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Assessment of contaminants in blue sharks from the Northeast Atlantic: Profiles, accumulation dynamics, and risks for human consumers☆

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ABSTRACT

Chemical pollution is a major threat to marine ecosystems, and top predators such as most shark species are extremely vulnerable to being exposed and accumulating contaminants such as metals and persistent organic pollutants (POPs). This work aimed to study the degree, composition, and the sources of contamination in the blue shark (*Prionace glauca*) inhabiting the Northeast Atlantic, as well as the potential risk faced by human consumers. A total of 60 sharks were sampled *in situ* aboard fishing vessels, and the concentrations of a set of metals and POPs were analysed in various tissues and complemented with stable isotope analyses. High levels of contaminants were found in most sharks sampled. The concentrations of most metals were higher in the muscle when compared with the liver. Regarding the dangers to consumers posed by the concentrations of arsenic (As), mercury (Hg), and lead (Pb), over 75% of the sharks presented muscle concentrations of at least one contaminant above the legal limits for human consumption, and a risk assessment determined that consumption of meat of these sharks exceeding 0.07 Kg per week could potentially expose human consumers to dangerous amounts of methylmercury (MeHg). Additionally, the assessment of single contaminants may lead to an underestimation of the risk for the human health. Finally, the overall accumulation of contaminants seems to be mostly influenced by the sharks' geographical distribution, rather than sex, size, or trophic level of their prey.

1. Introduction

Marine ecosystems constantly face contaminants input deriving mostly from the world's industries (Wilhelmsson et al., 2013), with waters closer to more urbanized and industrialized areas (i.e., each country's Exclusive Economic Zone (EEZ)) being, in theory, more susceptible to presenting higher levels of contamination when compared with more oceanic areas. Metals such as arsenic (As), lead (Pb), and mercury (Hg) are notorious for their negative health impacts (Boening, 2000; Ishaque et al., 2020; Neff, 1997), but other less studied chemical elements have been receiving increased attention by researchers studying teleosts (da Silva et al., 2022) and elasmobranchs (Hauser-Davis et al., 2021). The class of persistent organic pollutants (POPs) also represents a substantial threat to marine ecosystems (Wenning and Martello, 2014), particularly contaminants such as polychlorinated biphenyls (PCBs), polybrominated diphenyl ethers (PBDEs), and polychlorinated dibenzo-p-dioxins and polychlorinated dibenzo-p-furans (PCDD/Fs). In the oceans, metals and POPs progressively accumulate in marine organisms, potentially impairing their health.

Due to particular biological characteristics, some organisms (i.e. top predators) are particularly susceptible to accumulate these contaminants through bioaccumulation (Gray, 2002) and biomagnification (Suedel et al., 1994). Consequently, many shark species have elevated concentrations of the aforementioned contaminants (Tiktak et al., 2020), suffering impacts in many different processes essential for their health (Alves et al., 2022). Consales and Marsili (2021) have alerted to

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the lack of information regarding pollution and its threats to sharks in the IUCN Red List assessments and for the importance of updating their conservation status having into consideration this extra threat sharks are facing, given their importance to the homeostasis of their respective ecosystems (Ferretti et al., 2010), and the fact that numbers of these fishes have been declining for decades (Pacoureau et al., 2021). Additionally, shark meat is eaten by human populations around the world (Dent and Clarke, 2015), making the levels of contaminants in these animals a public health concern. This tendency to accumulate detectable and often elevated concentrations of contaminants, as well as their ecological and socioeconomic relevance, makes sharks suitable target species for biomonitoring studies (Alves et al., 2022). The blue shark (*Prionace glauca*) is one of the most landed and consumed sharks worldwide (Dent and Clarke, 2015), including in Portugal (Alves et al., 2020) where the species is consumed domestically and also exported to countries like Spain and Italy. The contaminants in these sharks may therefore impact human health. Since Portugal's fish consumption *per capita* is one of the highest in the world (FAO, 2020), consumers may be particularly exposed to contamination. A recent systematic review by TikTak et al. (2020) found blue shark to be the most targeted species by researchers investigating contaminants in elasmobranchs, with some studies reporting worrying concentrations. Case in point, a previous study found that juvenile blue sharks landed in Portuguese ports possessed levels of contaminants (i.e. Hg) above the maximum values deemed safe and legal for human consumption (Alves et al., 2016). More recently, Muñoz-Arnanz et al. (2022) reported elevated concentrations of POPs in blue sharks captured in the Northeast Atlantic and stated that contamination seemed to have increased when compared with previous results (Alves et al., 2016).

Although an increasing number of studies have been tracking the concentrations of pollutants in the tissues of these sharks, much less is known about how these contaminants behave along spaciotemporal scales in marine ecosystems. Understanding where, when, and how the animals accumulate contaminants is an important part of tackling the marine pollution issue, to better protect both the ecosystems and fish consumers. Assimilation techniques such as stable isotope analysis (SIA) are valuable tools in this regard, and may be used to highlight patterns of feeding that contribute to contaminant accumulation (Hussey et al., 2012). Because in elasmobranchs the muscle has been reported to have a turnover rate of isotope assimilation of about 420 days, while blood's is about 280 days (Matich et al., 2011), the combined use of these tissues has the potential to give insight on feeding patters during different timescales (potentially with some overlap).

The present work aims to build on the findings of Muñoz-Arnanz et al. (2022) and assess other relevant contaminants in the same sharks, as well as study the factors that may have influenced the detected concentrations in both works. Therefore, the main aims of the present work were to 1) assess contamination levels in blue sharks destined to human consumption, 2) estimate the risk of exposure to human consumers, and 3) identify potential physiological, and environmental variables affecting the accumulation of different contaminants. To do this, metals, POPs, and stable isotope ratios (i.e., $\delta^{13}C$, and $\delta^{15}N$) were assessed in samples collected from blue sharks varying in size, sex, and location of capture.

2. Material and methods

2.1. Sampling

Between March and December 2019, 60 blue sharks were sampled aboard a commercial longliner targeting swordfish in the Northeast Atlantic. Individuals were caught as bycatch and sampled opportunistically after being sacrificed by fishermen upon landing on vessel. Samples for contaminant assessment (liver and muscle) and SIA (blood and muscle) were collected from each specimen. Samples for the analysis of metals were collected using ceramic cutlery and stored in plastic tubes. Samples used for the analysis of POPs were collected using inox cutlery, rinsed with acetone, wrapped in tin foil and stored in individual tubes (Muñoz-Arnanz et al., 2022). The sampling station and all equipment used were thoroughly cleaned with ethanol between each dissection. All sampled individuals' size, sex, and position of capture were recorded, and samples were stored at −20 °C. Specimens were categorized according to sex (male and female) and sampling location (inside EEZ, ≤ 200 nautical miles (NM) from shore – inside Portugal's EEZ; outside EEZ, > 200 NM from shore – outside Portugal's EEZ, Fig. 1). Additionally, based on the methodology used by Vandeperre et al. (2014a), samples were categorized into different size/maturity groups based on fork length (FL): small juveniles (SJ, <130 cm FL), large juveniles (LJ, 130 cm ≥ FL < 183 cm, for males, and 130 cm ≥ FL < 180 cm, for females), and adults (AD, ≥183 cm FL, for males, and ≥180 cm FL, for females). The group of sampled individuals comprised 38 males and 22 females. The mean size (FL) was 161.6 ± 45.9 cm and ranged between 101 cm and 251 cm. Males included 4 small juveniles, 17 large juveniles and 17 adults, and females comprised 12 small juveniles, 10 large juveniles and no adults. Overall, 40 sharks were sampled inside EEZ, and 20 outside EEZ.

2.2. Contaminant chemical analysis

For the analysis of metals, white muscle and liver samples from each specimen were freeze dried before being digested with an acid mixture composed by HNO_3:$HClO_4$ (7:1) in a total volume of 2 mL of final reaction mixture. Mineralization procedure occurred in Teflon reactor at 110 °C for 3 h. After mineralization, an ultra-pure acid Gallium solution (internal standard) was added to the samples. Then, 5 μL of the mineralization product were placed in a siliconized quartz disc and evaporated at 80 °C. Quartz discs holding evaporated samples were then placed in a sample carrier, along with three reference samples (As, Ni, and a multi-element sample). Samples were analysed using a Total X-Ray Fluorescence Spectrometer Bruker Picofox S2 (Bruker Nano Analytics, Germany), with a measuring time of 800 s per sample, using gallium as internal standard. For each specimen, the concentrations of silver (Ag), aluminium (Al), As, calcium (Ca), cadmium (Cd), cobalt (Co), chromium (Cr), copper (Cu), iron (Fe), Hg, potassium (K), manganese (Mn), sodium (Na), nickel (Ni), phosphorus (P), Pb, sulphur (S), antimony (Sb), selenium (Se), tin (Sn), strontium (Sr), titanium (Ti), vanadium (V), and zinc (Zn) were assessed. Concentration values were reported as $\mu g \cdot g^{-1}$ of dry weight (dw). Extraction efficiency was confirmed through the analysis of International certified reference materials (ERM-BB422 Fish Muscle), being all analysed values within the certified values range. Although a suitable standard reference material for Hg was unavailable, our methodology and results compare well with what has been generally practiced in similar studies analysing metals in fish muscle by Total Reflection X-ray Fluorescence spectroscopy (TXRF) (e.g. da Silva et al., 2020; Duarte et al., 2022). The method's mean limits of detection (LOD) values for each element can be seen in Table S1. For more details on the methodological procedures and on the analytical accuracy of the reference material, see TXRF technical report as supplementary material. For the comparison with legislated recommended maximum levels for human consumption and with mean concentrations reported in other studies, the concentrations detected in this work were converted to $\mu g \cdot g^{-1}$ of wet weight (ww) using the conversion factor of 0.25 (Hauser-Davis et al., 2021). Since Se can counteract some of the negative effects of Hg (Ibrahim et al., 2019; Ralston and Raymond, 2010), the Se/Hg molar ratios were calculated for the muscle and liver samples using the total Hg, as described by Burger and Gochfeld (2013).

The methodological procedures for the POP analyses are described in detail in Muñoz-Arnanz et al. (2022). Succinctly, approximately 1.5 g of liver were analysed by gas chromatography using a high-resolution mass spectrometer (GC-HRMS) to assess the concentrations of a total of 61 POPs (6 non-dioxin-like PCBs (NDL-PCBs), 12 dioxin-like PCBs (DL-PCBs), 26 PBDEs, 7 polychlorinated dibenzodioxins (PCDDs), and

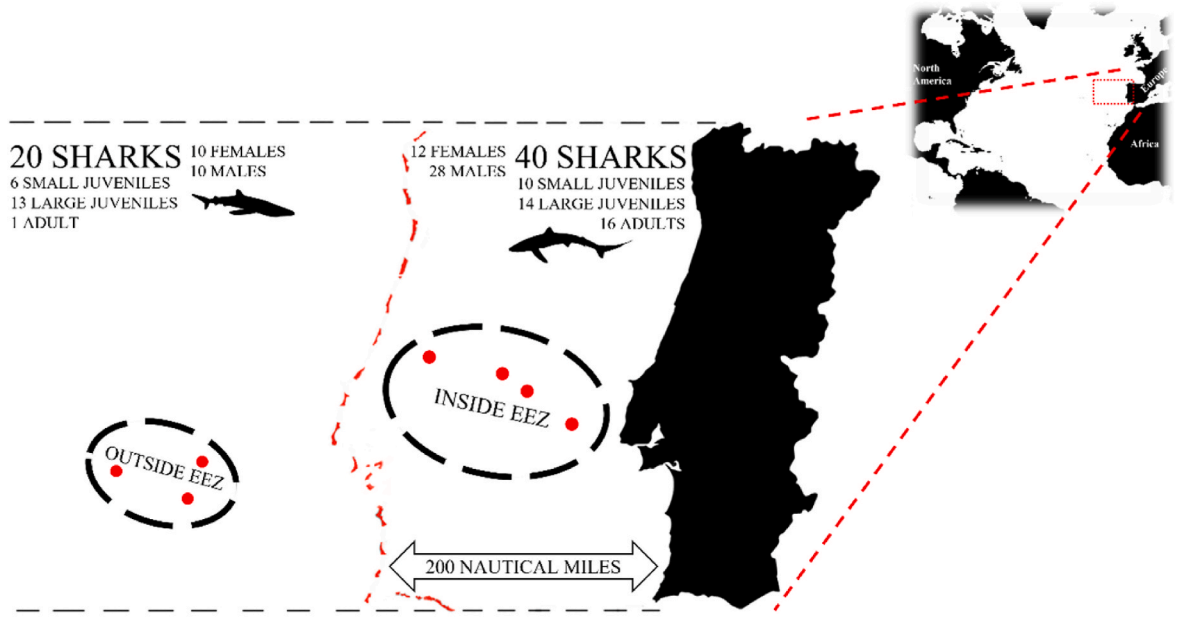

Fig. 1. Illustration of the sampling zones in the Northeast Atlantic Ocean, off the coast of Portugal (inside EEZ and outside EEZ) and descriptive summary of the number of blue sharks (*Prionace glauca*) sampled in each zone in relation to sex (males and females) and size/maturity groups (adults, large juveniles and small juveniles). Red dots represent individual sampling spots. (For interpretation of the references to colour in this figure legend, the reader is referred to the Web version of this article.)

