nous n’avons donc pas pu implémenter ESCROC sur nos données. Aussi, dans le cadre de ce CHOPIN, la méthode LMEC a été privilégiée.

De plus, selon les familles, les valeurs de TMF ne sont pas tout à fait convergentes avec les diagnostics établis au regard des BMF. Ainsi, quel que soit le mode de calcul du BMF, tous les congénères de PCB apparaissent bioamplifiés entre proies benthiques et soles G0 alors que seul le CB-153 ressort si l’on considère les TMF. Différentes hypothèses ont été avancées pour expliquer les différences entre BMF et TMF (ex : forte contamination de certains bivalves situés en bas de chaine trophique). Les approches sont plus cohérentes entre elles pour le HBCDD (jamais bioamplifié ici) et les PFAS. Pour ces derniers, tous les composés fréquemment détectés dans les soles apparaissent bioamplifiés au regard des valeurs de TMF et de BMF obtenues même si la conclusion est plus nuancée lorsque l’on tient compte du bol alimentaire dans le calcul du BMF.