EStimating Contaminants tRansfers Over Complex food webs (ESCROC): An innovative Bayesian method for estimating POP's biomagnification in aquatic food webs

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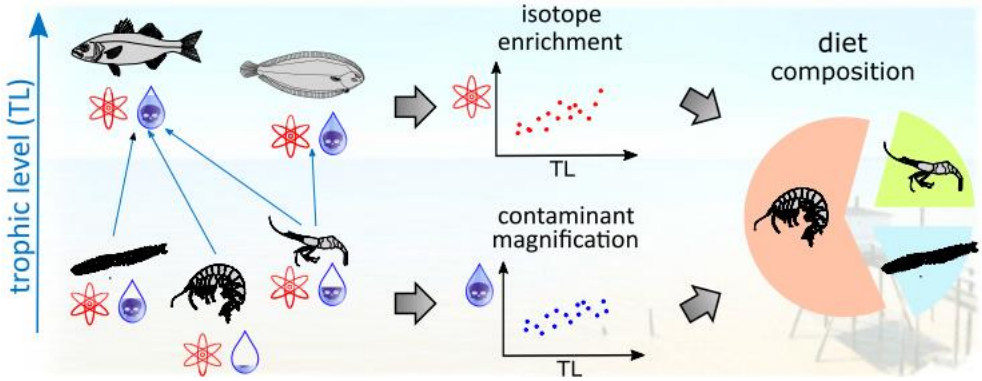
ABSTRACT

Pollution greatly impacts ecosystems health and associated ecological functions. Persistent Organic Pollutants (POPs) are amongst the most studied contaminants due to their persistence, bioaccumulation, and toxicity potential. Biomagnification is often described using the estimation of a Trophic Magnification Factor (TMF). This estimate is based on the relationship between contamination levels of the species and their trophic level. However, while the estimation can be significantly biased in relation to multiple sources of uncertainty (e.g., species physiology, measurement errors, food web complexity), usual TMF estimation methods typically do not allow accounting for these potential biases. More accurate and reliable assessment tool of TMFs and their associated uncertainty are therefore needed in order to appropriately guide chemical pollution management. The present work

proposes a relevant and innovative TMF estimation method accounting for its many variability sources. The ESCROC model (EStimating Contaminants tRansfers Over Complex food webs), which is implemented in a Bayesian framework, allows for a more reliable and rigorous assessment of contaminants trophic magnification, in addition to accurate estimations of isotopes trophic enrichment factors and their associated uncertainties in food webs. Similar to classical mixing models used in food web investigations, ECSROC computes diet composition matrices using isotopic composition data while accounting for contamination data, leading to more robust food web descriptions.

As a demonstration of the practical application of the model, ESCROC was implemented to revisit the trophic biomagnification of 5 polyfluoroalkyl substances (PFAS) in a complex estuarine food web (the Gironde, SW France). In addition to the TMF estimate and 95% confidence intervals, the model provided biomagnification probabilities associated to the investigated contaminants —for instance, 92% in the case of perfluorooctane sulfonate (PFOS) — that can be interpreted in terms of risk assessment in a precautionary approach, which should prove useful to environmental managers.

Graphical abstract



Highlights

- Estimating the trophic magnification potential of chemicals is a key issue for management
- This biomagnification potential is usually estimated through a Trophic Magnification Factor (TMF)
- ESCROC is an innovative Bayesian mixing model for estimating TMFs
- It provides rigorous diagnoses on contamination and associated uncertainty
- The example of PFASs in the Gironde estuarine food web was used as a case study

Keywords

Trophic magnification – food webs – Bayesian mixing model – organic micropollutants – stable isotopes - Gironde estuary

1 Introduction

Increased nutrients, pollutants, and agrochemicals due to industries, urbanization and agriculture exert dramatic impacts on ecosystems (Köhler and Triebskorn, 2013; Verhoeven et al., 2006). Aquatic ecosystems and, among them, coastal and estuarine ecosystems are particularly vulnerable to these changes: they are increasingly exploited and polluted, and their biodiversity is decreasing (Budzinski et al., 1997; Matthiessen and Law, 2002). At the individual and population scales, some pollutants can lead to deleterious effects, such as altered metabolism, immunotoxicity, endocrine disruption or neurotoxicity (Köhler and Triebskorn, 2013). This issue is exacerbated by the fact that some pollutants tend to be accumulated by organisms, a process known as bioaccumulation. Bioaccumulation is a fundamental process in environmental toxicology and risk assessment because it controls the internal dose of potential toxicants in individual organisms (Arnot and Gobas, 2004). Additionally, some contaminants also become ecologically harmful because they accumulate through food webs, a process known as biomagnification. In these instances, pollutants found at low concentrations in natural environments can achieve harmful concentration for high-order organisms including human beings (Kelly et al., 2007; Köhler and Triebskorn, 2013; Van Oostdam et al., 2005). Studies aquatic ecosystems –marine systems (e.g. Romero-Romero et al., 2017; Sun et al., 2017), lakes (e.g. Liu et al., 2018; Mazzoni et al., 2018), coastal environments (Bodin et al., 2007; Loizeau et al., 2001a; Loizeau et al., 2001b; Munschy et al., 2011) and rivers (Lopes et al., 2011)- but also in terrestrial environments (e.g. Daley et al., 2011; Wang and Gao, 2016) demonstrated such biomagnification process occurs for hydrophobic organohalogenated contaminants. Being potentially persistent, bioaccumulative and toxic (due for instance to eurotoxic properties and/or endocrine disruption), Persistent Organic Pollutants (POPs) are of particular concern (see the Stockholm Convention on POPs as amended in 2009 - UNEP, 2009) for human health (Belpaire et al., 2016; Berger et al., 2009) as well as for animal populations' viability (Gilliers et al., 2006a; Gilliers et al., 2006b; Rochette et al., 2010). In this context, a comprehensive understanding of the ecodynamics of human-induced chemicals in coastal and

84 estuarine ecosystems is needed to better manage the ecological functions associated with these
85 areas.

86 Potential for bioaccumulation in organisms and biomagnification in food webs differ depending
87 on the investigated contaminants, environmental contexts, and physiological characteristics of species
88 (Bodiguel et al., 2009; Connolly and Glaser, 2002; Gobas, 1993). Therefore, accurate *in situ* assessment
89 of bioaccumulation and biomagnification potential of pollutants in aquatic food webs is required, in
90 order to inform management actions.

91 Most empirical approaches used to understand trophic transfers of pollutants rely on the
92 estimation of a Trophic Magnification Factor (TMF) from field data (Borgå et al., 2012). The TMF is
93 used to assess the biomagnification of a given pollutant in a food web and to define environmental
94 quality norms in some instances (see french envrionmental quality norms (NQE) - Migne-Fouillen et
95 al., 2010). It is usually corresponds to the slope of the statistical regression between the chemical
96 concentration and the trophic level of organisms within a food web. Although TMF is increasingly used
97 to describe trophic dynamics of xenobiotics, its estimates present many uncertainties, reviewed by
98 Borga et al. (2012) and Mackay et al. (2016). Among the manyassumptions, potential bias, pitfalls, and
99 vigilance points reported in these reviews, uncertainty in measurements of contaminant
100 concentrations, temporal or spatial variability of these concentrations, , inter and intraspecific
101 variability in the bioaccumulation process, and uncertainty about the food web structure and trophic
102 levels of individuals were emphasized. Recently, Munoz et al. (2017) evaluated different statistical
103 methods to address these above-mentioned sources of uncertainty and bias, based on data on 19
104 polyfluoroalkyl substances (PFASs) in the Gironde estuarine food web. The statistical approaches
105 compared included linear mixed models from the 'NADA' (Lee, 2017) and 'LMEC' (Vaida and Liu, 2012)
106 R-packages, accounting for censored responses and a random effect 'species', respectively. Both
107 methods however assumed that the trophic level of each individual —and consequently, the structure
108 of the trophic network— are perfectly known, an assumption that rarely verifies in the real world, and
109 particularly not in the Gironde estuary.