10 polychlorinated dibenzofurans (PCDFs)). For this work, only the sums of the main families of POPs measured were used. Concentration values were reported as $pg{\cdot}g^{-1}$ of wet weight (ww).

2.3. Stable isotopes analyses (SIA)

Stable isotopes of carbon ($\delta^{13}C$) and nitrogen ($\delta^{15}N$) were analysed in the muscle and blood of each specimen. Samples were lyophilized and grounded to fine powder in a mill (Mixed Mill MM 400 (Retsch, Haan, Germany)). Lipids were removed from muscle by successive rinses in a 2:1 chloroform-methanol solution, and dried at 50 °C for 48 h. The C:N mass ratios of blood (mean ± SD: 2.8 ± 0.2) and delipidated muscle (3.1 ± 0.1) indicate low lipid content (i.e. < 3.5) of the samples analysed (Post et al., 2007). For each isotopic ratio, approximately 0.3 mg of powdered tissue were loaded into small tin cups, and then combusted at 1800 °C using a Flash EA1112 Series elemental analyser (Thermo Italy, Rhodano, Italy) coupled on line via Finnigan ConFlo II interface to a Thermo Delta V mass spectrometer (Bremen, Germany). Isotope ratios are presented in the usual δ notation based on the Vienna-PeeDee Belemnite (V-PDB) for carbon, and atmospheric N_2 (AIR) for nitrogen and expressed as ‰. $\delta^{13}C$ or $\delta^{15}N = [(Rsample/Rstandard) - 1]$, where R = 13C/12C or 15N/14N, respectively. A precision of <0.2‰ for both $\delta^{13}C$ and $\delta^{15}N$ values were obtained by measuring replicates of internal laboratory standards (acetanilide).

2.4. Statistical analyses

Density plots and Shapiro-Wilks' tests were used to assess the normality of contaminant concentration data. Given the non-normal distribution, non-parametric tests were performed to assess the existence of correlations (Kendal's Tau) and statistical differences (Wilcoxon rank sum) between the accumulation levels and the variables under study. To highlight the differences between the concentrations of metals measured in muscle and liver tissues, the fold change (FC) was calculated according to Marques et al. (2021) as [(muscle mean concentration/liver mean concentration)-1]. A fold change of 1 means that the mean concentration for that element in muscle is double of what was verified in liver, whereas a value of −1 is indicative of the opposite (i.e., the negative values mean that the concentration was higher in the liver X number of times). Stable isotope data were analysed in the context of isotopic niche width using a Bayesian framework (Stable Isotope Bayesian Ellipses in R: SIBER; Jackson et al., 2011). The area of the standard ellipse (SEAc, an ellipse obtained by Bayesian inference containing 40% of the data regardless of sample size and corrected for small sample sizes) was adopted to compare niche width between groups (i.e., sex, size/maturity, and zone of capture).

The significance level for all statistical tests was set at $p < 0.05$. Due to the complex and constrained nature of our sample set, the existence of different accumulation patterns within sex, size/maturity, and zone of capture was investigated using Principal Component Analysis (PCA). Where applicable, results are expressed as mean ± standard deviation (SD). All statistical analysis were performed on R Version 4.0.3 (R Core Team, 2020), using the integrated development environment RStudio Version March 1, 1093 (RStudio Team, 2020).

Following the recommendations made by the European Food Safety Authority (EFSA, 2010), an alternative approach to traditional substitution methods was used to analyse elements presenting left-censored data (i.e., concentrations below the LOD). Packages in R created specifically for the assessment of left-censored data were used, namely the NADA (Lee, 2020) and NADA2 (Helsel, 2021). To compare our data with examples found in the literature, summary statistics (i.e., mean and SD) for metals with percentages of concentrations below the LOD <80% were estimated applying the Kaplan-Meier method using the "cenfit" function form the NADA package. For metals with percentages of concentrations below the LOD levels ≥80% and <100%, the mean concentrations of the samples with detectable concentrations were used (along with the indication of respective uncensored number of samples). Non-parametric methods were applied to test for the existence of correlations (Kendal's Tau) and differences (Peto-Peto), using the "ATSmini" and "cen1way" functions from the NADA2 package, respectively. As the NADA and NADA2 packages do not allow to perform a PCA, and this multivariate analysis requires a complete database without empty cells to perform the correlation matrix (Kleinbaum and Kupper, 1978), a substitution method was used: censored values were substituted by LOD/2, a method proven to be adequate when performing PCA (Farnham et al., 2002). To reduce the potential bias associated with

substitution methods, only elements presenting ≤20% censorship were used.

2.5. Risk assessment for human health through dietary exposure

To assess the safety of blue shark meat consumption, the levels of As, Hg, and Pb measured in white muscle were compared with limits stipulated by international regulatory agencies (European Council, 1993; FAO/WHO, 2016; National Research Council of the National Academies, 2014) and used to calculate risk factors, to estimate whether consumption poses a risk to consumers. The values determined for As and Hg were first converted into inorganic arsenic (iAs) and methylmercury (MeHg), respectively. This conversion was made based on previous studies which estimated that iAs represent in average 3% of total As in shark muscle (Bosch et al., 2016; Denobile, 2007; Muñoz et al., 2000), and that MeHg accounts for 90% of total Hg (Branco et al., 2007; Kim et al., 2016; Storelli et al., 2001).

Since the consumption of blue shark meat can be estimated and considering the contamination of the sharks, the safe limits for consumption of these sharks' meat were determined. Fig. 2 summarizes the various parameters involved in the risk assessment. The provisional tolerable weekly intake (PTWI) and the hazard quotient (HQ) for the Portuguese population were estimated as in Marques et al. (2021). The estimated PTWI was calculated as PTWI = (c x AvC)/bw, where c is the detected mean contaminant concentration, AvC is the average fish consumption per week in Portugal, and bw represents the average body weight on a European adult (i.e., 70 kg) (EFSA, 2012). The HQ was calculated as HQ = EDI/RfD, where EDI is the estimated daily intake (calculated as PTWI/7), and RfD is the oral reference dose of the U.S. EPA for each element (0.3 μg kg bw^{-1}. day^{-1} for iAs, and 0.1 and MeHg (IRIS, 2022)).

The PTWI and the HQ were estimated for the Portuguese population using both a worst case scenario approach (i.e., assuming all the fish consumed was blue shark), and using a more realistic estimate of the blue shark consumption. For the worst case scenario, a *per capita* apparent consumption of fishery and aquaculture products of 60.92 kg $year^{-1}$ (and 1.168 kg $week^{-1}$; data from 2018) was considered (European Commission Directorate-General for Maritime Affairs and Fisheries, 2021). The estimation of real blue shark consumption values was done using data available for this species provided by Statistics Portugal (INE, 2021). Using 2018 as the reference year, the consumption of blue shark meat in Portugal was estimated according to EUMOFA (2020), using the following formula: Estimated Consumption = (Landings + Imports) - Exports. The obtained value of 133 tonnes corresponds to 0.021% of the total fish and seafood supply in Portugal (i.e. 626 000 tonnes $year^{-1}$ (European Commission Directorate-General for Maritime Affairs and Fisheries, 2021; FAO, 2020)). The obtained PTWI values were compared with the available limits recommended by FAO/WHO (FAO/WHO CAC, 2018). Regarding HQ, a value < 1 means that adverse health effects are not expected, while a HQ ≥ 1 indicates potential health risks (EPA, 2011). Finally, the maximum safe weekly consumption (MSWC) was calculated using the equation MSWC = (bw x maximum recommended PTWI)/c, following the method described by Chouvelon et al. (2009), where bw is the average body weight on a European adult (i.e., 70 kg) (EFSA, 2012), PTWI is a) for MeHg, the recommended maximum level of 1.6 μg kg bw^{-1}. $week^{-1}$(JECFA, 2007); b) for iAs, the reference dose of 0.3 μg kg bw^{-1}. day^{-1} (EPA, 2002) multiplied by 7 for a result per week (since the maximum recommended PTWI for iAs was withdrawn (EFSA, 2009)), and C represents the detected mean element concentration.

3. Results

3.1. Contaminant levels

A descriptive summary of the results for all metals measured in the blue sharks' muscle and liver samples, divided per sampling zone, sex and size group, are presented in Supplementary Table S2 and Supplementary Table S3, respectively. The presence of Ag and Cd was not detected in any of the tissues for any specimen and, therefore, those elements were removed from the aforementioned tables. The concentrations of Sb and V were also 100% below LOD in the liver samples and were therefore removed from Supplementary Table S3. Additionally, concerning all samples and both tissues analysed, Al, Co, Cr, Mn, Ni, Pb, Sb, Sn, Sr, and V presented percentages of censorship that varied between 3.3 and 96.7% (Table S1, supplementary material). In relative terms, the accumulation patterns of the chemical elements were very similar for both tissues analysed, with both tissues presenting the same detected elements with the four highest mean concentrations (i.e., Na, K, P, and S) and the lowest (i.e., Co, Cr, Mn, and Ni). However, most elements presented significant differences in absolute concentration between tissues ($p < 0.05$), preferably accumulating in the muscle tissue (Fig. 3).

All sharks presented negative Se/Hg ratios in the muscle, and only

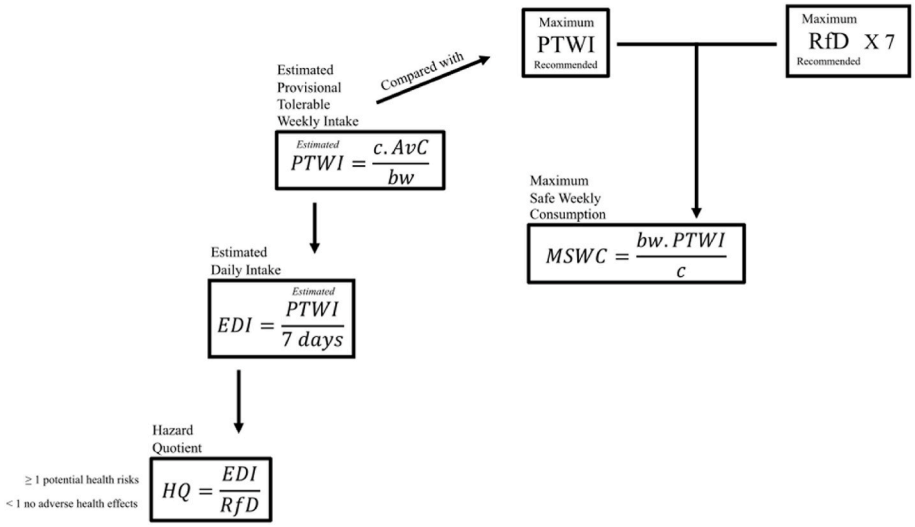

Fig. 2. Diagram explaining the various parameters involved in the health risk assessment. PTWI – Provisional Tolerable Weekly Intake; EDI – Estimated Daily Intake; HQ – Hazard Quotient; MSWC – Maximum Safe Weekly Consumption; c – mean element concentration in shark muscle; AvC – average fish consumption per week; bw – average body weight for European adults; RfD – reference dose for oral exposure.

