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RESEARCH ARTICLE

# Mercury screening in highly consumed sharpnose sharks (*Rhizoprionodon lalandii* and *R. porosus*) caught artisanally in southeastern Brazil

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Sharpnose sharks *Rhizoprionodon lalandii* and *R. porosus* are frequently captured in fishing activities in Brazil and are significantly consumed by humans, especially in southeastern Brazil. Both species lack population data and suffer intense fishing pressures and habitat degradation, consequently hindering adequate management and conservation actions. In this context, this study aimed to assess mercury (Hg) contamination in *R. lalandii*, and *R. porosus* sampled off the coast of Rio de Janeiro, addressing both animal health and public health risks. Sharks were obtained from two artisanal fishing colonies in southeastern Brazil (Copacabana and Recreio dos Bandeirantes), located on the coastal zone adjacent to Guanabara Bay, one of the most important, productive, and contaminated estuaries in Brazil, and a further three artisanal fishing colonies from the Região dos Lagos area (Saquarema, Cabo Frio and Rio das Ostras). Hg concentrations in liver, muscle, and brain in *R. lalandii* ($n = 24$) and *R. porosus* ($n = 20$) specimens were determined by inductively coupled plasma mass spectrometry. A gravid female measuring 112 cm from Copacabana is the first record for an individual of this size for *R. lalandii*. No correlation between length and muscle Hg concentrations was observed, and no differences between Hg concentrations for muscle or liver were found between male and female juveniles from either Cabo Frio or Rio das Ostras. No differences in Hg loads were observed herein for both assessed species. Low Hg bioaccumulation in juveniles and nongravid female muscle tissue was noted compared to significantly higher Hg concentrations in gravid females. Hg was detected in all embryos, indicating potential maternal offloading. As Hg thresholds for sharks in particular have not yet been established, whether the Hg concentrations detected in brain pose neurotoxic risks for these animals is not known. Public health concerns concerning adult *R. lalandii* consumption from Copacabana, however, are significant.

**Keywords:** Hg contamination, Public health assessment, Elasmobranchii

## 1. Introduction

Mercury (Hg) is a highly toxic and persistent element, which is biomagnified throughout trophic webs (Lavoie et al., 2013). Even sublethal concentrations of this contaminant in animal tissues pose significant risks to animal health, especially during the embryonic and

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juvenile phases, potentially leading to significant neurotoxicity and motor, sensory, and reproductive damage (McKinney et al., 2016).

Coastal elasmobranchs, particularly vulnerable to a contaminant like mercury due to their long life spans, slow growth, late sexual maturity, and low fecundity (Camhi et al., 1998), occupy key niches in trophic webs and perform top or intermediate functions (Heupel et al., 2014). Increasingly, however, they have been overfished worldwide, either as bycatch or targeted for human consumption. Thus, the public health implications regarding elasmobranch Hg contamination need to be considered, especially in Brazil, one of the world's largest consumers of elasmobranch meat and other by-products (Bornatowski et al., 2018).

Sharpnose sharks *Rhizoprionodon lalandii* (Brazilian sharpnose shark) and *R. porosus* (Caribbean sharpnose shark), two small coastal sharks, are distributed throughout the entire Brazilian coast and occupy intermediate positions in neritic systems (Compagno, 2001). Both are frequently captured in fishing activities in Brazil (Lessa et al., 1999; Compagno, 2001; Andrade et al., 2008). *R. lalandii* is distributed from Nicaragua to southern Brazil and is very prominent in Brazil's fishing scenario, accounting for 60% of all elasmobranch captures in the south and southeastern Brazil (Bornatowski et al., 2012). It is classified as "Data Deficient" by the International Union for Conservation of Nature (Rosa et al., 2004; IUCN, 2020). In contrast, a wider northern distribution range is observed for *R. porosus*, from Panama to southern Brazil, with a current classification of "Least Concern" by the IUCN (Lessa et al., 2006; MMA, 2018). Sharpnose shark captures in Rio de Janeiro, SE Brazil, are significant, with an estimated total of 5,212.8 kg landed in this category in 2016 (FIPERJ, 2016), by both artisanal fisheries and industrial fleets. Artisanal fisheries in Brazil are defined as small-scale fisheries involving fishing households, as opposed to commercial companies, and are usually organized in fishing colonies or associations with a local leader as representative; they carry out low-technology fisheries activities, using multigear and presenting high species overlap, mainly near the coast (Silva Junior et al., 2008; Prestrelo et al., 2019).