Theoretically, estimating the trophic level of a consumer species (or an individual) requires that its diet be estimated, i.e., the proportion, in biomass, of each prey and its respective trophic level, in the overall consumer's diet. This is usually done through stomach content analyses, which reflect the quantitative and qualitative ingestion of species at a specific time, but sometimes raise problems of prey identification, suffers from some biases such as differential digestibility, and requires many samples to be analyzed. Stable isotope analyses represent more integrative records of food intake over longer time scale (Post, 2002) and are now widely used to explore food web structure (Boecklen et al., 2011; Layman et al., 2012). Stable isotopes of nitrogen are discriminated during digestion and assimilation, leading to an enrichment of the $^{15}N/^{14}N$ ratio ($\delta^{15}N$) of the consumer with respect to its prey. Since the enrichment factor is generally in the range 3 –4 ‰ (DeNiro and Epstein, 1981; Minagawa and Wada, 1984; Peterson and Fry, 1987), linear regressions can be used to convert stable isotope composition into trophic level (Post, 2002). This is the usual method chosen to estimate trophic level when investigating contaminant biomagnification (Borgå et al., 2012). Combined with the consideration of the isotopic ratio of other elements (e.g. $^{13}C/^{12}C$ or $^{34}S/^{33}S$), it can be used to estimate the diet of species among a set of potential prey. This is the approach used by MixSIR, SIAR and MixSIAR, the mixing models classically used to estimate a species diet based on its isotopic composition, which is assumed to be a mixture of the isotopic compositions of the different prey (Moore and Semmens, 2008; Parnell et al., 2008; Parnell et al., 2010; Parnell et al., 2013; Stock et al., 2016). This approach allows accounting for the uncertainty of stable isotope enrichment factors, and for intra- and interspecific variability. However, since the number of isotopic tracers is small (i.e., usually two) compared to the number of potential prey, isotope mixing models are generally underdetermined (Fry, 2013; Phillips and Gregg, 2003; Phillips et al., 2014). Consequently, precise estimates of enrichment factors must be provided for the mixing model to work, although these estimates are very sensitive parameters (Bond and Diamond, 2011). Furthermore, it is not possible to carry out the analysis using those mixing models over a full trophic network but only one predator after another. Finally, since biomagnification and food web analyses are generally carried out into two

136 independent steps, it is usually impossible (or very difficult) to propagate the uncertainty on estimated
137 trophic levels to the estimation of biomagnification factors. For both scientific and management
138 reasons, it is therefore necessary to provide an alternative rigorous method based on sound statistics
139 to evaluate biomagnification factors (TMF) and associated uncertainty.
140 The present work aimed at presenting such alternative method combining both biomagnification and
141 food web analyses into a single model. We assume that using contaminants as additional diet tracers
142 within trophic networks could mitigate the issue of underdetermination of mixing models. Conversely,
143 incorporating the inference of trophic levels within biomagnification analysis allows propagating the
144 uncertainty over trophic levels when estimating biomagnification factors such as TMF. Diet tracers
145 such as isotopes can then be used to estimate contaminant transfers. Our model aims at (1)
146 accounting for most of the sources of variability listed above on both biomagnification and isotopic
147 fractionation, (2) estimating diets and related uncertainty for all predators of the food web at once, (3)
148 estimating the biomagnification of contaminants and related uncertainty. Our model, named ESCROC
149 (EStimating Contaminants tRansfers Over Complex food webs), is based on a generic mixing model,
150 similar to those used for deriving diet composition from isotopic data, but allows incorporating
151 contamination measures.
152 To illustrate the relevance of this model, a large dataset describing the occurrence of a family
153 of Persistent Organic Pollutants (POPs) in the Gironde estuary was used. Located on the French
154 Atlantic coast, in SW France and largest estuary in Western Europe (Lobry et al., 2003), the Gironde
155 estuary case study is especially relevant since POPs are now an increasing issue in this area (Munoz et
156 al., 2017; Tapie et al., 2011). Among those substances, the target selected compounds were
157 polyfluoroalkyl substances (PFASs). Few studies have addressed the contamination of estuarine food
158 webs by these emerging contaminants (de Vos et al., 2008; Naile et al., 2012; Munoz et al. 2017). As
159 the dataset was previously described and analyzed by Munoz et al. (2017) with a set of various
160 modelling approaches, this would also allow a comparison of the outcomes provided by the ESCROC
161 model with those obtained with more traditional methods.

2 Material and methods / description of the model

2.1 Context on TMF estimation

2.1.1 Basis of TMF estimation

Basically, TMF estimation consists in assessing the average factor of change in contaminant concentration per Trophic Level (TL). In that sense, it is similar to the enrichment factor in isotopic analysis. Considering biomass distribution in aquatic food webs as well as contaminants transfer efficiencies, the relationship between contaminant concentrations [C] and TL has an exponential form (Borgå et al., 2012). Therefore, TMF estimations are based on the linear regression between Log-transformed contaminant concentrations $Log_{10}[C]$ and TL. The TMF is subsequently obtained as 10^b, with b being the slope of the linear regression (Eq. 1, Figure 1) usually estimated from simple regression models.

$$\mathrm{Log}_{10}[\mathrm{C}] = \mathrm{a} + \mathrm{b} \bullet \mathrm{TL} \qquad \text{Eq. 1}$$

Modifications to improve TMF estimation calculation were however recently proposed to better account for different sources of uncertainty and variability. For instance, Munoz et al. (2017) suggested to use linear mixed models with censored data (to take into account non-detected data), with random effects to integrate both inter-specific variabilities in physiological properties, errors in chemical concentration measurements and potentially low sampling effort.. These methods however still assumed that trophic levels were perfectly known.

If initially, the trophic positions of organisms in the food webs were directly assessed using stable N isotope ratios ($\delta^{15}N$), most recent studies (see Borga et al. 2012 or for instance Munoz et al. 2017) refined the estimation of trophic position by using integer-based TL. The TL of a primary producer being fixed to 1 by convention, a primary consumer has a TL of 2. Thus, for secondary consumers, the trophic level of a particular individual (TL_i see Eq. 2) is estimated using the difference between its own stable N isotope ratio ($\delta^{15}N_i$, obtained using tissue measures of $^{15}N/^{14}N$) and a source isotope ratio $\delta^{15}N_{base}$ at the base of the food web weighted by the trophic enrichment between TLs ($\Delta^{15}N$). Different sources for benthic and pelagic food chains ($\delta^{15}N_{base1}$ and $\delta^{15}N_{base2}$) are often considered in marine coastal and/or estuarine environments, to reflect the complexity of trophic food webs in those systems. Each individual has to be allocated to one or the other of these food chains through an α coefficient (from totally benthic: α=1 to totally pelagic α=0), which has to be fixed *a priori* using expert knowledge on species' feeding ecology and food web structure.

$$TL_i = 2 + \frac{[\delta^{15}N_i - \{\alpha \bullet \delta^{15}N_{base1} + (1-\alpha) \bullet \delta^{15}N_{base2}\}]}{\Delta^{15}N} \quad \text{Eq. 2}$$

Then, a linear mixed model has to be fitted for each food web (benthic and pelagic) independently, by selecting species based on α values For species feeding on both pelagic and benthic preys and sources, α values can be comprised between 0 and 1. When α>0.5 species are allocated to benthic food chain and to the pelagic one for α<0.5 for pelagic.

Estimating trophic levels is therefore not straightforward, and subjected to multiple sources of uncertainty, including measurements of $\delta^{15}N$ of individuals and sources at the basis of the food web, and estimations of α values associated to each species considered in the food web. Moreover, while a linear increase in $\delta^{15}N$ with TL probably oversimplified the mechanism of isotope discrimination (Hussey et al., 2014) and equation 2 is not necessarily relevant in a situation where trophic chains are intertwined in complex interaction trophic networks.

2.2 ESCROC modeling framework

The proposed model (ESCROC) was developed in a Bayesian framework. The Bayesian theorem allows (1) combining objectively different core metrics accounting for their sensitivity and variability and (2) providing rigorous uncertainty quantification.

The approach in ESCROC was based on the same conceptual framework as stable isotope mixing models such as MixSIAR (Parnell et al., 2013; Stock et al., 2016). In such models, consumer species are assumed to feed on a combination of prey items (or sources) that are all known, and that the isotopic composition $y_{i,e,t}$ of an individual *i* of species *e* for tracer *t* results from the combined isotopic composition of the assimilated prey items.

2.2.1 Model formulation

In ESCROC, we combine isotope values and contaminant concentrations (in log scale) as chemical tracers of food web structure.

We can therefore describe the mean concentration *Y* of a tracer *t* for a species *e* as a combination of its consumed prey (*p*) concentrations. In lieu of raw tracer concentrations, scaled values were used by subtracting the average value and dividing by the standard deviation: $y'_{i,e,t} = (y_{i,e,t} - \bar{y_t})/\sigma_t)$ so that all tracers share a common scale. This facilitates the integration of prioris and statistical inference of the model (Bolker et al., 2013).

Based on stable isotope mixing model assumptions, the tracer composition of a predator was calculated using the tracer compositions of its prey items:

$$Y_{e,t}=\frac{\sum_{p\in prey(e)}\left(\rho_{e,p}\cdot q_{p,t}\cdot\left(Y_{p,t}+\Delta_t^{'}\right)\right)}{\sum_{p\in prey(e)}\left(\rho_{e,p}\cdot q_{p,t}\cdot\right)}+E_{e,t}\ \text{with}\ E_{e,t}\sim N(0,s_t^2) \qquad \text{Eq. 3}$$

where

$Y_{e,t}$ is the average value of tracer (either contaminant or isotope) *t* value for species *e*

$\rho_{e,p}$ is the dietary contribution of prey *p* for consumer *e*

$q_{p,t}$ is the concentration of *t* in prey *p*

$Y_{p,t}$ is the measured mean tracer *t* value for prey *p*

Δ_t' is the enrichment factor for tracer *t.* Note that this corresponds to the enrichment for the scaled values, which can be converted to the enrichment in the original scale: $\Delta_t = \sigma_t \cdot \Delta_t'$

$E_{e,t}$ is the species random effect for species *e* and marker *t* that accounts for inter-specific physiological variability

As ESCROC is implemented in a Bayesian framework, priors can be defined for unknown parameters. Priors, corresponding to possible *a priori* distributions of the parameters, can be constructed using knowledge from various sources (expert knowledge, meta-analyses, other field data…). In the absence of external knowledge, uninformative or weakly informative priors can be built.