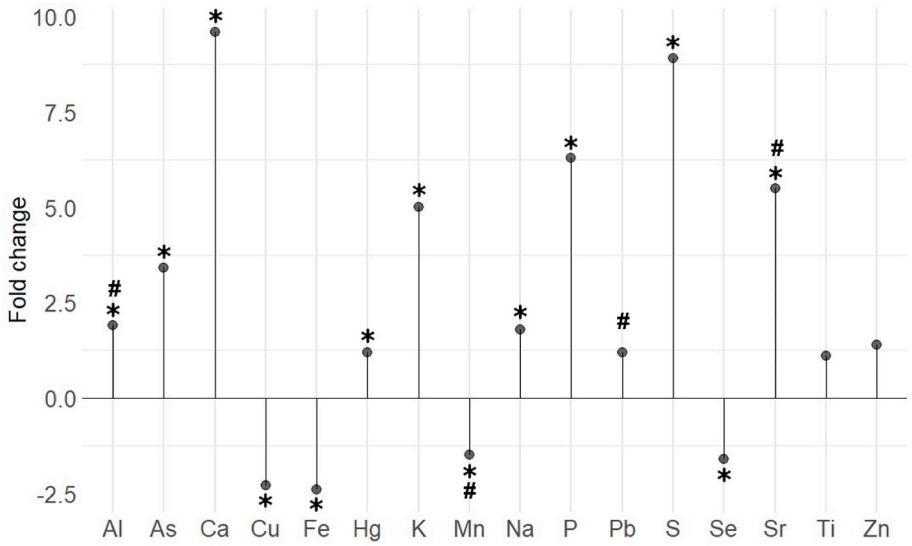

Fig. 3. Differences between mean values of elements measured in muscle and liver samples of blue sharks (*Prionace glauca*), represented by fold change. Fold change of 1 means that the mean concentration for that element in muscle is double of what was verified in liver, whereas a value of −1 is indicative of the opposite. * indicates significant differences (Wilcoxon rank sum for non-censored elements, Peto-Peto for censored elements, all $p < 0.05$). # indicates censorship. Only elements presenting less than 50% censorship on each tissue were included.

two countered that result in liver (Supplementary Tables S2 and S3). Regarding POPs, the concentrations for the sum of the different congeners within the main POP families measured in liver samples can be seen in Supplementary Table S4 (only values measured in liver are presented given its higher relevance as preferential accumulation tissue; more details on POPs measured in the sampled sharks can be found in Muñoz-Arnanz et al., 2022). The POPs with the highest mean concentrations were NDL-PCBs (151,000 pg g^{-1} ww), and DL-PCBs (72,000 pg g^{-1} ww), followed by PBDEs (15,000 pg g^{-1} ww), PCDFs (8.3 pg g^{-1} ww) and PCDDs (2.5 pg g^{-1} ww).

3.2. Human health risk assessment

The concentrations of As, Hg, and Pb detected in each shark's muscle were analysed from a consumer's health perspective and compared with relevant legal limits for direct human consumption and food production. Regarding As, 3 out of 60 samples presented concentrations above the 25 μg g^{-1} ww limit imposed in the E.U. for fish-based feed according to the Commision Directive (EU) 2009/141/EC (2009). A total of 32 out of 60 samples were above the Hg legal limit for human consumption of 1.0 μg g^{-1} ww, for high-level pelagic predators (like sharks) established in Council Regulation (EU) No 1881/2006 (2006). The mean Hg value was almost double the limit (i.e., 1.9 μg g^{-1} ww), with a maximum of 13.3 μg g^{-1} ww. Concerning Pb, 35 out of 60 samples surpassed the legal threshold of 0.30 μg g^{-1} ww in muscle meat of fish, with mean and maximum concentrations of 0.7 μg g^{-1} ww and 3.1 μg g^{-1} ww, respectively. The overall distribution of the concentrations of Hg and Pb can be seen in Fig. 4, and in both cases the median concentrations detected were above the legal limits for human consumption (represented by the dotted lines).

The calculated human health risk parameters for iAs, MeHg, and Pb can be seen in Table 1. When assuming a fish diet 100% comprised of blue shark meat, iAs and MeHg presented values of PTWI and HQ above those considered safe. Using a more conservative and realistic approach, considering the estimated real consumption of blue shark meat in Portugal (i.e., 0.021% of total fish consumption), in the unlikely event that consumption is evenly distributed throughout all population, both PTWI and HQ for all elements were under the set safety limits. Although the PTWI safe limits for Pb have been withdrawn it is unclear if the values found may pose a risk for consumers. Considering the mean concentrations detected in the shark samples, the maximum safe weekly consumption (MSWC) of this fish meat for iAs and MeHg would be around 0.55 kg $week^{-1}$ and 0.07 kg $week^{-1}$, respectively.

A detailed analysis on the safety of the POP levels in the shark samples has been described in detail in Muñoz-Arnanz et al. (2022). Overall, results showed that more than half of liver samples exceeded the maximum levels allowed for NDL-PCBs, DL-PCBs, and PCDD/Fs,

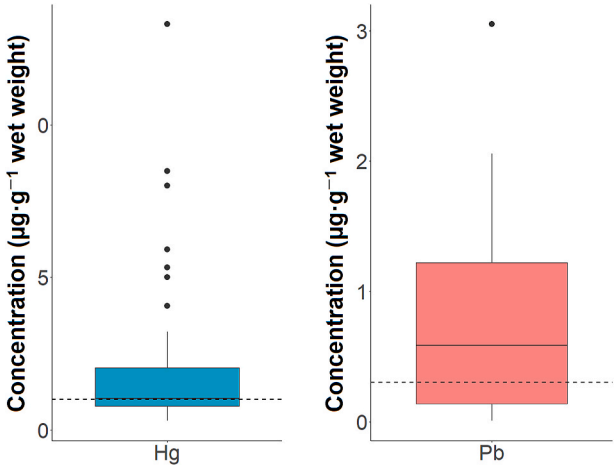

Fig. 4. Boxplots of concentrations of mercury (Hg) and lead (Pb) detected in the muscle of blue sharks (*Prionace glauca*) captured in the Northeast Atlantic. Dotted lines represent legal limits for human consumption (Hg = 1.0 μg g^{-1}, Pb = 0.3 μg g^{-1}, Commission Regulation1881/2006). (For interpretation of the references to colour in this figure legend, the reader is referred to the Web version of this article.)

Table 1
Human health risk assessment calculations for the consumption of blue shark (*Prionace glauca*) meat (present study) and maximum safe limits used in each metal comparison or risk calculation. All values are in wet weight. iAS = inorganic arsenic; MeHg = methylmercury; Pb = total lead. PTWI = Provisional Tolerable Weekly Intake; HQ = Hazard Quotient. Results exceeding the regulatory limits or with a HQ > 1 are highlighted in bold.

Metal	PTWI (μg kg bw^{-1} $week^{-1}$)			HQ	
	Maximum safe limit (FAO/WHO CAC, 2018)	100% Blue shark	0.021% Blue shark	100% Blue shark	0.021% Blue shark
iAs	withdrawn	4.5	0.0009	**2.1**	0.0005
MeHg	1.6	**28.6**	0.006	**40.9**	0.009
Pb	withdrawn	11.83	0.0025		

which may be of importance to account when considering the consumption of oil-related products.

3.3. Demographic analysis of contamination

To assess the relationship between the contaminant levels of the sharks and the factors that may have contributed to differences in their accumulation, different multivariate analyses were performed using the contaminant accumulation data on the tissues where contaminants accumulated preferentially (i.e., muscle for metals, and liver for POPs). Regarding the metals, several biplots were created for the same PCA analysis, each emphasizing a different factor (i.e., sex – Fig. 5A, size/maturity – Fig. 5B, and zone of capture – Fig. 5C). Axis 1 and 2 of the PCA explained most of the variability observed (82.8%). Axis 1 was influenced by the concentrations of all elements excluding Pb (which was responsible for most of the variability observed in axis 2). The analysis also indicated strong positive correlations between most of the chemical elements, being Pb the one with the most different accumulation behaviour. The PCA analysis showed no relevant patterns of accumulation associated with differences in sex (Fig. 5A). Size/maturity also appeared to have no clear influence on element accumulation (Fig. 5B), something that was corroborated by the lack of correlations between the size of the sharks and the concentrations of metals measured in the muscle samples (Kendal's Tau, all $p > 0.05$). However, differences between animals caught inside EEZ and outside EEZ were apparent (Fig. 5C). The disposition of the vectors (metals) in relation to the dotted samples indicates that the animals from inside EEZ (i.e., closest to the shore) tend to have higher concentrations of metals in their muscle, when compared to the animals from outside EEZ. One sided Wilcoxon rank sum tests ($p < 0.05$) showed that sharks caught inside EEZ presented significantly higher mean concentrations of Cu (W = 732, $p = 4.3e\text{-}09$), Fe (W = 653, $p = 1.7e\text{-}05$), Na (W = 643, $p = 3.8e\text{-}05$), Se (W = 676, $p = 2.4e\text{-}06$), and Ti (W = 589, $p = 1.3e\text{-}0.3$).

The biplots resulting from the multivariate analysis regarding the POP concentrations measured in the liver samples can be seen in Fig. 6. The first two axis of the PCA biplot also explained most of the variability (72.1%). POPs were displayed into two different and mostly unrelated groups: one formed by PCBs (both dioxin like and non-dioxin like) and PBDEs, and another formed by PCDDs and PCDFs. The biplots did not show a clear separation between sex groups (Fig. 6A), nor in relation to size/maturity (Fig. 6B), although positive correlations could be found between the size of the sharks and the concentrations of PCBs, PBDEs, and PCDD/Fs (Muñoz-Arnanz et al., 2022). In accordance with the observations for the metals concentrations in muscle, differences between zones of capture seemed to impart the most influence (Fig. 6C), with higher concentrations being also associated with animals caught inside the EEZ. One sided Wilcoxon rank sum tests ($p < 0.05$) revealed significantly higher concentrations of NDL-PCBs (W = 650, $p = 2.2e\text{-}05$), DL-PCBs (W = 605, $p = 5.1e\text{-}04$) and PBDEs (W = 617, $p = 2.4e\text{-}04$) in sharks from inside EEZ. Although not significant, PCDFs were also tendentially higher in sharks from inside EEZ (W = 505, $p = 5.1e\text{-}02$).

3.4. Isotopic profiles

The isotopic patterns were very similar in both tissues assessed and were strongly and positively correlated ($\delta^{13}C$: tau = 0.611, $p = 5.4e\text{-}12$;

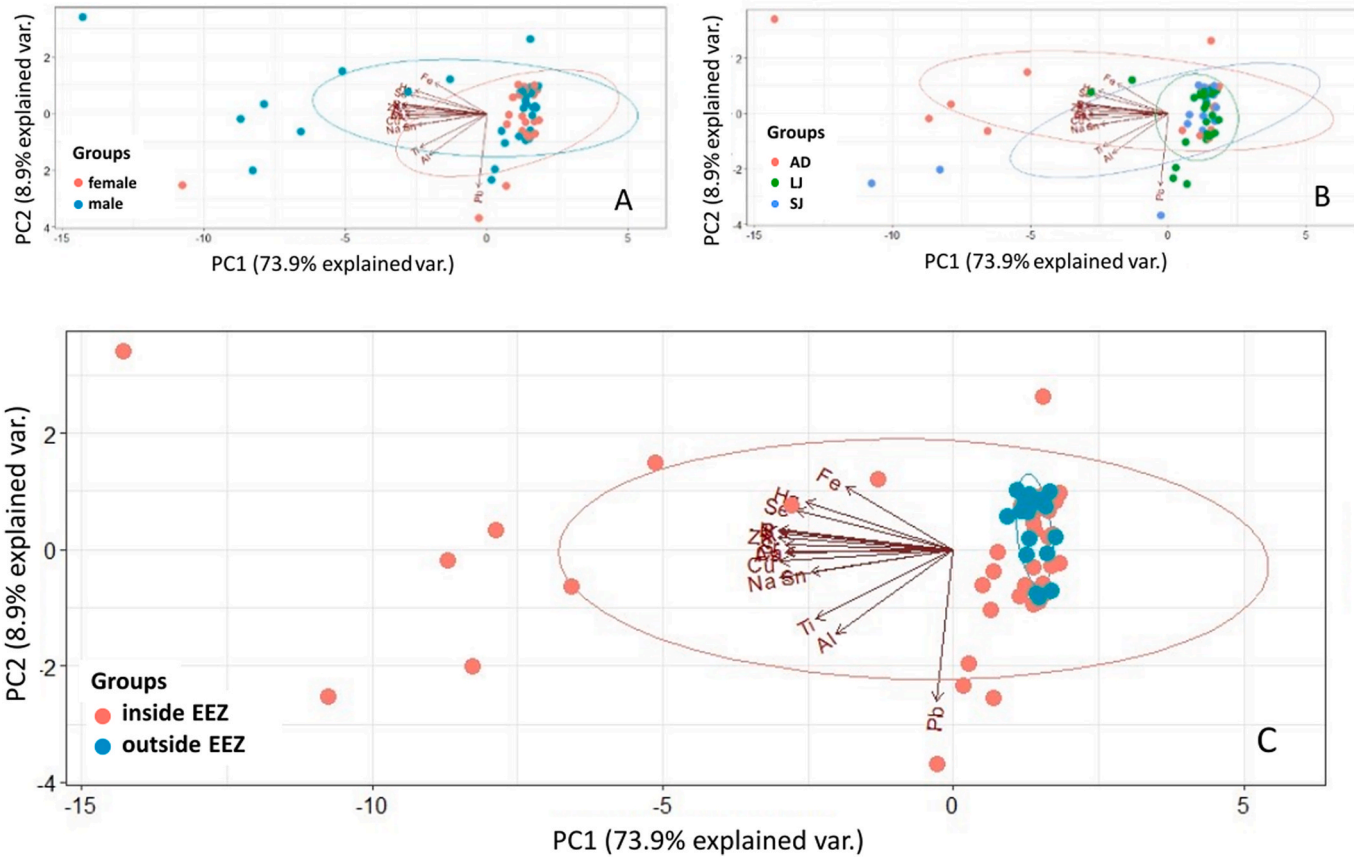

Fig. 5. Biplots with axis 1 and 2 from the same Principal Component Analysis (PCA) performed for concentrations of metals measured in muscle samples from blue sharks (*Prionace glauca*), highlighting: A) the different sexes – pink = female samples, blue = male samples; B) the different size/maturity groups – pink = adults (AD), green = large juveniles (LJ), blue = small juveniles (SJ); C) the different zones of sampling – pink = individuals sampled from inside EEZ (≤200 NM), blue = individuals sampled from outside EEZ (>200 NM). (For interpretation of the references to colour in this figure legend, the reader is referred to the Web version of this article.)

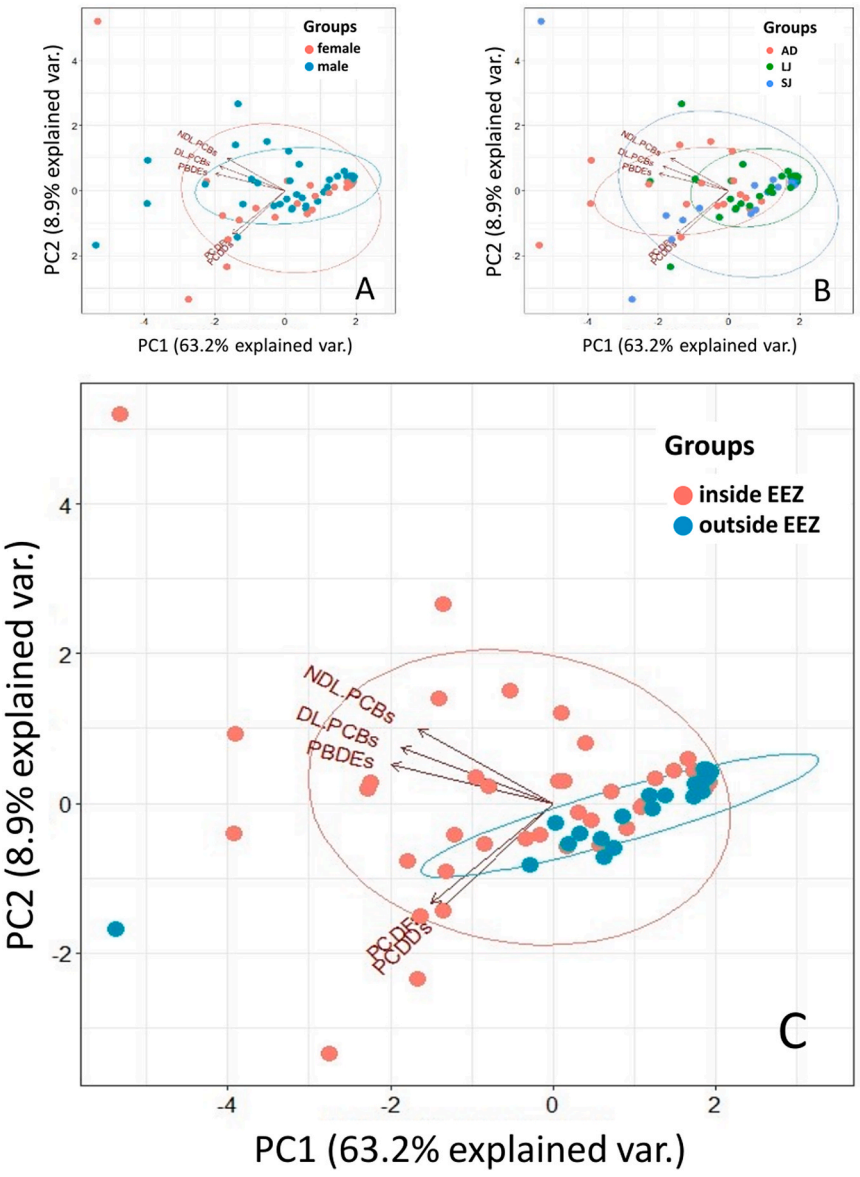

Fig. 6. Biplots with axis 1 and 2 from the same Principal Component Analysis (PCA) performed for concentrations of Persistent Organic Pollutants (POPs) measured in liver samples from blue sharks (*Prionace glauca*), highlighting: A) the different sexes – pink = female samples, blue = male samples; B) the different size/maturity groups – pink = adults (AD), green = large juveniles (LJ), blue = small juveniles (SJ); C) the different zones of sampling – pink = individuals sampled from inside EEZ (≤200 NM), blue = individuals sampled from outside EEZ (>200 NM). (For interpretation of the references to colour in this figure legend, the reader is referred to the Web version of this article.)

δ^{15}N: tau = 0.57, p = 9.6e-11). The correlations found between the isotopic values and the contaminants can be seen in Supplementary Table S5. Overall, most correlations were found between nitrogen ratios measured in both tissues and the concentrations of metals measured in muscle. All correlations found in both tissues were negative, and most were observed for nitrogen. No correlations were detected between isotopic ratios and concentrations of POPs. Finally, sex, size/maturity, and sampling zone groups showed no clear segregation of isotopic profiles (Supplementary Fig. S1), and Wilcoxon rank sum tests confirmed the lack of differences between groups of each category (all $p > 0.05$).

4. Discussion

4.1. Contamination profile and implications for human consumers

The overall preferential accumulation of metals in muscle is in accordance with what was observed in a recent study by Hauser-Davis et al. (2021), which assessed the concentrations of a large set of elements (22) in the same species, in the North-western Atlantic. In the present study, Cu, Fe, Mn, and Se did not follow this trend and instead accumulated preferentially in the liver. Higher concentrations of these essential elements in liver are not unusual for this species (Alves et al., 2016; Hauser-Davis et al., 2021; Stevens and Brown, 1974), and have been also observed for other shark species (Boldrocchi et al., 2019; Company et al., 2010; Gilbert et al., 2015). Elements such as Fe and Mn are known to be present in high concentrations in blood so their presence in a highly irrigated tissue such as the liver is expected. Additionally, some of these elements (e.g., Se) have important roles in detoxification processes naturally occurring in the liver (Schlenk et al., 2008). The mean concentrations of metals measured in muscle and liver tissues were compared with values available in the literature for other blue shark studies (Table 2). Overall, the mean concentrations found in the present work are within the ranges reported in the literature. However, the mean concentration of Hg measured in the present study is one of the highest reported. Furthermore, the mean concentration of Hg in the liver is the highest reported amongst all the studies, regardless of the geographic area of the sampling, although studies reporting values for concentrations measured in liver are much less common (6 for liver vs. 21 for muscle). Also, the study reporting the highest concentrations in shark muscle from the North Atlantic (Matos et al., 2015) did not report values for corresponding liver samples.

Table 2

Comparison of concentrations of metals found in muscle and liver samples from blue sharks (*Prionace glauca*). Metals were ordered by decreasing number of references; metals with the same number of references were ordered alphabetically. Data is presented as mean ± standard deviation, as mean, or range of values (minimum-maximum), as found on the literature. By default, concentrations are expressed in μg g^{-1} of dry weight (dw). When available, information on the size of the sharks was presented in cm as range of values (minimum-maximum), as mean ± standard deviation, or as mean, as found on the literature. To reduce the size of the numbers without losing information, values were rounded to one significant figure. FL = fork length; TL = total length. a = concentrations are expressed in μg g^{-1} of wet weight (ww); TXRF = Total Reflection X-ray Fluorescence spectroscopy; AAS = atomic absorption spectrometry; ICP-MS = Inductively coupled plasma mass spectrometry; FI-CVAFS = flow injection cold vapor atomic fluorescence spectrometry; CVAAS = Cold vapor atomic absorption spectrometry; AFS = Atomic fluorescence spectrometry; GC/ECD = gas chromatography/electron capture detection.

Muscle	Sampling area	Method	Size (cm)	N	Hg	Cu	Pb	Se	As	Zn	Fe	Mn	Ni	Cr	Al	Co	Sr	Ti	Sb	V	Ca	K	Na	P	S	Sn	Reference
	North Atlantic	TXRF	101.3-250.7 (FL)	60	1.9 ± 2.3	0.20 ± 2	0.7 ± 0.7	0.2 ± 0.3	8.9 ± 8.9	2.9 ± 3.4	2.5 ± 3.6	0.05 ± 0.08	0.02 ± 0.02	0.1 ± 0.2	30.1 ± 31.9	0.02 ± 0.003	0.6 ± 0.8	0.2 ± 0.2	0.1 ± 0.1	0.1 ± 0.02	18.8 ± 36.8	269.1 ± 312.5	2287.5 ± 2015.1	237.2 ± 366.9	169.8 ± 215.6	2.9 ± 3.7	Present study
		ICP-MS	209.3–288.3 (FL)	5	1.3 ± 0.5	0.6 ± 0.7	0.1 ± 0.03	3.1 ± 2.5	60.1 ± 34.04	7.8 ± 4.7	3.9 ± 2.7	0.1 ± 0.03	0.1 ± 0.1	0.5 ± 0.3	1.5 ± 0.6	0.005 ± 0.002	0.3 ± 0.2	19.7 ± 17.9	0.005 ± 0.002	0.03 ± 0.02							Hauser-Davis et al. (2021)
		AAS	79-284 (TL)	40	a 0.5																						Biton-Porsmoguer et al. (2018)
		AAS	145.32 ± 5.44 (TL)	30	0.3 ± 0.02	0.7 ± 0.2		0.5 ± 0.02	10.02 ± 0.7	3.9 ± 0.2				0.5 ± 0.03			0.4 ± 0.03										Torres et al. (2017)
		ICP-MS	112-167 (TL)	20	1.4 ± 0.8	1.2 ± 0.6	0.1 ± 0.1	0.3 ± 0.9	78.2 ± 21.9	24.6 ± 15.5	28.2 ± 26.2	0.6 ± 0.6	0.3 ± 0.6	2.6 ± 3.3	23.8 ± 47.01											Alves et al. (2016)
		ICP-MS		15	2.3 ± 0.7			0.3 ± 0.1																		Matos et al. (2015)
		AAS	84-239 (FL)	37	a 0.2–1.3			a 0.1–0.3																		Branco et al. (2007)
		AAS	97-258 (FL)	50	a 0.2–1.8																					Branco et al. (2004)
		AAS	203–219	5		0.2					6.3	1.6	2.6													Vas (1991)
		AAS	3.3–56.2 (kg)			4.4				35																Stevens and Brown (1974)
	South Atlantic	FI-CVAFS	77-137 (TL)	27	a 1.1 ± 0.6																					de Carvalho et al. (2014)
		AAS		47	0.8 ± 0.5																					Dias et al. (2008)
		CVAAS		30	a 0.4 ± 0.3																					Másrico et al. (2007)
	North Pacific	AAS	203 ± 22.2 (TL)	10	0.4 ± 0.2																					Kazama et al. (2020)
		AAS	117-269 (TL)	44	1.03 ± 0.1	1.6 ± 0.1		0.2 ± 0.02	6.7 ± 0.6	6.1 ± 0.4	27.4 ± 3.6															Barrera-García et al. (2012)
		AAS	206.2 ± 52.8 (TL)	21	1.9 ± 1.5																					Maz-Courrau et al. (2012)
		AAS	113-287 (TL)	38	1.4 ± 1.6			0.1 ± 0.05																		Escobar-Sánchez et al. (2011)
	Central Pacific	HR-ICP-MS	173 (TL)	30	0.4 ± 0.1	22.7 ± 15.9	2.9 ± 2.8	5.3 ± 1.7	114.9 ± 83.3	169.2 ± 81.8	445.3 ± 673.6	7.4 ± 11.3	2.1 ± 2.9	2.1 ± 4.8		0.3 ± 0.07											Álvaro-Berlanga et al. (2021)
	South Pacific	ICP-MS	122.6–200.6 (TL)	14			0.01 ± 0.01																			Cordero-Maldonado and Espinoza (2022)
		AFS	146.30–234.20 (TL)	25	a 0.1–0.5		a 0.04–0.3																			Reátegui-Quispe and Pariona-Velarde (2020)
		AAS		39	0.01 ± 0.1		2.2 ± 0.8																			Lopez et al. (2013)
		AAS	89-335 (TL)	74	0.6																					Davenport (1995)
	Sea of Japan ±	GC/ECD	110 ± 20	15	a 2.3 ± 1.1																					Kim et al. (2016)