$Y_{e,t}$ as described in Eq3 therefore corresponds to a weighted average of the prey tracer compositions (with weights corresponding to the importance of the prey in the predator's diet and to the concentration of the tracer in the prey), to which we added an enrichment and a species effect. If $q_{p,t}$ are equal among prey items, equation 3 simplifies to:

$$Y_{e,t}=\sum_{p\in prey(e)}\left(\rho_{e,p}\cdot Y_{p,t}\right)+\Delta_t^{'}+E_{e,t}\ \text{with}\ E_{e,t}\sim N(0,s_t^2) \qquad \text{Eq. 4}$$

Note that the model does not work if two species are both prey and predator of each other because equations 3 or 4 become circular.

Similarly, the variance of the values of the tracer *t* for a species *e*, denoted $V_{e,t}$, can be calculated from the variances of the different prey:

$$V_{e,t}=\frac{\sum_{p\in prey(e)}\left(\rho_{e,p}^2\cdot q_{p,t}^2\cdot V_{p,t}\right)}{\left[\sum_{p\in prey(e)}\left(\rho_{e,p}\cdot q_{p,t}\right)\right]^2}+B_t \quad \text{Eq. 5}$$

With B_t a variable to add potential noise at each trophic level.

If the concentration parameters $q_{p,t}$ are all equals, then the equation simplifies to:

$$V_{e,t}=\sum_{p\in prey(e)}\left(\rho_{e,p}^2\cdot V_{p,t}\right)+B_t$$

In equation 3, if $Y_{e,t}$ is the concentration of a contaminant (in log10 scale) and assuming that all concentration parameters are equal (as in equation 4), then Δ_t corresponds to the enrichment in contaminant between a prey and its predator, i.e., the enrichment along the food web when trophic level increases by 1. Consequently, it corresponds to the slope of the line in Figure 1 and we obtain $\Delta_t=\log_{10}(TMF)$.

The measured values of a given tracer on a given dataset are supposed to follow a Normal law whose parameters are calculated from previous equations. We thus assume that the distribution of the tracer *t* value, for an individual *i* of the species *e* can be written as:

$$y_{i,e,t}\sim Normal(Y_{e,t},V_{e,t}) \quad \text{Eq. 6}$$

In case of the concentration measurement of a given tracer is null or not reliable for analytical reasons (e.g. below the detection threshold of a given measurement method), it can be considered as a left-

censored data. The cumulative distribution of the normal distribution is thus used instead of the density of probability.

2.2.2 Model calibration and priors construction

We propose here a selection of possible priors that can be implemented for most of the model applications, but adaptation can be made in relation with available data or expert knowledge.

Selection of priors for main parameters

Priors can be supplied for the enrichment factors either on the transformed scale or on the original scale. Informative priors for Nitrogen and Carbon TEF (Δ_N and Δ_C see Eq. 3) can be inferred from the literature (e.g. Post 2002). For instance, these priors can be implemented as follows:

$$\Delta_N \sim \text{Normal}(3,1) \quad \text{Eq. 7}$$

$$\Delta_C \sim \text{Normal}(0,1) \quad \text{Eq. 8}$$

corresponding to a TEF value for N around 3 and around 0 for C.

A non-informative prior can be used for the enrichment factors of all tracers corresponding to PFAS concentrations:

$$\Delta'_t \sim \text{Normal}(0,10) \quad \text{Eq. 9}$$

Finally, non-informative priors can be used for other parameters (residual variation and random effect) of model formulation (Eq. 3 and Eq.):

$$B_t \sim \text{Inverse Gamma}(0.01, 0.01) \quad \text{Eq. 10}$$

$$\sigma_t \sim \text{Uniform}(0.01, 10) \quad \text{Eq. 11}$$

Diet matrix

Information on trophic interactions should be implemented in the model. At first, we can only specify if a prey *p* can be predated by a consumer *e* based on the evidence of predator-prey relationships using field data.

An uninformative prior for the dietary contribution of prey *p* for consumer *e* ($\rho_{e,p}$ see Eq. 3) can then be constructed by assuming that it follows a Dirichlet distribution:

$$\{\rho_{e,1}, \cdots, \rho_{e,n}\} \sim \text{Dirichlet}(\{\lambda_{e,1}, \cdots, \lambda_{e,n}\}) \quad \text{Eq. 12}$$

where $\lambda_{e,p} = 1$ if *e* feeds on *p* and $\lambda_{e,p} = 0$ otherwise; n being the number of species in the considered food web.

Informative priors can be implemented if external data, such as stomach contents, are available. However, considering the implementation framework, trophic loops cannot be included in the food web description.

Constructing priors for tracer values using data

Average tracer compositions of predators are calculated from prey tracer compositions. However, if a species does not have prey (e.g. primary producers), or if some prey of a species are not present in the dataset (i.e., no contamination data or isotopic measurements), it is necessary to provide priors for

this species. We will see in the case study how an informative prior can be constructed. However, in the absence of external data, a weakly informative prior can be implemented as follows:

$$Y_{e,t} \sim \text{Normal}\ (0,10) \quad \text{Eq. 13}$$

$$V_{e,t} \sim \text{Gamma}\ (0.01,0.0) \quad \text{Eq. 14}$$

2.2.3 Outputs and Implementation

Outputs

Main outputs of ESCROC consist in posterior distributions of the estimated parameters. Three main types of outputs can be obtained from ESCROC: diet compositions of each consumer of the investigated food web, enrichment factors for each tracer, and TMF estimates (with associated credibility interval).

As a mixing model, ESCROC allows estimating a distribution of the proportion of each prey in the diet of the predators in the investigated food web.

Furthermore, by considering posterior distribution of enrichment factors (see Eq. 3, Figure 4) for N and C isotopic ratios, ESCROC allowed re-estimating TEF values, which are usually empirically fixed in the literature on isotope-based trophic studies, with aforementioned uncertainty. TEF can indeed be estimated using the median of the distribution and 2.5% and 97.5% quantiles, providing associated 95%-credibility intervals. In the same way, estimated posterior distributions of enrichment factors for contaminants can be used to estimate TMFs and associated credibility intervals. In this case, TMFs corresponds to $10^{\Delta t}$. Similarly, an estimation of TMFs can be provided using the median of the distribution and bounds of the 95%-credibility interval, computed from 2.5% and 97.5%-quartiles. Associated with TMF estimates, the probability of a contaminant to be biomagnified in the

investigated food web can then be computed by estimating the probability of Δ_t to be positive (*i.e.* corresponding to a TMF value greater than 1).

Implementation

ESCROC was implemented using the R software (R Development Core Team, 2006) and the integrated development environment (IDE) R-studio (the model being run using *coda* and *runjags* packages). For the Bayesian part of the model, the JAGS language was used (Plummer et al., 2016). The model convergence can be checked using Gelman and Rubin tests (Brooks and Gelman, 1997; Gelman and Rubin, 1992). A first beta version of the R-package (escrocR) implementing the method is available on GitHub (https://github.com/Irstea/escroc). It can be cited as follows:

Hilaire Drouineau, Marine Ballutaud and Jeremy Lobry (2018). EscrocR: a R package implementing the model ESCROC. R package version 0.0.0.9000.

2.3 Illustrative example: PFAS in the Gironde estuarine food web

In this illustrative example, the method was applied to a dataset on PFAS contamination in the Gironde estuarine food web. The main aims were to estimate (1) TMF values for a set of PFAS previously described and analyzed (Munoz et al. 2017), (2) enrichment factors of two isotopes of nitrogen and carbon and (3) the diets of all species within the trophic network.

2.3.1 Dataset

Data used in this study are taken from Munoz (2015) and Munoz et al (2017). Samples were collected between May and November 2012 in the mesohaline zone of the Gironde estuary. Amongst the initial dataset of 147 biological samples from 18 species or group of species, a subset of data was used

362 comprising 138 samples from 16 species, for which both isotopic data ($\delta^{15}N$ and $\delta^{13}C$) and PFAS
363 concentrations were available. We selected 5 PFAS (L-PFOS, FOSA, PFOA, PFNA and PFUnDA, see Table
364 S1) based on their occurrence in biota samples, the existence of censored data (considering PFAS with
365 different proportions of censored data), the TMF values previously estimated, and the variety of
366 chemical structures (ie., fluoroalkyl chain length or functional group - Table 1).
367

368 *2.3.2 Specific priors*

369 The priors for diet compositions were directly built from the trophic network illustrated in Figure 2.
370

371 For two groups (gammarids and copepods), instead of using uninformative prior as proposed in
372 equations 13 and 14, we built an informative prior to take advantage of pre-existing data on nitrogen
373 and carbon isotopic ratios (David, unpublished data). We specified a normal prior parameterized with
374 the mean and the standard errors in pre-existing available tracer composition data:
375

$$\frac{Y_{e,t-\bar{y}_t}}{\sigma_t} \sim \text{Normal}\left(\mu_{e,t}, se_t^2\right) \quad \text{Eq. 15}$$

376

377 with

378 e species or group of organism (gammarid or copepods)

379 t type of chemical tracer (carbon or nitrogen)

380 μ_{et} arithmetic mean of tracer values in an independent dataset

381 se_{et} associated standard error

382

383 Regarding the variances, the estimator of the variances in the samples follows the distribution:

384

$$\upsilon_{e,t} \sim \frac{V_{e,t}}{\sigma_t^2 \cdot n_{e,t}\text{-}1} \chi^2_{(n_{e,t}\text{-}1)} \quad \text{Eq. 16}$$

with

$v_{e,t}$ the estimator of the variance in the samples

$n_{e,t}$ number of samples

e species or group of organism (gammarid or copepods)

t type of chemical tracer (carbon or nitrogen)

Specific gammarid and copepod priors were thus built as follows:

$$\frac{1}{V_{e,t}} \sim \frac{\upsilon_{e,t}}{\sigma_t^2 \cdot n_{e,t}\text{-}1} \chi^2_{(n_{e,t}\text{-}1)} \quad \text{Eq. 17}$$

2.3.3 Outputs and model implementation

We set $q_{p,t}$=1 for all species in the model. We also computed an index α_e for each predator to compare to the α values (see section 2.1.1) arbitrarily assigned in the Munoz et al. dataset (2017).

$$\alpha_e = \sum_{p \in prey(e)} (\rho_{e,p} \cdot \alpha_e)$$

For gammarids and copepods, we set α_e=0 (pelagic), and we set α_e=0 (benthic) for nereids and crabs.

The model was fitted using a MCMC (Monte Carlo Markov Chain) method. Three chains were used in parallel with 1 million preliminary iterations – burnin – followed by 150,000 iterations to assess posterior distributions. We run the model with JAGS software version 4.1.0 and check convergence using the Gelman-Rubin diagnostic (gelman.test< 1.05).

3 Results

3.1 Model calibration and convergence

Values of Gelman indices confirmed the model convergence for 68 parameters out of 70. The only two parameters for which the model did not converge corresponded to tracer composition of prey items with 100% of censored data.

The model fitted observations as suggested by the plot of predicted *posterior* distributions of mean species tracer composition against observed values (Figure 3).

3.2 Outputs

3.2.1 Diet compositions

Diet compositions were estimated for each predator of the Gironde estuarine food web for which both isotope and PFAS data were available (see Figure 4 as an example). A global diet matrix can then be obtained by compiling all the diet values estimated by ESCROC using both N and C isotopic ratios and the 5 PFAS concentrations (Table 2).

For most species, the coefficients relating the species to the pelagic and benthic food webs were rather consistent with the expert knowledge used in Munoz et al. (2017) though the model tends to consider less species as benthic (Table 3). Those results confirm that it is very difficult to separate species into a set of two independent food-webs, as required by usual TMF estimation methods.

3.2.2 Enrichments and TMF

TEF values and the associated uncertainty were estimated, as well as TMFs estimates and biomagnification probabilities (Figure 4 and Table 4).

Non-informative priors were used for the PFAs enrichment factors, as highlighted by the flat curve lines in Figure 4. In these cases, ESCROC provided informative posterior estimates for contaminants TMFs. Conversely, informative priors based on literature were used for isotopes TEFs. Although the estimated posterior distribution appeared consistent for $\delta^{15}N$, it was significantly different for $\delta^{13}C$ even if the classical value used in the literature and in our prior definition (TEF = 0) is comprise in the posterior credibility interval.

4 Discussion

4.1 Limits and benefits of the ESCROC model formulation

4.1.1 Bayesian framework, a priori information and uncertainty propagation

ESCROC was implemented in an innovative and flexible Bayesian framework to estimate TMFs and associated uncertainty. This modeling approach presents several advantages.

Unlike frequentist methods previously mentioned (e.g. linear models and mixed models), the Bayesian framework allows to account rigorously for uncertainty propagation in measurements (e.g., TL estimates) to TMF estimates. Starrfelt et al. (2013), for instance, recommended the use of Bayesian inference to account for measure uncertainty in contaminants concentrations and isotope ratios ($\delta^{15}N$), as well as for variability and uncertainty related to TL estimates, in order to improve the precision of TMF estimations. ESCROC goes even further, by providing credibility intervals for all parameters, in particular for contaminants TMFs and isotopes TEFs. By doing so, ESCROC represents a noteworthy methodological advance as compared to traditional methods for TMF estimation.

Based on both a contaminant propagation model and an isotope mixing model, ESCROC combines and enriches both modeling approaches. The contaminant propagation model therefore benefits from integrated diet estimations. As such, TMF estimates are of more generic nature, as they no longer depend on a pre-specified trophic chain structure that the model user provides (and usually simplify). This model rather accounts for the whole food web complexity at once, and provides generic TEF estimates. Similar to widely-used mixing models (Parnell et al., 2008; Parnell et al., 2010; Parnell et al., 2013; Stock et al., 2016), ESCROC allows estimating diet compositions in investigated food webs (See Supplementary Materials S2 for a preliminary comparison of both approaches). It allows going even further than classical mixing models. Indeed, using contaminants in addition to isotopes data increases the number of chemical tracers and allow the estimation of isotope enrichment factors, a significant improvement as compared to the use of fixed *a priori* -values from the literature (usually from Post (2002)), values which are known to not perform as well in various environmental contexts or for contrasted food webs). Furthermore, our modelling approach partly addresses some of the recommendations listed by Hussey et al. (2014) for estimating isotope discrimination. The full Bayesian estimation framework indeed provides a pragmatic and very flexible estimation of enrichment factors. The framework is thus generic and can be applied in all ecosystem contexts. Furthermore, a random effect 'Species' is added in the model. Thus, although the TEF and TMF estimation are still considered globally constant through the food web, they are modulated species by species. By doing this, ESCROC did not specifically consider that isotope discrimination varies with trophic position but it allowed the estimations to vary for every species.

In the Gironde estuarine case study, the TEF for $\delta^{13}C$ estimated from ESCROC considerably differs from the one in literature. This is mostly explained by the fact we used data from studies on marine environments to compute the prior distribution while estuarine ecosystems are usually enriched on continental organic carbon which signature is different. However the model allowed not only estimating more accurately the TEF distribution for $\delta^{13}C$ in the estuarine context but the estimates remains consistent with the reference literature (e.g. Post, 2002). This result further highlights the

need for accurate TEF estimates based on the best available knowledge, as already advocated in multiple reviews of isotope-based ecological studies (Martinez del Rio et al., 2009; Layman et al., 2012; Bastos et al., 2017).

Additionally, prior knowledge incorporation is a really significant advantage of both ESCROC and MixSIAR approaches (Parnell et al., 2010; Parnell et al., 2013). Indeed, the Bayesian framework is especially well-adapted to integrate *a priori* information such as expert knowledge or external datasets. For instance, in the present case study, external information about copepods isotopic composition were combined in an informative prior as available data were uncertain. Similarly, expert knowledge and food web data (such as stomach contents) could have been used to compute more informative prior on trophic interactions, instead of using an uninformative prior for the contribution of each prey in each consumer diet.

4.1.2 Computing TMF and associated bioaccumulation probabilities

Although the initial purpose of the model was to compute TMF estimates and associated uncertainty, the ECROC modeling approach provides a comprehensive framework for the understanding of contaminants transfers in a complex food web. In relation with the Bayesian framework used, ECSROC indeed provides a biomagnification probability, which corresponds to the probability of a particular contaminant to be biomagnified in the investigated food web. This innovative feature is especially relevant for risk assessment. In fact, this probability directly expresses the risk a manager would take by classifying a given contaminant as “biomagnifiable” in a given food web. As such, ESCROC model represents an important tool to support decision making. For instance, estimated thresholds of risks could be used to define contamination levels for which additional monitoring is required, as well as levels for which specific management measures appear mandatory.

4.1.3 Limitations

Despite the aforementioned advances and advantages, ESCROC modeling approach also presents two major limitations.

First, considering the implementation framework, trophic loops cannot be considered in the food web description, although such phenomena may exist in nature. We can cite cannibalism as an illustrative example, a process relatively common in aquatic food webs, in which adult consumers sometimes prey upon juveniles from the same species (Livingston, 2002). In our case example, we assumed such flows to be be negligible or, at least much weaker than direct prey-predator trophic flows. This assumption seems reasonable with regards to available knowledge on food webs in our study system (see for instance Lobry et al., 2008; Selleslagh et al., 2012; Tecchio et al., 2015 for French estuaries).

Another limitation of the ESCROC modeling approachlies in technical aspects. First, computing time can reveal quite long depending on food web complexity and computer devices used. Second, data about multiple tracers need to be included to avoid any underdetermination issues, as TMFs and TEFs are estimated together and at the whole trophic network scale, SSince, the number of tracers (either isotopes or contaminants) should be large enough to avoid any underdetermination issue. This implies considerable efforts in sample collection, preparation, and chemical analysis, an even higher than for a classical mixing model such as MixSIAR.