(*continued on next page*)

Table 2 (*continued*)

	Mediterranean	AAS	80.5–212.0 (TL)	23					7.2 ± 3.1																Storelli and Marcotrigiano (2004)
	Adriatic Sea	AAS			0.4																				Storelli et al. (2001)
	South Indian	AAS	160-269 **(FL)**	31	5.3 ± 2.2																				Kiszka et al. (2015)
Liver	North Atlantic	**TXRF**	101.3-250.7 (FL)	60	1.6 ± 0.7	0.4 ± 0.3	0.6 ± 0.2	0.3 ± 0.2	2.6 ± 1.9	2.1 ± 1.3	5.9 ± 7.02	0.1 ± 0.1	0.02 ± 0.01		15.5 ± 15.9	0. ± 0.01	0.1 ± 0.1	0.2 ± 0.1			1.9 ± 2.1	53.8 ± 28.8	1281.5 ± 287.6	18.9 ± 12.3	37.8 ± 24.6	0.6 ± 0.7	Present study
		ICP-MS	209.3–288.3 (FL)	8	0.3 ± 0.2	1.1 ± 1.1	0.04 ± 0.01	1.3 ± 0.4	23.4 ± 13.1	6.5 ± 3.9	38.6 ± 33.6	0.5 ± 0.3		0.2 ± 0.1	0.2 ± 0.2	0.03 ± 0.02	0.2 ± 0.1	2.3 ± 0.9	0.001 ± 0.0003	0.02 ± 0.02						Hauser-Davis et al. (2021)
		ICP-MS	112-167 (TL)	20	0.3 ± 0.4	6.8 ± 3.9	1.3 ± 4.4		39.9 ± 27.8	43.9 ± 39.7	99.9 ± 55.8	2.5 ± 1.1	0.04 ± 0.2	1.6 ± 0.1	24.4 ± 40.1												Alves et al. (2016)
		AAS	84-239 (FL)	37	a 0.03–0.9			a 0.5–3.0																		Branco et al. (2007)
		AAS	203–219	5		0.7	1.1				4.02	0.4	3.2													Vas (1991)
		AAS	3.3–56.2 (kg)			5.7				39																Stevens and Brown (1974)
	Pacific	AAS	117-269 (TL)	35	0.2 ± 0.4	9.3 ± 8.4	0.4 ± 0.4	1.7 ± 0.6	10.6 ± 4.8	49.9 ± 27.1	195.7 ± 95.6															Barrera-García et al. (2013)
		AAS		39	0.1 ± 0.03		1.6 ± 0.3																			Lopez et al. (2013)
		AAS	203 ± 22.2 (TL)	10	0.1 ± 0.03																					Kazama et al. (2020)
	Mediterranean	AAS	80.5–212.0 (TL)	23					5.9 ± 2.7																	Storelli and Marcotrigiano (2004)

The high concentrations of Hg found in these sharks are concerning and may have been impacting their health, since elevated concentrations of this contaminant have been linked with the impairment of essential functions in many different fishes (Boening, 2000). Specifically, Hg has been linked with the induction of CYP1A expression in rectal gland cells from *Squalus acanthias* (Ke et al., 2002), and to a glutathione reduction in cerebrospinal fluid of *Rhizoprionodon terraenovae* (Ehnert-Russo and Gelsleichter, 2020). Additionally, all sampled sharks presented negative Se/Hg ratios in muscle (Supplementary Table S2), and only two opposed that result in liver (Supplementary Table S3). These results are similar to observations reported for blue shark meat tested in Spain by Olmedo et al. (2013), and negative Se/Hg ratios are also known in other shark species (Kaneko and Ralston, 2007; Olmedo et al., 2013). Since the Se/Hg ratio can be used to estimate the toxicity of the measured Hg, and considering the fact that high levels of Hg seem to normally exist in sharks (Tiktak et al., 2020), it is possible that these animals rely on mechanisms other than Se to counter Hg's effects. These negative Se/Hg ratios are also relevant to human consumers since they will face the risks of unsequestered Hg, having to rely on other sources of Se to counter the effects of this toxic element (Ralston and Raymond, 2010; Raymond and Ralston, 2020).

In addition to Hg, the muscle samples analysed in this work contained other elements known for their toxicity to humans, namely As and Pb (Järup, 2003). Despite being classified as Near Threatened by the IUCN Red List since 2009 (Rigby et al., 2019), blue shark is one of the most traded species in the global market for shark products (Dent and Clarke, 2015). Its trade is very common in some European countries (Henriques et al., 2021) and, even in those where it is not, its meat can still be found in many supermarkets and restaurants (in its original form or in derivatives). To safeguard consumers from the dangers of contamination, there are guidelines and limits for the concentration of elements such as As, Hg, and Pb in fish muscle deemed for human consumption. In the EU, the limits for these contaminants in food are set in the Commission Regulation (EC) No 1881/2006 (European Commission, 2006). The limit of As in fish meat for human consumption has been withdraw, meaning all concentrations detected in the present work are legal for human consumption in the EU. It is important to consider that this withdrawal happened not due to a lack of concern with As, but rather due to difficulties in its assessment in some types of food (such as fish meat), and to increasing evidence that exposure to concentrations lower than the previously set limit may elicit harm to consumers (Commission Regulation (EC) No 2015/1006). While there is currently no European legislation setting a limit for As in fish meat, there are some potential exportation destination countries that have those limits established (Petursdottir et al., 2015). The mean iAs value in this study (0.27 μg g^{-1} ww) is well below the limit imposed in Australia and New Zealand (i.e., 2 μg g^{-1} ww), but more than double the one existing in China (0.1 μg g^{-1} ww). In the EU, there are currently only As limits for fish destined for feed production. In fact, the anglers from the boat where the sampling took place admitted that a small portion of the blue sharks caught are occasionally destined for that use (personal observation). In that regard, present samples were mostly within the established safety limits for feed production, with only three animals surpassing the 25 μg g^{-1} ww corresponding limit, a much better result than the one obtained by Marques et al. (2021) using samples from another shark species (*Scyliorhinus canicula*), very commonly landed in Portugal. Nevertheless, present results showed that more than half of the sampled sharks would be in nonconformity if introduced in the supply chain and tested by food safety authorities for either Hg or Pb. While the legal limits are set for As, Hg, and Pb individually, safety assessments should consider all these and other contaminants together. For example, although the percentage of animals exceeding the legal limits for human consumption for Hg and Pb individually were 53% and 58%, respectively, the percentage of sharks that presented illegal concentrations for at least one of the two elements was 78%. It is important to keep in mind that concentrations of some of the contaminants included in our assessment (i.e., iAs, and MeHg) were estimated and that their real values can be slightly different. However, the fact that the value used to estimate MeHg in our samples (i.e., 90%, Branco et al., 2007; Kim et al., 2016; Storelli et al., 2001) is well supported in the literature, and the fact that the estimated concentrations are way above the safe limits for consumption indicate that slight differences in real concentration due to errors from the estimation would not change the overall conclusion that most of the sampled sharks could be considered unsafe for human consumption. These results raise concerns regarding the apparent lack of scrutiny that blue shark meat has been subjected to over the years. Using Hg as an example, it is possible to observe that from the available studies on Hg quantification in this shark species (Table 2), seven of them (including this study) report blue sharks specifically caught in the North Atlantic and all identified sharks with concentrations of Hg above the legally established limit for human consumption in the EU.

The concentration of contaminants such as As, Hg, and Pb in food is one of the factors influencing consumers' susceptibility to contaminant intake, with other factor being the frequency with which they eat the contaminated food. Using a worst case scenario approach, considering that the Portuguese are amongst the highest fish consumers in the world (FAO, 2020), some risk parameters fell expectedly way above the safety limits (Table 1). However, according to Almeida et al. (2015), despite the variety of fish and fishery products consumed, the Portuguese market trends are dominated by just a few species. Adjusting the value of consumed blue shark to a more realistic estimate (i.e., 0.021% of all fish consumed), both PTWI and HQ fell under the set safety limits. It is important to note however, that while an exclusive consumption of blue shark meat is highly unlikely to occur, certain communities living near shore and in close relationship with the fishing sector may eat blue shark meat much more frequently than the average Portuguese population. According to Statistics Portugal (INE), blue shark is almost entirely traded as frozen fish steaks (INE, 2021). In fact, this is the most common presentation in supermarkets, where portions (i.e., steaks) weight on average 0.26 Kg and thus, by eating just one steak of blue shark in a month may be enough to surpass the safe limit for this element (according to the MSWC of 0.07 kg/week), a conclusion already reported on previous publications on this topic (Bernardo, 2017; Matos et al., 2015). Regarding the concentrations of POPs measured in these sharks, a detailed discussion can be found in the work by Muñoz-Arnanz et al. (2022). Concerning the risk to human consumers, the analysis concluded that the consumption of these sharks' meat posed no serious threat to human consumers. However, neither the risk assessment in the present work neither the one performed by Muñoz-Arnanz et al. (2022) take into consideration the effects of simultaneous exposure to all the contaminants present in these sharks' meat. Caution should be taken when interpreting these data since the assessment of single contaminants may lead to an underestimation of the risk for the human health, as the simultaneous effects from several different contaminants may greatly increase the danger to consumers (Drakvik et al., 2020; Yáñez et al., 2002). Therefore, reduced intake of this kind of fishes should be promoted as part of a healthier diet.

4.2. Drivers of contaminant body burden

Due to the inherent characteristics of bioaccumulation and biomagnification, large and older sharks are expected to possess higher concentrations of chemical elements than their smaller and younger counterparts. Because of this, the size of these predators tends to be positively associated with the concentrations of hard to eliminate contaminants such as heavy metals and POPs (Tiktak et al., 2020), and there are reports of positive correlations between the size of blue sharks and the concentrations of heavy metals (Alves et al., 2016; Branco et al., 2007; Márcico et al., 2007), and POPs (Alves et al., 2016) in their tissues. Most POPs detected in the same individuals used for this study (Muñoz-Arnanz et al., 2022), were shown to positively correlate with the size of the sharks but such relationship was not observed for any of

the metals measured in the muscle samples. While unexpected, this result is not unheard and similar examples can be found in the literature (Boldrocchi et al., 2019; Dias et al., 2008). Several factors may have contributed to the present results. It is possible that unexpected high contamination levels in small sharks, and the opposite in large adults, can occur due to variations in feeding preferences, for example. As sharks grow they become able to eat larger prey, leading to positive associations between size and trophic level (i.e. δ^{15}N) (Kiszka et al., 2015), but our analysis showed no correlation between the size of the sharks and the isotopic ratios measured. Additionally, because larger prey often possess higher concentrations of contaminants, it was expected that the values of δ^{15}N would be positively correlated with the concentrations of contaminants in the sampled sharks, as was previously observed by other authors (Branco et al., 2004; Kiszka et al., 2015; Maz-Courrau et al., 2012), but no such correlations were found. In fact, only negative correlations between δ^{15}N and metals in the muscle which are generally non- toxic (i.e., P, K, Ti, Cu, Se, and Sr) were detected. Factors such as regional nutrient dynamics could potentially help justify the lack of correlations between isotopes and pollutants, but prey choice could also play a role in explaining our results. Larger blue sharks tend to eat more small bony fish and less crustaceans, when compared with smaller ones. This decrease in crustaceans, which can be rich in elements such as Cu (Taylor and Anstiss, 1999), and P and K (Gökoğlu and Yerlikaya, 2003), may be one of the causes for the negative correlations observed with these elements.

Consisting mostly of small fish, squid, and crabs, the feeding preferences of blue sharks may also vary according to sex, life stage, availability of prey, and individual preferences (Compagno et al., 2005). In sample sets with small to moderate sample numbers such as the one in the present work, the individual preferences of each shark should also be taken into consideration, as they can greatly impact the results. Differences in preferred feeding ground may expose sharks differently to contamination, masking expected positive associations between size and contamination (i.e., smaller sharks from more contaminated areas may have higher concentrations of contaminants in their tissues than larger sharks spending most of their time in more pristine locations). Since a great deal of the contamination derives from feeding, the variability in the resources exploited may affect not only the isotopic signature of the sharks, but their degree of exposure to contamination. No differences were found in both isotopes within any of the groups considered (i.e., sex, size/maturity, or zone of capture (Supplementary Fig. S1)). Unlike what was generally observed for the concentrations of contaminants, the isotopic profiles were very similar in both zones of sampling. This indicates that sharks from both zones had been feeding on prey with similar isotopic profiles (at least on average, during periods over one year). It also means that bioaccumulation may have a high effect in the concentration of contaminants over long time frames in the muscle of this species, leading to difficulties in detecting linear relationships with stable isotopes (i.e., trophic ecology). The high degree of consistency detected in the isotopic values between muscle and blood indicates high individual specialisation in both prey (i.e., δ15N) and habitat (i.e., δ13C) for periods longer than one year (Matich et al., 2011). Therefore, these animals may be subjected to chronic exposure to the contamination levels existing in their specific ecological niches, and this may help to explain the observed differences in contamination from the sharks captured in side and outside the EEZ. The spatial and temporal segregations of blue sharks are complex, as they vary both by influence of sex and size/maturity. In the Atlantic, females are known to give birth off the Iberian coast (Stevens, 1990) and in the Azores region (Queiroz et al., 2005), and pups tend to remain in their birth area for at least one or two years (Queiroz et al., 2005; Vandeperre et al., 2014b). Larger and older sharks tend to embark on large seasonal trips between mating seasons (Queiroz et al., 2005; Vandeperre et al., 2014a), but there is evidence pointing to a high degree of site fidelity, with the sharks often returning to their original pupping grounds (Vandeperre et al., 2014a). Based on the information from the aforementioned studies one can hypothesize that the blue sharks sampled in this work could have spent most of their lives in their corresponding zone of sampling (i.e., either inside or outside EEZ). It also suggests that blue sharks that happen to be born in areas more prone to contamination will be much more susceptible to prolonged exposure to contamination than blue sharks born in other more pristine areas. Therefore, it can be hypothesised that the differences observed here between the sharks sampled in each of the sampling zones may be due to the differences in the contamination levels of each zone, which seems to assume a higher relevance in explaining accumulation than sex or size. It should be highlighted that blue sharks from inside EEZ present generally higher concentrations of metals and POPs when compared with sharks from outside EEZ, and that the area inside EEZ is the closest to urbanized areas and human industries, a pattern that has been observed before for other elasmobranchs (Lyons et al., 2014).

5. Conclusions

The present study is an important step towards a better understanding of the contamination status of the blue shark populations in the Northeast Atlantic and of the risks it represents. Getting commercial fishing crews to cooperate in studies such as this one is not easy and is still the biggest obstacle for scientists wanting to use samples from freshly caught sharks. It is therefore essential to increase the efforts to establish good relations between academia and the fishing industry as the cooperation between the two is essential for the conservation of marine resources and the protection of human seafood consumers. Results show that commercially caught blue sharks in the Northeast Atlantic possess elevated concentrations of contaminants. Around 78% of sharks exceeded the legal limits for human consumption for either Hg or Pb. The muscle mean concentrations of Hg, in particular, are amongst the highest reported for the species. The present study's risk assessment indicates that although not widely and frequently consumed in the country, blue shark meat may be posing a serious threat to consumers if consumed regularly (i.e., one steak per month). However, risk assessments such as the one here presented still suffer from a degree of uncertainty that make objective recommendations hard to make, and for instance the future use of machine learning techniques could improve the accuracy and reliability of these assessments (Rajkumar et al., 2000; Ru et al., 2017). Regarding the factors potentially affecting the contamination detected in the sharks, individual spatial segregation seems to have more influence in driving contamination exposure than sex and size/maturity, with most of the differences found pointing to higher concentrations of metals and POPs in sharks caught in areas closer to the Portuguese mainland. The results also lead to the conclusion that future biomonitoring studies planning to use blue sharks to monitor pollution should make an effort to collect animals of similar size. Lastly, future studies should be conducted to assess if the sharks presenting higher levels of contamination were under a higher degree of stress when compared with the sharks from the more pristine area, and which may ultimately have an impact in these threated populations.

Author statement

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Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

Data will be made available on request.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.envpol.2022.120467.

References

Almeida, C., Karadzic, V., Vaz, S., 2015. The seafood market in Portugal: driving forces and consequences. Mar. Policy 61, 87–94. https://doi.org/10.1016/j.marpol.2015.07.012.

Álvaro-Berlanga, S., Calatayud-Pavía, C.E., Cruz-Ramírez, A., Soto-Jiménez, M.F., Liñán-Cabello, M.A., 2021. Trace elements in muscle tissue of three commercial shark species: Prionace glauca, Carcharhinus falciformis, and Alopias pelagicus off the Manzanillo, Colima coast, Mexico. Environ. Sci. Pollut. Res. 28, 22679–22692. https://doi.org/10.1007/s11356-020-12234-5.

Alves, L.M.F., Correia, J.P.S., Lemos, M.F.L., Novais, S.C., Cabral, H., 2020. Assessment of trends in the Portuguese elasmobranch commercial landings over three decades (1986–2017). Fish. Res. 230, 105648 https://doi.org/10.1016/j.fishres.2020.105648.

Alves, L.M.F., Lemos, M.F.L., Cabral, H., Novais, S.C., 2022. Elasmobranchs as bioindicators of pollution in the marine environment. Mar. Pollut. Bull. 176, 113418 https://doi.org/10.1016/j.marpolbul.2022.113418.

Alves, L.M.F., Nunes, M., Marchand, P., Le Bizec, B., Mendes, S., Correia, J.P.S., Lemos, M.F.L., Novais, S.C., 2016. Blue sharks (Prionace glauca) as bioindicators of pollution and health in the Atlantic Ocean: contamination levels and biochemical stress responses. Sci. Total Environ. 563 (564), 282–292. https://doi.org/10.1016/j.scitotenv.2016.04.085.

Barrera-García, A., O'Hara, T., Galván-Magaña, F., Méndez-Rodríguez, L.C., Castellini, J. M., Zenteno-Savín, T., 2013. Trace elements and oxidative stress indicators in the liver and kidney of the blue shark (Prionace glauca). Comp. Biochem. Physiol. Mol. Integr. Physiol. 165, 483–490. https://doi.org/10.1016/j.cbpa.2013.01.024.

Barrera-García, A., O'Hara, T., Galván-Magaña, F., Méndez-Rodríguez, L.C., Castellini, J. M., Zenteno-Savín, T., 2012. Oxidative stress indicators and trace elements in the blue shark (Prionace glauca) off the east coast of the Mexican Pacific Ocean. Comp. Biochem. Physiol. C Toxicol. Pharmacol. 156, 59–66. https://doi.org/10.1016/j.cbpc.2012.04.003.

Bernardo, I.A.S., 2017. Avaliação de riscos e benefícios da população portuguesa, crianças em idade pré-escolar e adultos, associados ao consumo de espécies de peixe alvo. Universidade de Lisboa.

Biton-Porsmoguer, S., Bănaru, D., Boudouresque, C.F., Dekeyser, I., Bouchoucha, M., Marco-Miralles, F., Lebreton, B., Guillou, G., Harmelin-Vivien, M., 2018. Mercury in blue shark (Prionace glauca) and shortfin mako (Isurus oxyrinchus) from north-eastern Atlantic: Implication for fishery management. Mar. Pollut. Bull. 127, 131–138. https://doi.org/10.1016/j.marpolbul.2017.12.006.

Boening, D.W., 2000. Ecological effects, transport, and fate of mercury: a general review. Chemosphere 40, 1335–1351. https://doi.org/10.1016/S0045-6535(99)00283-0.

Boldrocchi, G., Monticelli, D., Omar, Y.M., Bettinetti, R., 2019. Trace elements and POPs in two commercial shark species from Djibouti: implications for human exposure. Sci. Total Environ. 669, 637–648. https://doi.org/10.1016/j.scitotenv.2019.03.122.

Bosch, A.C., O'Neill, B., Sigge, G.O., Kerwath, S.E., Hoffman, L.C., 2016. Heavy metals in marine fish meat and consumer health: a review. J. Sci. Food Agric. 96, 32–48. https://doi.org/10.1002/jsfa.7360.

Branco, V., Canário, J., Vale, C., Raimundo, J., Reis, C., Canario, J., Vale, C., Raimundo, J., Reis, C., 2004. Total and organic mercury concentrations in muscle tissue of the blue shark (Prionace glauca L.1758) from the Northeast Atlantic. Mar. Pollut. Bull. 49, 871–874. https://doi.org/10.1016/j.marpolbul.2004.09.002.

Branco, V., Vale, C., Canário, J., Santos, M.N. dos, Branco, V., Vale, C., , Can??rio, Santos, J., dos, M.N., 2007. Mercury and selenium in blue shark (Prionace glauca, L. 1758) and swordfish (Xiphias gladius, L. 1758) from two areas of the Atlantic Ocean. Environ. Pollut. 150, 1–8. https://doi.org/10.1016/j.envpol.2007.01.040.

Burger, J., Gochfeld, M., 2013. Selenium and mercury molar ratios in commercial fish from New Jersey and Illinois: variation within species and relevance to risk communication. Food Chem. Toxicol. 57, 235–245. https://doi.org/10.1016/j.fct.2013.03.021.

Chouvelon, T., Warnau, M., Churlaud, C., Bustamante, P., 2009. Hg concentrations and related risk assessment in coral reef crustaceans, molluscs and fish from New Caledonia. Environ. Pollut. 157, 331–340. https://doi.org/10.1016/j.envpol.2008.06.027.

Compagno, L.J.V., Dando, M., Fowler, S., 2005. Sharks of the World, 2005th, Pape ed. Princeton University Press.

Company, R., Felícia, H., Serafim, A., Almeida, A.J., Biscoito, M., Bebianno, M.J., 2010. Metal concentrations and metallothionein-like protein levels in deep-sea fishes captured near hydrothermal vents in the Mid-Atlantic Ridge off Azores. Deep-Sea Res. Part I Oceanogr. Res. Pap. 57, 893–908. https://doi.org/10.1016/j.dsr.2010.02.005.

Consales, G., Marsili, L., 2021. Assessment of the conservation status of Chondrichthyans: underestimation of the pollution threat. Eur. Zool. J. 88, 165–180. https://doi.org/10.1080/24750263.2020.1858981.