4.2 Diagnosis about PFAS in the Gironde estuary

The computed TMF values of the five investigated PFAS are not significantly greater than 1 with a α-risk at 95%. This implies that, considering the results of the present study, none of the five investigated contaminants can be considered as 'biomagnifiable' in the Gironde estuarine food web. However, when considering the biomagnification probabilities associated with TMF estimates, results are more contrasted. The diagnosis actually depends on the risk-level a manager is ready to accept. For instance, if a risk-level was fixed to 30% in a precautionary approach, PFUnDA, FOSA, and more

528 particularly L-PFOS would be considered to be biomagnified in the food web with 72%, 86% and 92%
529 of certainty, respectively (Table 4). These considerations give sense to the computation of
530 biomagnification probabilities within the ESCROC modeling tool.
531
532 Our ESCROC-based diagnosis about PFAS contamination in the Gironde estuarine food web are slightly
533 different from previous assessment from Munoz et al. (2017). In the latter study, the 5 investigated
534 contaminants were considered to have been magnified at both the benthic chain and the whole food
535 web levels, whereas only one TMF estimates (FOSA) was greater than one when considering the
536 pelagic chain. Differences between Munoz et al. (2017) and our interpretation probably arises from
537 differences in the methodological approaches. First, pelagic and benthic data were combined in
538 ESCROC's estimates as well as in the Munoz's pooled estimates (Table S17 in Munoz et al., 2017). As
539 we saw in section 3.2.1, such combination is probably more realistic than separating benthic and
540 pelagic food chains, and this difference can lead to significant contrasts in TMF estimates. Second,
541 the Bayesian model formulation provides a better integration of uncertainty propagation than
542 traditional linear models. This leads to greater associated uncertainties and larger credibility intervals
543 than the confidence intervals estimated with the LMEC method used in Munoz et al. (2017) for pooled
544 TMFs. Third, Munoz et al. (2017) study was based on TMF estimates only whereas ESCROC provides
545 both TMFs estimates and biomagnification probabilities. Considering only TMFs estimates however,
546 both studies also led to contradictory results. When considering biomagnification probabilities as well,
547 our and Munoz et al. (20174)'s diagnoses remain different, but partly converge for PFUnDA, FOSA and
548 L-PFOS (see above). Finally, previous diagnoses from Munoz et al. (2017) based on classical TMF
549 estimations through linear regression appear questionable, with regards to aforementioned statistical
550 consideration, although such a method can still be seen as a simple and useful approach to perform
551 comparative studies for comparing biomagnification of selected chemicals in a given ecosystem.
552 Nevertheless, the results obtained in the present study also plead for a precautionary approach when

553 interpreting contaminant transfer data, and for using biomagnification probabilities rather than TMF
554 values alone.
555

556 ### 4.3 Perspectives for the ESCROC modeling approach

557 More than an innovative estimation framework for TMFs in complex food webs, ESCROC can also be
558 viewed as an improved mixing model for food web analyses. Considering more chemical tracers than
559 the classical N and C isotopes indeed clearlyimproves diet matrix estimations. Results obtained for the
560 Gironde estuarine food web are in line with those previously obtained by Pasquaud et al. (2008) and
561 Pasquaud et al. (2010) using both stomach contents and isotope data, and by Lobry et al. (2008) using
562 literature compilation and mass-balance modeling. Moreover, in the ESCROC modeling approach
563 presented herein, tracers are contaminants, but other kind of isotopes (e.g. $\delta^{34}S$) can be used as well.
564 As highlighted by Mackay et al. (2016), several processes related for instance to hydrophobicity or
565 rates of biotransformation and growth can influence contaminant biomagnification. However, as far as
566 they biomagnify, any type of tracer can be used in the ESCROC modeling framework. In their *Best*
567 *practice in Ecopath with Ecosim food-web models for ecosystem-based management*, Heymans et al.
568 (2016) underlined that: "Diet estimates for functional groups can also be obtained from stable isotopic
569 analyses using Bayesian isotopic mixing models." By providing rigorous estimates of diet matrices (and
570 associated uncertainty) based on chemical tracers (eventually combined with expert knowledge
571 and/or external information), diet composition estimates from ESCROC modeling could so be used to
572 calibrate diet matrices in mass-balanced food web models.
573

574 Other perspectives could also relate to mass-balance equations (similar than the ones used in
575 Ecopath) which could also be implemented in the ESCROC model formulation, in order to provide an
576 innovative modeling framework of 'biomass propagation'. This would allow a very integrated view of

aquatic ecosystem food webs, with simultaneous estimations of biomass, contaminants, and isotopes transfers..

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Table 1. Data summary with Average [Min; Max] values for each tracer (5 PFAS, d13C and d15C). n: number of samples on which contaminants and isotopes were measured for each species. – corresponds to censored values (*i.e*. values below the limit of quantification for each specific contaminant). Contaminant concentrations are given in ng g^{-1} wet weight of the whole-body (from Munoz et al., 2017).

Species	L-PFOS (ng g^{-1})		FOSA (ng g^{-1})		PFOA (ng g^{-1})		PFNA (ng g^{-1})		PFUnDA (ng g^{-1})		$\delta^{13}C$ (‰)		$\delta^{15}N$ (‰)		n
Anchovy	3.58	[1.6;6.3]	1.52	[0.9;2.2]	0.12	[-;0.1]	0.25	[0.0;0.1]	0.31	[-;0.2]	-18.71	[-19.4;-18.2]	7.99	[8.8;12.0]	6
Brown shrimp	6.57	[3.8;8.0]	3.75	[1.5;5.4]	0.37	[0.3;0.5]	1.44	[0.8;2.1]	0.51	[0.5;0.6]	-18.07	[-18.7;-17.1]	12.60	[11.5;13.3]	3
Common seabass	6.68	[3.0;14.3]	1.39	[0.4;2.2]	0.30	[-;0.2]	0.39	[0.1;0.5]	0.63	[0.3;1.4]	-17.58	[-19.8;-15.7]	12.65	[10.7;15.0]	9
Copepods	1.24	[0.8;1.6]	0.33	[0.3;0.4]	0.31	[0.1;0.5]	0.56	[-;0.1]	0.32	[0.1;0.2]	-23.43	[-26.4;-21.6]	9.04	[10.6;13.1]	3
Crabs	2.42	[1.8;3.0]	0.22	[0.2;0.2]	2.42	[1.7;3.0]	1.40	[0.8;2.3]	0.31	[-;0.4]	-14.84	[-15.8;-13.3]	6.03	[7.9;10.1]	3
Flounder	5.71	[0.7;21.7]	0.90	[0.1;3.8]	0.40	[-;1.6]	1.18	[0.2;7.9]	0.73	[0.1;1.9]	-18.00	[-23.8;-14.1]	10.53	[10.7;15.2]	13
Gammarids	2.36	[1.5;2.8]	0.52	[0.4;0.7]	1.00	[0.3;2.1]	0.48	[0.3;0.6]	0.44	[0.4;0.5]	-22.51	[-24.2;-19.4]	9.10	[8.4;9.1]	3
Goby	2.35	[2.0;2.4]	0.18	[0.2;0.2]	-	[-;-]	0.14	[0.1;0.2]	0.41	[-;0.4]	-19.21	[-19.2;-19.0]	11.53	[11.2;11.8]	3
Meagre	4.39	[2.5;10.7]	3.30	[2.3;5.4]	-	[-;-]	0.34	[-;0.3]	0.39	[0.2;0.8]	-16.89	[-18.4;-16.3]	14.19	[13.1;14.8]	12
Mullet	2.53	[0.8;4.0]	0.36	[0.1;0.8]	-	[-;-]	0.36	[-;0.3]	0.94	[-;4.0]	-21.55	[-28.2;-16.5]	10.11	[8.7;13.6]	12
Mysids	3.14	[2.4;3.8]	1.20	[0.9;1.5]	0.86	[0.1;0.1]	0.15	[0.1;0.2]	0.15	[0.1;0.2]	-21.35	[-22.8;-19.9]	10.75	[8.2;13.3]	2
Nereis	2.90	[2.0;21.0]	0.59	[0.4;0.8]	5.21	[3.6;8.2]	6.01	[3.7;8.3]	0.49	[0.1;0.3]	-16.40	[-18.2;-15.2]	4.85	[9.9;10.7]	5
Oyster	0.52	[0.1;0.1]	0.74	[0.4;0.8]	0.16	[-;0.0]	0.11	[-;0.0]	0.16	[-;0.0]	-19.66	[-21.2;-18.7]	7.86	[6.1;8.5]	4
Scrobicularia	0.31	[0.2;0.5]	0.27	[0.2;0.3]	0.26	[-;0.0]	0.31	[0.0;0.0]	0.49	[0.0;0.1]	-16.25	[-17.5;-15.5]	7.65	[7.6;7.7]	3
Sole	9.12	[0.7;19.2]	1.22	[0.1;2.4]	0.83	[0.0;2.5]	3.73	[0.2;11.8]	0.47	[0.1;1.3]	-14.19	[-20.8;-14.3]	13.11	[11.1;14.9]	31
Spotted seabass	4.87	[2.2;10.5]	2.22	[1.0;4.2]	0.41	[-;0.7]	0.27	[-;0.7]	0.39	[0.1;1.0]	-16.45	[-20.1;-14.8]	13.78	[11.8;15.4]	28
Sprat	1.64	[0.3;3.8]	3.03	[1.7;4.7]	-	[-;-]	0.15	[-;0.1]	-	[-;-]	-17.19	[-17.4;-16.8]	11.60	[11.3;11.9]	3
White shrimp	3.02	[2.7;3.0]	3.24	[2.7;3.6]	0.39	[0.3;0.4]	0.39	[0.3;0.4]	0.42	[0.3;0.5]	-14.00	[-20.9;-19.4]	8.00	[10.5;11.2]	3

Table 2. Diet matrix of the Gironde estuarine food web. Each value corresponds to the proportion of the prey in line in the diet of the predator in column. It was computed as the median value of the proportion of each of the listed preys in the diet composition of the predators estimated by ESCROC using both N and C isotopic ratios and 5 PFAS concentrations.