Cordero-Maldonado, C., Espinoza, P., 2022. Cadmium and lead levels in muscle tissue of blue shark (Prionace glauca) in the Southeastern Pacific Waters. Mar. Pollut. Bull. 177, 113523 https://doi.org/10.1016/j.marpolbul.2022.113523.

da Silva, J.M., Alves, L.M.F., Laranjeiro, M.I., Bessa, F., Silva, A.V., Norte, A.C., Lemos, M.F.L., Ramos, J.A., Novais, S.C., Ceia, F.R., 2022. Accumulation of chemical elements and occurrence of microplastics in small pelagic fish from a neritic environment. Environ. Pollut. 292, 118451 https://doi.org/10.1016/j.envpol.2021.118451.

da Silva, J.M., Alves, L.M.F., Laranjeiro, M.I., Silva, A., Angélico, M.M., Norte, A.C., Lemos, M.F.L., Ramos, J.A., Novais, S.C., Ceia, F.R., 2020. Mercury levels in commercial mid-trophic level fishes along the Portuguese coast – relationships with trophic niche and oxidative damage. Ecol. Indicat. 116, 106500 https://doi.org/10.1016/j.ecolind.2020.106500.

Davenport, S., 1995. Mercury in blue sharks and deepwater dogfish from around Tasmania. Austr. Fish. 54 (3), 20–22.

de Carvalho, G.G.A., Degaspari, I.A.M., Branco, V., Canário, J., de Amorim, A.F., Kennedy, V.H., Ferreira, J.R., 2014. Assessment of total and organic mercury levels in blue sharks (Prionace glauca) from the south and Southeastern Brazilian coast. Biol. Trace Elem. Res. 159, 128–134. https://doi.org/10.1007/s12011-014-9995-6.

Denobile, M., 2007. Estudo da ocorrência de compostos arseniais, mercuriais e selênio em cações comercializados na cidade de São Paulo. Univ. São Paulo.

Dent, F., Clarke, S., 2015. State of the global market for shark products. FAO Fish. Aquac. Tech. Pap. No. 590, 187.

Dias, A.C.L., Guimarães, J.R.D., Malm, O., Costa, P.A.S., 2008. Mercúrio total em músculo de cação Prionace glauca (Linnaeus, 1758) e de espadarte Xiphias gladius Linnaeus, 1758, na costa sul-sudeste do Brasil e suas implicações para a saúde pública. Cad. Saúde Pública 24, 2063–2070. https://doi.org/10.1590/S0102-311X2008000900012.

Drakvik, E., Altenburger, R., Aoki, Y., Backhaus, T., Bahadori, T., Barouki, R., Brack, W., Cronin, M.T.D., Demeneix, B., Hougaard Bennekou, S., van Klaveren, J., Kneuer, C., Kolossa-Gehring, M., Lebret, E., Posthuma, L., Reiber, L., Rider, C., Rüegg, J., Testa, G., van der Burg, B., van der Voet, H., Warhurst, A.M., van de Water, B., Yamazaki, K., Öberg, M., Bergman, Å., 2020. Statement on advancing the assessment of chemical mixtures and their risks for human health and the environment. Environ. Int. 134, 105267 https://doi.org/10.1016/j.envint.2019.105267.

Duarte, B., Duarte, I.A., Caçador, I., Reis-Santos, P., Vasconcelos, R.P., Gameiro, C., Tanner, S.E., Fonseca, V.F., 2022. Elemental fingerprinting of thornback ray (Raja clavata) muscle tissue as a tracer for provenance and food safety assessment. Food Control 133, 108592. https://doi.org/10.1016/j.foodcont.2021.108592.

EC, 2006. COMMISSION REGULATION (EC) No 1881/2006 of 19 December 2006 setting maximum levels for certain contaminants in foodstuffs.

EC, 2015. Commission regulation (EU) 2015/1006. Off. Off. J. Eur. Union L 161/14, 14–16.

EFSA, 2009. Scientific opinion on arsenic in food. EFSA panel on contaminants in the food chain (CONTAM). EFSA J 7 (10), 199. https://doi.org/10.2903/j.efsa.2009.1351, 1351.

EFSA, 2010. Management of left-censored data in dietary exposure assessment of chemical substances. EFSA J. https://doi.org/10.2903/j.efsa.2010.1557.

EFSA, 2012. Guidance on selected default values to be used by the EFSA Scientific Committee, Scientific Panels and Units in the absence of actual measured data. EFSA J. 10, 1–32. https://doi.org/10.2903/j.efsa.2012.2579.

Ehnert-Russo, S.L., Gelsleichter, J., 2020. Mercury accumulation and effects in the brain of the Atlantic Sharpnose shark (Rhizoprionodon terraenovae). Arch. Environ. Contam. Toxicol. 78, 267–283. https://doi.org/10.1007/s00244-019-00691-0.

EPA, 2002. Chemical Assessment Summary: Arsenic (Inorganic), Iris.

EPA, 2011. An Overview of Methods for EPA's National-Scale Air Toxics Assessment. EPA, p. 220. https://www.epa.gov/sites/production/files/2015-10/documents/2005-nata-tmd.pdf.

Escobar-Sánchez, O., Galván-Magaña, F., Rosíles-Martínez, R., 2011. Biomagnification of mercury and selenium in blue shark Prionace glauca from the Pacific Ocean off