Prey \ Predators	Anchovy	Common seabass	Spotted seabass	Flounder	Goby	Meagre	Mullet	Sole	Sprat	White shrimp	Brown shrimp	Mysids	Gammarids	Copepods	Nereis	Crab
Anchovy	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Common seabass	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Spotted seabass	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Flounder	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Goby	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Meagre	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Mullet	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Sole	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Sprat	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
White shrimp	0.00	0.08	0.23	0.00	0.00	0.17	0.00	0.10	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Brown shrimp	0.00	0.09	0.00	0.00	0.00	0.16	0.00	0.13	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Mysids	0.41	0.38	0.15	0.00	0.83	0.11	0.06	0.00	0.00	0.49	0.54	0.00	0.00	0.00	0.00	0.00
Gammarids	0.12	0.29	0.19	0.71	0.17	0.09	0.00	0.47	0.59	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Copepods	0.46	0.00	0.13	0.00	0.00	0.10	0.94	0.00	0.41	0.51	0.46	1.00	0.00	0.00	0.00	0.00
Nereis	0.00	0.06	0.28	0.05	0.00	0.35	0.00	0.12	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Crab	0.00	0.00	0.00	0.22	0.00	0.00	0.00	0.07	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00

Table 3. α_e coefficients (1=benthic, 0=pelagic) estimated by the model (quantiles of the posterior distributions) for the different species and comparison with the values estimated a priori by Muñoz et al. (2017).

	ESCROC (quantiles of posterior distributions)			Muñoz et al. (2017)
Species	2.5%	50%	97.5%	
Anchovy	0.00	0.00	0.00	0.00
Common seabass	0.00	0.06	0.26	0.13
Spotted seabass	0.06	0.28	0.52	0.23
Flounder	0.11	0.29	0.64	0.82
Goby	0.00	0.00	0.00	0.05
Meagre	0.10	0.35	0.60	0.44
Mullet	0.00	0.00	0.00	0.96
Sole	0.04	0.22	0.51	0.67
Sprat	0.00	0.00	0.00	0.00
White shrimp	0.00	0.00	0.00	0.00
Brown shrimp	0.00	0.00	0.00	0.00
Mysids	0.00	0.00	0.00	0.00
Gammarids	0.00	0.00	0.00	0.00
Copepods	0.00	0.00	0.00	0.00
Nereis	1.00	1.00	1.00	1.00
Crab	1.00	1.00	1.00	1.00

Table 4. Estimates and associated 95% -credibility interval of isotopes TEF and contaminants TMF in the investigated Gironde estuary food web with associated biomagnification probabilities. Munoz et al. (2017) are TMF values via LMEC methods when pooling all samples. See text for details.

Tracers	Median	Bounds of the 95% credibility interval		Biomagnification probability	Munoz et al. (2017)
Isotopes TEF					
$\delta^{15}N$	2.76	1.55	3.80	NA	
$\delta^{15}C$	1.60	-0.23	3.09	NA	
Contaminants TMF					
L-PFOS	1.65	0.77	3.28	0.92	1.5 [1.5;1.6]
FOSA	2.29	0.48	6.50	0.86	1.9 [1.9;2.0]
PFOA	0.28	0.04	1.50	0.06	2.0 [1.9;2.1]
PFNA	0.69	0.13	4.22	0.32	1.5 [1.4;1.6]
PFUnDA	1.30	0.47	3.02	0.72	1.1 [1.0;1.2]

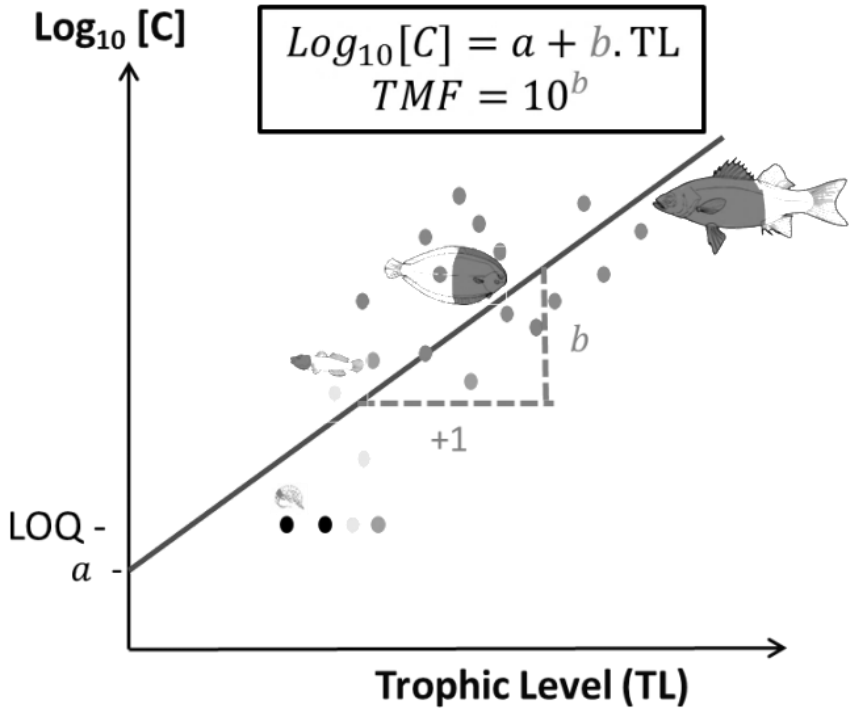


Figure 1. Basis of Trophic Magnification Factor (TMF) estimation. $Log_{10}[C]$: log-transformation of the contaminant concentrations [C]; a: intercept of the regression ; b: slope of the regression. LOQ: limit of quantification. See text for details.

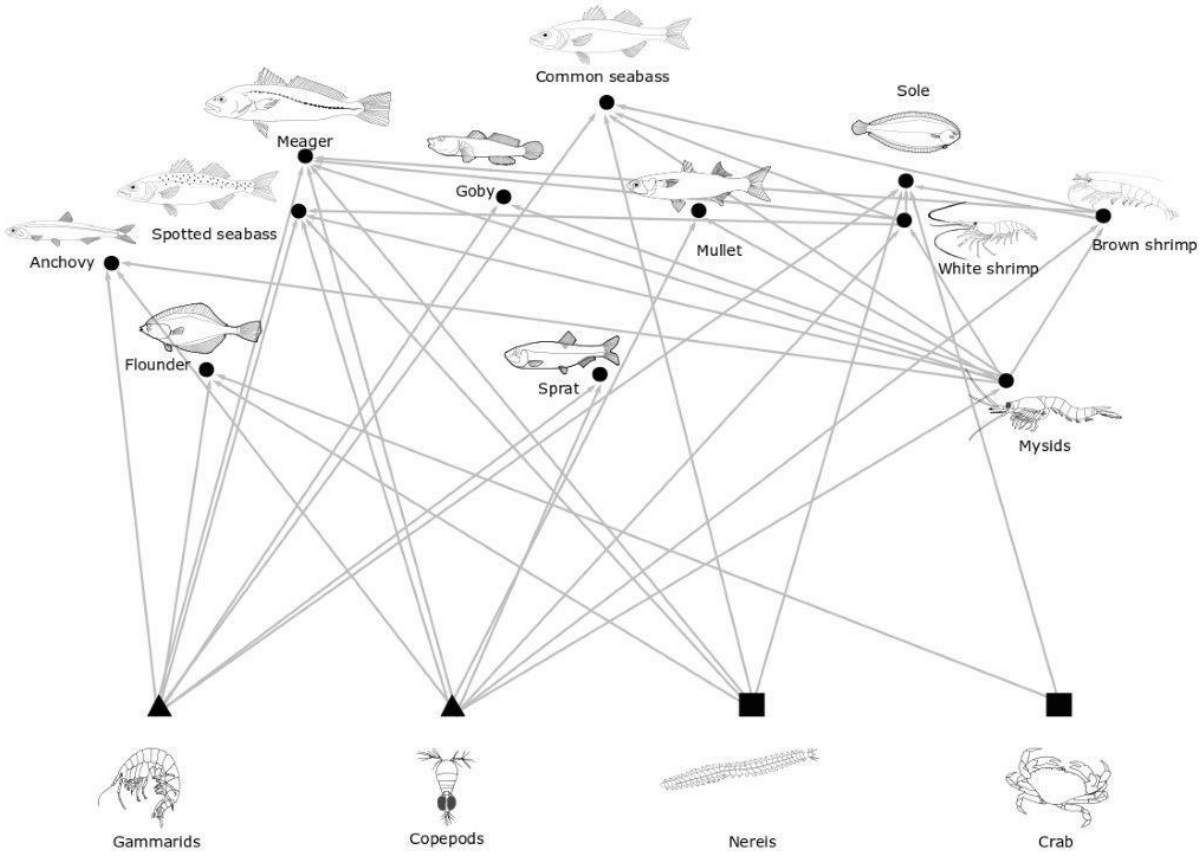


Figure 2. Synoptic diagram of the Gironde estuarine food web (from Pasquaud et al., 2010)

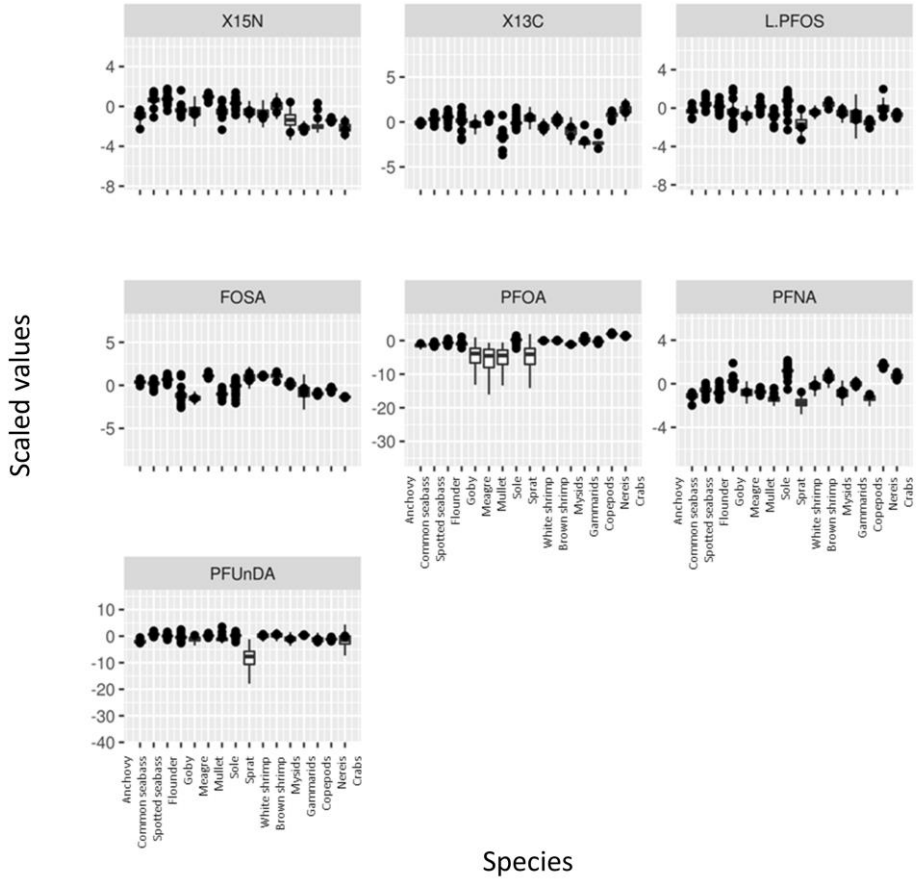


Figure 3. Predicted *posterior* distributions (boxplot) and observations (points) of tracer values by species. Values are scaled (see text for details)

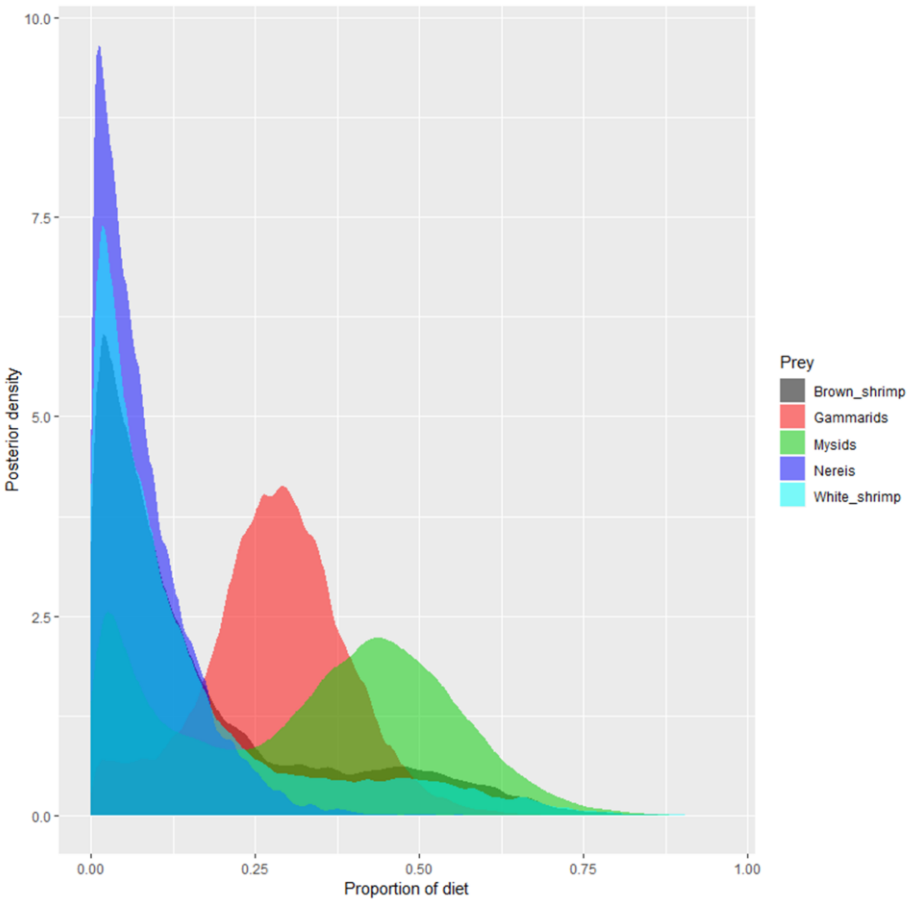


Figure 4. Probability density (in y-axis) of the proportion (in x-axis) of each of the listed preys in the diet composition of the common seabass in the Gironde estuary estimated by ESCROC using both N- and C- isotopic ratios and 5 PFAS concentrations.