Mexico. Biol. Trace Elem. Res. 144, 550–559. https://doi.org/10.1007/s12011-011-9040-y.
EUMOFA, 2020. The EU Fish Market 2020. European Union. https://doi.org/10.2771/664425.
European Commission Directorate-General for Maritime Affairs and Fisheries, 2021. The EU Fish Market: 2020 Edition. Publications Office of the European Union, Luxembourg https://doi.org/doi/10.2771/7095.
European Council, 1993. Council Regulation (EEC) No. 315/93 of 8 February 1993 laying down Community procedures for contaminants in food. Orkesterjournalen L 37, 1–5.
FAO/WHO, 2016. Joint FAO/WHO Expert Committee on Food Additives JECFA Fact Sheet. World Health Organization - Technical Report Series.
FAO/WHO CAC, 2018. Working document for information and Use in discussions on the GSCTF CF/5. In: CODEX Alimentarius Commission. CODEX Alimentarius Commission, Utrecht, pp. 1–169.
European Parliament, 2009. Commission Directive 2009/141/EC of 23 November 2009 amending Annex I to Directive 2002/32/EC of the European Parliament and of the Council as regards maximum levels for arsenic, theobromine, Datura sp., Ricinus communis L., Croton tiglium L. and Abrus pr. Off. J. Eur. Union L 308, 20–23.
FAO, 2020. FAO Yearbook. Fishery and Aquaculture Statistics 2018/FAO annuaire. Statistiques des pèches et de l'aquaculture 2018/FAO anuario. Estadísticas de pesca y acuicultura 2018. FAO, Rome. https://doi.org/10.4060/cb1213t.
Farnham, I.M., Singh, A.K., Stetzenbach, K.J., Johannesson, K.H., 2002. Treatment of nondetects in multivariate analysis of groundwater geochemistry data. Chemom. Intell. Lab. Syst. 60, 265–281.
Ferretti, F., Worm, B., Britten, G.L., Heithaus, M.R., Lotze, H.K., 2010. Patterns and ecosystem consequences of shark declines in the ocean. Ecol. Lett. 13 https://doi.org/10.1111/j.1461-0248.2010.01489.x no-no.
Gilbert, J.M., Reichelt-Brushett, A.J., Butcher, P.a., McGrath, S.P., Peddemors, V.M., Bowling, A.C., Christidis, L., 2015. Metal and metalloid concentrations in the tissues of dusky Carcharhinus obscurus, sandbar C. plumbeus and white Carcharodon carcharias sharks from south-eastern Australian waters, and the implications for human consumption. Mar. Pollut. Bull. 92, 186–194. https://doi.org/10.1016/j.marpolbul.2014.12.037.
Gökoğlu, N., Yerlikaya, P., 2003. Determinaton of proximate composition and mineral contents of blue crab (Callinectes sapidus) and swim crab (Portunus pelagicus) caught off the Gulf of Antalya. Food Chem. 80, 495–498. https://doi.org/10.1016/S0308-8146(02)00318-7.
Gray, J.S., 2002. Biomagnification in marine systems: the perspective of an ecologist. Mar. Pollut. Bull. 45, 46–52. https://doi.org/10.1016/S0025-326X(01)00323-X.
Hauser-Davis, R.A., Chávez Rocha, R.C., Saint'Pierre, T.D., Adams, D.H., 2021. Metal concentrations and metallothionein metal detoxification in blue sharks, Prionace glauca L. from the western North Atlantic OCEAN. J. Trace Elem. Med. Biol. 126813 https://doi.org/10.1016/j.jtemb.2021.126813.
Helsel, P.J., 2021. NADA2: Data Analysis for Censored Environmental Data.
Henriques, A.C., Sá, R., Grilo, C., 2021. Sharks and Rays : Guardians of the ocean in crisis Why are the guardians of the ocean in crisis ?.
Hussey, N.E., MacNeil, M.A., Olin, J.A., McMeans, B.C., Kinney, M.J., Chapman, D.D., Fisk, A.T., 2012. Stable isotopes and elasmobranchs: tissue types, methods, applications and assumptions. J. Fish. Biol. 80, 1449–1484. https://doi.org/10.1111/j.1095-8649.2012.03251.x.
Ibrahim, A.T.A., Banaee, M., Sureda, A., 2019. Selenium protection against mercury toxicity on the male reproductive system of Clarias gariepinus. Comp. Biochem. Physiol., Part C: Toxicol. Pharmacol. 225, 108583 https://doi.org/10.1016/j.cbpc.2019.108583.
INE, 2021. Estatísticas do comércio internacional de bens [WWW Document]. INE, Inst. Nac. Estatística.
IRIS, 2022. Integrated Risk Information System online database, U.S. Environmental Protection Agency [WWW Document]. Off. Res. Dev. Natl. Cent. Environ. Assess.
Ishaque, A., Ishaque, S., Arif, A., Abbas, H., 2020. Toxic EFFECTS of lead ON FISH and human. Biol. Clin. Sci. Res. J. 2020, 1–7. https://doi.org/10.54112/bcsrj.v2020i1.47.
Jackson, A.L., Inger, R., Parnell, A.C., Bearhop, S., 2011. Comparing isotopic niche widths among and within communities: SIBER - stable Isotope Bayesian Ellipses in R. J. Anim. Ecol. 80, 595–602. https://doi.org/10.1111/j.1365-2656.2011.01806.x.
Järup, L., 2003. Hazards of heavy metal contamination. Br. Med. Bull. 68, 167–182. https://doi.org/10.1093/bmb/ldg032.
JECFA, 2007. Safety evaluation of certain food additives and contaminants. Food Additives, World Health Organization food additives series.
Kaneko, J.J., Ralston, N.V.C., 2007. Selenium and mercury in pelagic fish in the central north pacific near Hawaii. Biol. Trace Elem. Res. 119, 242–254. https://doi.org/10.1007/s12011-007-8004-8.
Kazama, H., Yamaguchi, Y., Harada, Y., Kaneko, N., Mizushima, H., Tsuchiya, K., Nemoto, M., Takaku, Y., Sahoo, Y.V., Tanaka, M., 2020. Mercury concentrations in the tissues of blue shark (Prionace glauca) from Sagami Bay and cephalopods from East China Sea. Environ. Pollut. 266, 115192 https://doi.org/10.1016/j.envpol.2020.115192.
Ke, Q., Yang, Y., Ratner, M., Zeind, J., Jiang, C., Forrest, J.N., Xiao, Y.-F., 2002. Intracellular accumulation of mercury enhances P450 CYP1A1 expression and Cl− currents in cultured shark rectal gland cells. Life Sci. 70, 2547–2566. https://doi.org/10.1016/S0024-3205(02)01502-3.
Kim, S.J., Lee, H.K., Badejo, A.C., Lee, W.C., Moon, H.B., 2016. Species-specific accumulation of methyl and total mercury in sharks from offshore and coastal waters of Korea. Mar. Pollut. Bull. 102 https://doi.org/10.1016/j.marpolbul.2015.11.038.
Kiszka, J.J., Aubail, A., Hussey, N.E., Heithaus, M.R., Caurant, F., Bustamante, P., 2015. Plasticity of trophic interactions among sharks from the oceanic South-Western Indian Ocean revealed by stable isotope and mercury analyses. Deep-Sea Res. Part I Oceanogr. Res. Pap. 96, 49–58. https://doi.org/10.1016/j.dsr.2014.11.006.
Kleinbaum, D.G., Kupper, L.L., 1978. Applied Regression Analysis and Other Multivariable Methods. Duxbury Press, Boston, Massachusetts, USA, p. 556.
Lee, L., 2020. NADA: Nondetects and Data Analysis for Environmental Data. CRAN.
Lopez, S.A., Abarca, N.L., Meléndez, R.C., 2013. Heavy metal concentrations of two highly migratory sharks (Prionace glauca and Isurus oxyrinchus) in the Southeastern pacific waters: Comments on public health and conservation. Trop. Conserv. Sci. 6, 126–137. https://doi.org/10.1177/194008291300600103.
Lyons, K., Lavado, R., Schlenk, D., Lowe, C.G., 2014. Bioaccumulation of organochlorine contaminants and ethoxyresorufin- o -deethylase activity in southern California round stingrays (Urobatis halleri) exposed to planar aromatic compounds. Environ. Toxicol. Chem. 33, 1380–1390. https://doi.org/10.1002/etc.2564.
Marques, A.F.S., Alves, L.M.F., Moutinho, A., Lemos, M.F.L., Novais, S.C., 2021. Scyliorhinus canicula (Linnaeus, 1758) metal accumulation: a public health concern for Atlantic fish consumers? Mar. Pollut. Bull. 169, 112477 https://doi.org/10.1016/j.marpolbul.2021.112477.
Másrico, E.T., Machado, M.E.S., Knoff, M., Clemente, S.C.S., Oswaldo, F., De Janeiro, C.-R., 2007. Communication Total mercury in sharks along the southern Brazilian Coast [Mercúrio total em tubarões capturados na costa brasileira]. Arq. Bras. Med. Vet. Zootec. 1593–1596.
Matich, P., Heithaus, M.R., Layman, C.A., 2011. Contrasting patterns of individual specialization and trophic coupling in two marine apex predators. J. Anim. Ecol. 80, 294–305. https://doi.org/10.1111/j.1365-2656.2010.01753.x.
Matos, J., Lourenço, H.M., Brito, P., Maulvault, A.L., Martins, L.L., Afonso, C., 2015. Influence of bioaccessibility of total mercury, methyl-mercury and selenium on the risk/benefit associated to the consumption of raw and cooked blue shark (Prionace glauca). Environ. Res. 143, 123–129. https://doi.org/10.1016/j.envres.2015.09.015.
Maz-Courrau, A., López-Vera, C., Galván-Magaña, F., Escobar-Sánchez, O., Rosíles-Martínez, R., Sanjuán-Muñoz, A., 2012. Bioaccumulation and biomagnification of total mercury in four exploited shark species in the baja California peninsula, Mexico. Bull. Environ. Contam. Toxicol. 88, 129–134. https://doi.org/10.1007/s00128-011-0499-1.
Muñoz-Arnanz, J., Bartalini, A., Alves, L., Lemos, M.F., Novais, S.C., Jiménez, B., 2022. Occurrence and distribution of persistent organic pollutants in the liver and muscle of Atlantic blue sharks: relevance and health risks. Environ. Pollut. 309, 119750 https://doi.org/10.1016/j.envpol.2022.119750.
Muñoz, O., Devesa, V., Suñer, M.A., Vélez, D., Montoro, R., Urieta, I., Macho, M.L., Jalón, M., 2000. Total and inorganic arsenic in fresh and processed fish products. J. Agric. Food Chem. 48, 4369–4376. https://doi.org/10.1021/jf000282m.
National Research Council of the National Academies, 2014. Review of EPA's Integrated Risk Information System (IRIS) Process Committee to Review the IRIS Process, first ed. National Academies Press, Washington D. C.
Neff, J.M., 1997. Ecotoxicology of arsenic in the marine environment. Environ. Toxicol. Chem. 16, 917–927. https://doi.org/10.1002/etc.5620160511.
Olmedo, P., Pla, A., Hernández, A.F., Barbier, F., Ayouni, L., Gil, F., 2013. Determination of toxic elements (mercury, cadmium, lead, tin and arsenic) in fish and shellfish samples. Risk assessment for the consumers. Environ. Int. 59, 63–72. https://doi.org/10.1016/j.envint.2013.05.005.
Pacoureau, N., Rigby, C.L., Kyne, P.M., Sherley, R.B., Winker, H., Carlson, J.K., Fordham, S.V., Barreto, R., Fernando, D., Francis, M.P., Jabado, R.W., Herman, K.B., Liu, K.-M., Marshall, A.D., Pollom, R.A., Romanov, E.V., Simpfendorfer, C.A., Yin, J. S., Kindsvater, H.K., Dulvy, N.K., 2021. Half a century of global decline in oceanic sharks and rays. Nature 589, 567–571. https://doi.org/10.1038/s41586-020-03173-9.
Petursdottir, A.H., Sloth, J.J., Feldmann, J., 2015. Introduction of regulations for arsenic in feed and food with emphasis on inorganic arsenic, and implications for analytical chemistry. Anal. Bioanal. Chem. 407, 8385–8396. https://doi.org/10.1007/s00216-015-9019-1.
Post, D.M., Layman, C.A., Arrington, D.A., Takimoto, G., Quattrochi, J., Montaña, C.G., 2007. Getting to the fat of the matter: models, methods and assumptions for dealing with lipids in stable isotope analyses. Oecologia 152, 179–189. https://doi.org/10.1007/s00442-006-0630-x.
Queiroz, N., Lima, F.P., Maia, A., Ribeiro, P.A., Correia, J.P., M. Santos, A., 2005. Movement of blue shark, Prionace glauca, in the north-east Atlantic based on mark-recapture data. J. Mar. Biol. Assoc. U. K. 85, 1107–1112. https://doi.org/10.1017/S0025315405012154.
R Core Team, 2020. R: A Language and Environment for Statistical Computing.
Rajkumar, T., Guesgen, H.W., Robinson, S., Fisher, G.W., 2000. A New dose model for assessment of health risk due to contaminants in Air. J. Air Waste Manage. Assoc. 50, 3–20. https://doi.org/10.1080/10473289.2000.10463993.
Ralston, N.V.C., Raymond, L.J., 2010. Dietary selenium's protective effects against methylmercury toxicity. Toxicology 278, 112–123. https://doi.org/10.1016/j.tox.2010.06.004.
Raymond, L.J., Ralston, N.V.C., 2020. Mercury: selenium interactions and health implications. Neurotoxicology 81, 294–299. https://doi.org/10.1016/j.neuro.2020.09.020.
Reátegui-Quispe, A., Pariona-Velarde, D., 2020. Determinación de plomo, cadmio, mercurio y Bases Volátiles Nitrogenadas Totales (NBVT) en el músculo de tiburón azul Prionace glauca procedente de la zona sur del Perú. Rev. Biol. Mar. Oceanogr. 54, 336. https://doi.org/10.22370/rbmo.2019.54.3.2029.
Rigby, C.L., Barreto, R., Carlson, J., Fernando, D., Fordham, S., Francis, M.P., Herman, K., Jabado, R.W., Liu, K.M., Marshall, A., Pacoureau, N., Romanov, E., Sherley, R.B., Winker, H., 2019. Prionace glauca, Blue Shark, the IUCN Red List of Threatened Species 2019.

Ru, G., Crescio, M.I., Ingravalle, F., Maurella, C., Gregori, D., Lanera, C., Azzolina, D., Lorenzoni, G., Soriani, N., Zec, S., Berchialla, P., Mercadante, S., Zobec, F., Ghidina, M., Baldas, S., Bonifacio, B., Kinkopf, A., Kozina, D., Nicolandi, L., Rosat, L., 2017. Machine Learning Techniques applied in risk assessment related to food safety. In: EFSA Support, vol. 14. Publ. https://doi.org/10.2903/sp.efsa.2017.EN-1254.

Schlenk, D., Celander, M., Gallagher, E.P., George, S., James, M., Kullman, S.W., Van Den Hurk, P., Willett, K., 2008. The Toxicology of Fishes, the Toxicology of Fishes. CRC Press. https://doi.org/10.1201/9780203647295.

Stevens, J.D., 1990. Further results from a tagging study of pelagic sharks in the north-east Atlantic. J. Mar. Biol. Assoc. U. K. 70, 707–720. https://doi.org/10.1017/S0025315400058999.

Stevens, J.D., Brown, B.E., 1974. Occurrence of heavy metals in the blue shark Prionace glauca and selected pelagic in the N. E. Atlantic Ocean. Mar. Biol. 26, 287–293. https://doi.org/10.1007/BF00391512.

Storelli, M., Marcotrigiano, G., 2004. Interspecific variation in total arsenic body concentrations in elasmobranch fish from the Mediterranean Sea. Mar. Pollut. Bull. 48, 1145–1149. https://doi.org/10.1016/j.marpolbul.2004.03.005.

Storelli, M.M., Stuffler, R.G., Marcotrigiano, G.O., 2001. Total mercury and methylmercury in tuna fish and sharks from the South Adriatic Sea. Ital. J. Food Sci. 13, 101–106.

Suedel, B.C., Boraczek, J.A., Peddicord, R.K., Clifford, P.A., Dillon, T.M., 1994. Trophic transfer and biomagnification potential of contaminants in aquatic ecosystems. In: Reviews of Environmental Contamination and Toxicology, pp. 21–89. https://doi.org/10.1007/978-1-4612-2656-7_2.

Taylor, H.H., Anstiss, J.M., 1999. Copper and haemocyanin dynamics in aquatic invertebrates. Mar. Freshw. Res. 48, 159–172. https://doi.org/10.1071/MF99117.

Team, Rs, 2020. RStudio. Integrated Development Environment for R.

Tiktak, G.P., Butcher, D., Lawrence, P.J., Norrey, J., Bradley, L., Shaw, K., Preziosi, R., Megson, D., 2020. Are concentrations of pollutants in sharks, rays and skates (Elasmobranchii) a cause for concern? A systematic review. Mar. Pollut. Bull. 160, 111701 https://doi.org/10.1016/j.marpolbul.2020.111701.

Torres, P., da Cunha, R., Rodrigues, A.D.S., 2017. Mid-Atlantic elasmobranchs: suitable metal scouts? Mar. Pollut. Bull. 117, 203–213. https://doi.org/10.1016/j.marpolbul.2017.01.058.

Vandeperre, F., Aires-da-Silva, A., Fontes, J., Santos, M., Serrão Santos, R., Afonso, P., 2014a. Movements of blue sharks (Prionace glauca) across their life history. PLoS One 9, e103538. https://doi.org/10.1371/journal.pone.0103538.

Vandeperre, F., Aires-da-Silva, A., Santos, M., Ferreira, R., Bolten, A.B., Serrao Santos, R., Afonso, P., 2014b. Demography and ecology of blue shark (Prionace glauca) in the central North Atlantic. Fish. Res. 153, 89–102. https://doi.org/10.1016/j.fishres.2014.01.006.

Vas, P., 1991. Trace metal levels in sharks from British and Atlantic waters. Mar. Pollut. Bull. 22, 67–72. https://doi.org/10.1016/0025-326X(91)90138-I.

Wenning, R.J., Martello, L., 2014. POPs in marine and freshwater environments. In: Environmental Forensics for Persistent Organic Pollutants. Elsevier, pp. 357–390. https://doi.org/10.1016/B978-0-444-59424-2.00008-6.

Wilhelmsson, D., Thompson, R.C., Holmström, K., Lindén, O., Eriksson-Hägg, H., 2013. Marine pollution. In: Sumaila, U.R., Diaz, R. (Eds.), Managing Ocean Environments in a Changing Climate. Elsevier, Boston, pp. 127–169. https://doi.org/10.1016/B978-0-12-407668-6.00006-9.

Yáñez, L., Ortiz, D., Calderón, J., Batres, L., Carrizales, L., Mejía, J., Martínez, L., García-Nieto, E., Díaz-Barriga, F., 2002. Overview of human health and chemical mixtures: problems facing developing countries. Environ. Health Perspect. 110, 901–909. https://doi.org/10.1289/ehp.110-1241270.