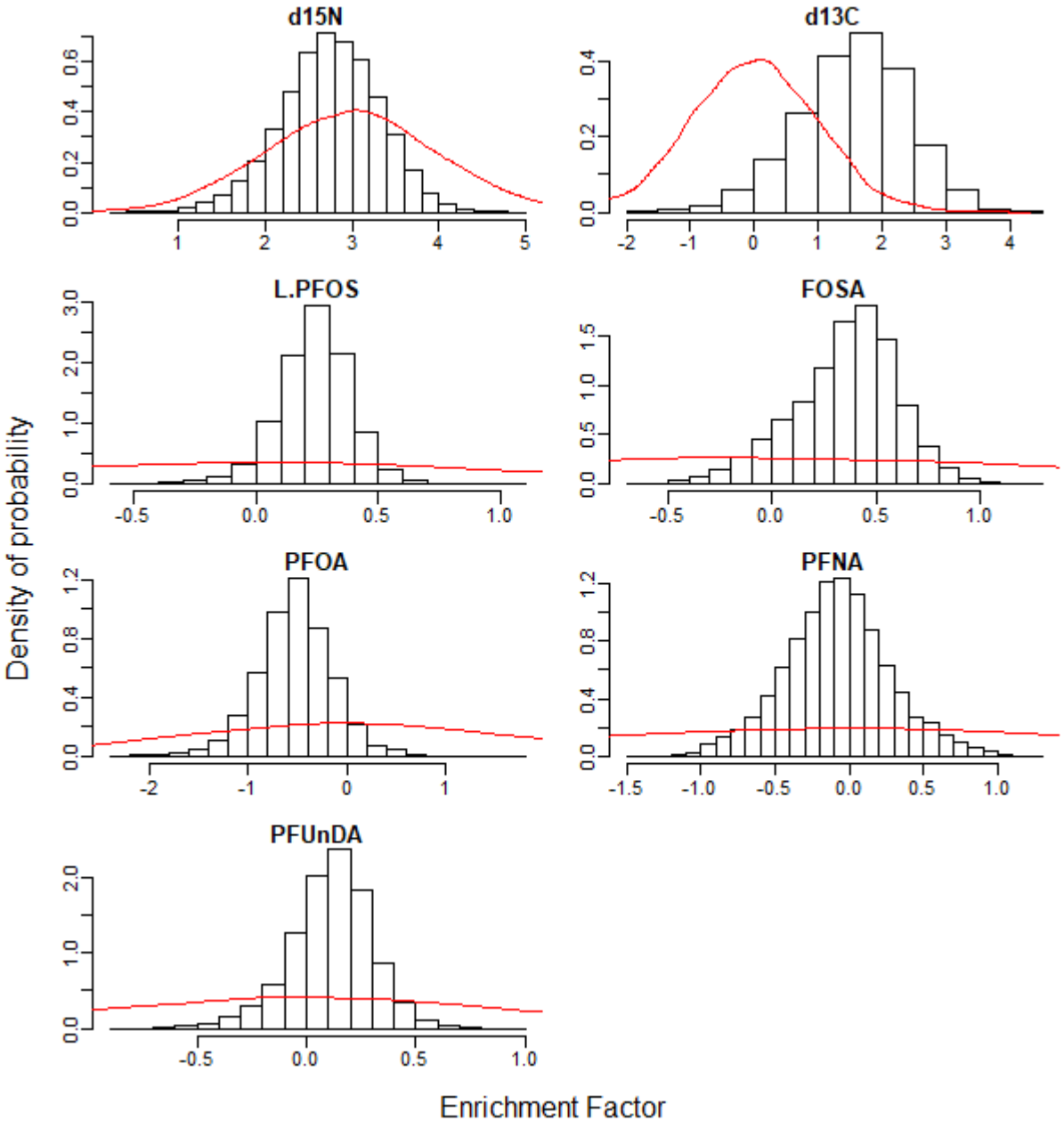


Figure 5. Graphical representation of priors (curves in red) and posteriors (histograms in black) distributions of enrichment factors for each investigated tracer (d15N: $\delta^{15}N$, d13C: $\delta^{13}C$, and contaminants concentrations: L-PFOS, FOSA, PFOA, PFNA and PFUnDA).

Table S 1. List of PFAS compounds targeted in the present study

Acronym	Compound name	Molecular formula
L-PFOS	n-perfluoro-1-octanesulfonic acid	$C_8F_{17}SO_3H$
FOSA	perfluorooctane sulfonamide	$C_8F_{17}\ SO_2NH_2$
PFOA	perfluoro-n-octanoic acid	$C_7F_{15}COOH$
PFNA	perfluoro-n-nonanoic acid	$C_8F_{17}COOH$
PFUnDA	perfluoro-n-undecanoic acid	$C_{10}F_{21}COOH$

S2. Comparisons between ECSROC and MixSIAR

As an illustrative example, we calibrated MixSIAR to estimate the sole diet, using the same data that were used to fit ESCROC.

To be consistent with our ESCROC approach (see text for details),

- We used exactly the same isotope data
- We used exactly the same a prior for diet composition than in ESCROC
- We used two alternative priors for N and C TEFs (Δ_N and Δ_C in Eq 7 and 8 in the text)
    - First, we used exactly the same priors as in ESCROC (see Eq. 7 and 8 in the text): $\Delta_N \sim \mathrm{Normal}(3,1)$ and $\Delta_C \sim \mathrm{Normal}(0,1)$
    - Second, we used priors corresponding to the TEF posterior distributions from ESCROC (see Figure 5 in the text)

Then, we also fitted ESCROC, but, contrary to the article, using only C and N (i.e., ignoring contaminants concentrations) data to directly compare with MixSIAR outputs. Note that with such a limited dataset, ESCROC is likely to be underdetermined.

The results presented in the figure S2 below are based on the direct plotting of the posterior distributions for both ESCROC and MixSIAR. They first highlight that:

(1) Adding contaminants in ESCROC allows to better discriminate the proportions of the main prey in the sole diet (Figures S2 A and C).
(2) Using the posterior distribution of TEF from ESCROC, MixSIAR provides very similar results to ESCROC (Figures S2 A and D).
(3) Using naive priors for TEF and using posterior distributions from ESCROC in MixSIAR provide quite different diet estimates (Figures S2 B and D). This is due to the fact that the naive prior is rather different from the posterior distribution (especially for C). This underlines that MixSIAR is especially sensitive to TEF prior specifications.

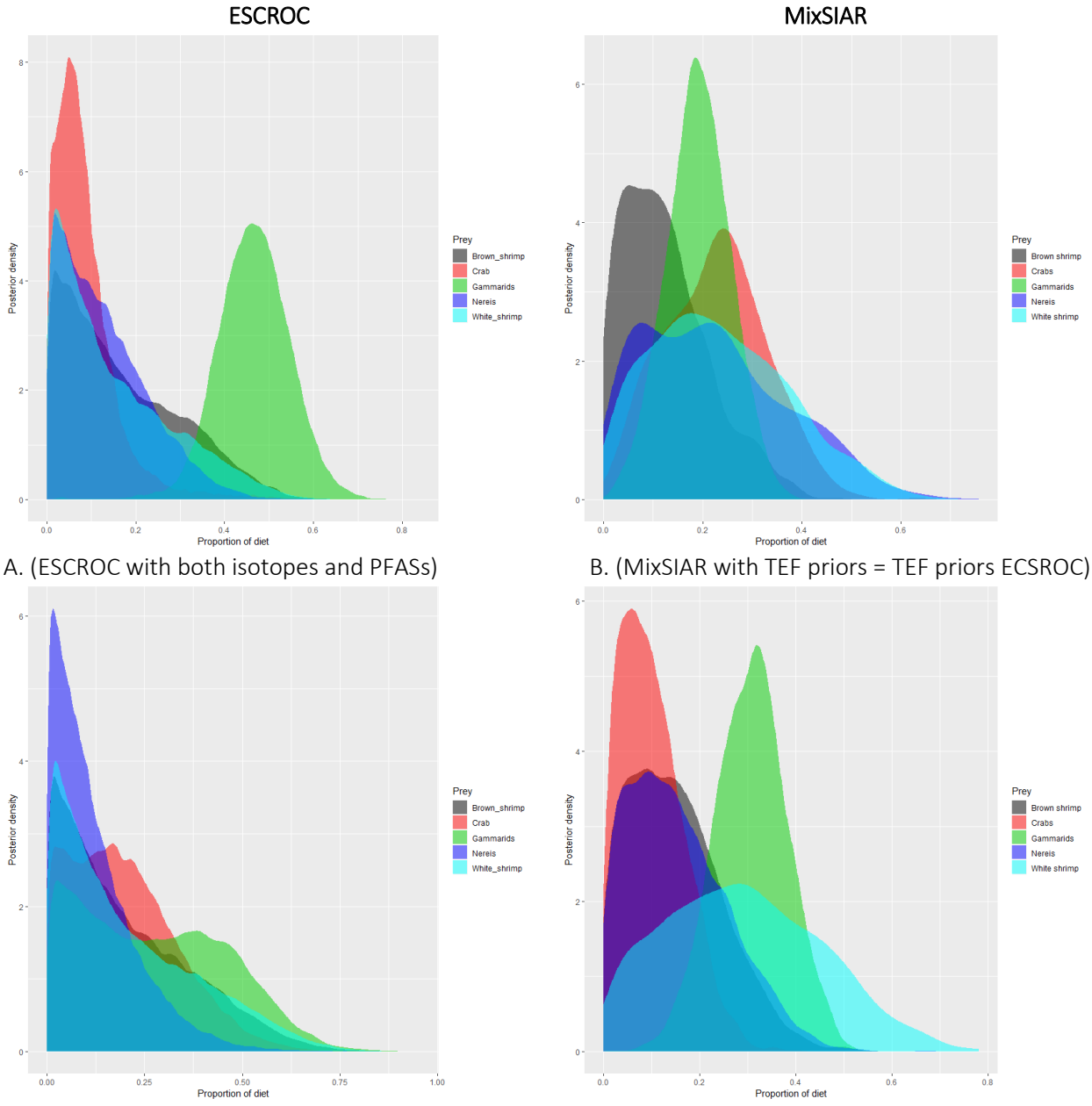
Outputs from ESCROC and MixSIAR (Figures S2 C and D) are different but not contradictory. Actually, they mainly differ in the proportion of white shrimps in the sole's diet. However, the prosterior density is very flat and rather uninformative for this particular species in the MixSIAR outputs and does not allow to really conclude on the proportion of this shrimp in the sole's diet. Indeed, most of the ESCROC simulated results could correspond to the MixSIAR posterior distribution.

Although based on the same transfer equations, these differences between ESCROC and MixSIAR could be explained by:

(1) the fact that ESCROC estimates are computed for the whole food web at one time. This allows using mutual information to compute more accurate estimates.

(2) the use of a random effect 'Species' Thus, although the TEF and TMF estimation are still considered globally constant through the food web, they are modulated species by species. By doing this, we did not specifically consider that isotope discrimination varies with Trophic Position but we allowed the estimation to vary for every species.

(3) a different way to account for uncertainty in both models. In particular, posterior sources and consumers' isotope compositions are computed in ESCROC using both observation data and parameters estimates. This leads to an a posterori quite different set of data between both approaches.



A. (ESCROC with both isotopes and PFASs)

B. (MixSIAR with TEF priors = TEF priors ECSROC)

C. (ESCROC using only C and N isotopes data)

D. MixSIAR (TEF priors = posteriors ESCROC)

Figure S2. Probability density (in y-axis) of the proportion (in x-axis) of each of the listed preys in the diet composition of the sole in the Gironde estuary. Comparisons of outputs from ESCROC (A. based on both isotopes and contaminants data; C. based only on isotopes data) and MixSIAR (B. using initial TEF priors implemented in ESCROC; C. using estimated posterior TEF estimations from ESCROC).