Lack of population data, fishing pressures, and habitat degradation pose major threats for both *R. lalandii* and *R. porosus*, hindering adequate management and conservation actions (Rosa et al., 2004; Lessa et al., 2006). Coastal areas usually display high levels of contamination due to anthropogenic activities (Fleming and Laws, 2006). However, assessments concerning Hg in surface waters and the water column in coastal water bodies in southeastern Brazil and off the Rio de Janeiro coast, in particular, are sorely lacking, as most evaluations are carried out in sediment. Sediment studies have reported higher Hg concentrations in water bodies located in the metropolitan region of the city of Rio de Janeiro, while lower levels have been reported further to the north of the state of Rio de Janeiro, indicating significant anthropogenic influence (Lacerda and Gonçalves, 2001).

In this context, coastal organisms are considered adequate bioindicators concerning environmental contamination, and assessments in this regard have become increasingly essential in understanding the dynamics of contaminants, such as metals, in aquatic ecosystems (Alves et al., 2016). Studies on sharks in general, however, are scarce compared to bony fish. Some assessments concerning Hg risks associated with shark consumption in Brazil have been carried out (Dias et al., 2008; De Carvalho et al., 2014; Hauser-Davis et al., 2020), reporting significant public health concerns, as Hg concentrations are mostly above the maximum permissible limits set for this toxic element. This situation becomes even more alarming when taking into account that shark meat intake has increased significantly in Brazil in recent years and is even served in public schools throughout the country, to children ranging in age from 6 to 17 years, as subsidized meat by the government (Bornatowski et al., 2018).

Considering the fact that sharpnose sharks are highly consumed by local human populations in Rio de Janeiro, this study aimed to assess muscle Hg contamination in *R. lalandii*, and *R. porosus* sampled off the coast of Rio de Janeiro, addressing both animal health and public health risks. Brain and liver Hg concentrations were also assessed to further discussions on animal health.

## 2. Methodology

### 2.1. Study area

The Copacabana and Recreio dos Bandeirantes artisanal fishing colonies, located in the coastal zone adjacent to Guanabara Bay (GB), southeastern Brazil, were chosen due to their high representativity regarding artisanal fisheries in this area. GB is one of the most important and productive estuaries in Brazil (da Silva et al., 2016), albeit highly contaminated by untreated sewage from both domestic and industrial sources, as well as by other anthropogenic sources (Fistarol et al., 2015). A further three artisanal fishing locations were selected from the Região dos Lagos area, namely Saquarema, Cabo Frio, and Rio das Ostras, located relatively near the oil and gas exploration area of Campos Basin, one of the highest productivity areas in Brazil. **Figure 1** presents all sampling locations included in the present study.

### 2.2. Sharpnose shark sampling and processing

*R. lalandii* ($n = 22$) and *R. porosus* ($n = 20$) specimens were purchased whole from artisanal fishers between February 2017 and September 2019, through visits at intervals ranging from 2 weeks to 1 month, determined by adequate oceanographic conditions for fishery activities. Because of these constraints, no systematic samplings were carried out.

All specimens were caught by gillnets, with meshes ranging between 40 mm and 120 mm (Copacabana, 50 mm; Cabo Frio, 45 and 65 mm; Recreio, 40 mm; and Rio das Ostras, 110 to 120 mm), positioned at mid-water depth or at the bottom. These mesh sizes are used throughout the sampling area and allow for comparisons with no fish mesh-size bias (Silva Junior et al., 2008; da Oliveira et al., 2016; Viana et al., 2017; Loto et al., 2018).

Fish were all purchased fresh, about 1 h after landing, at the landing sites themselves, which is where the fishers

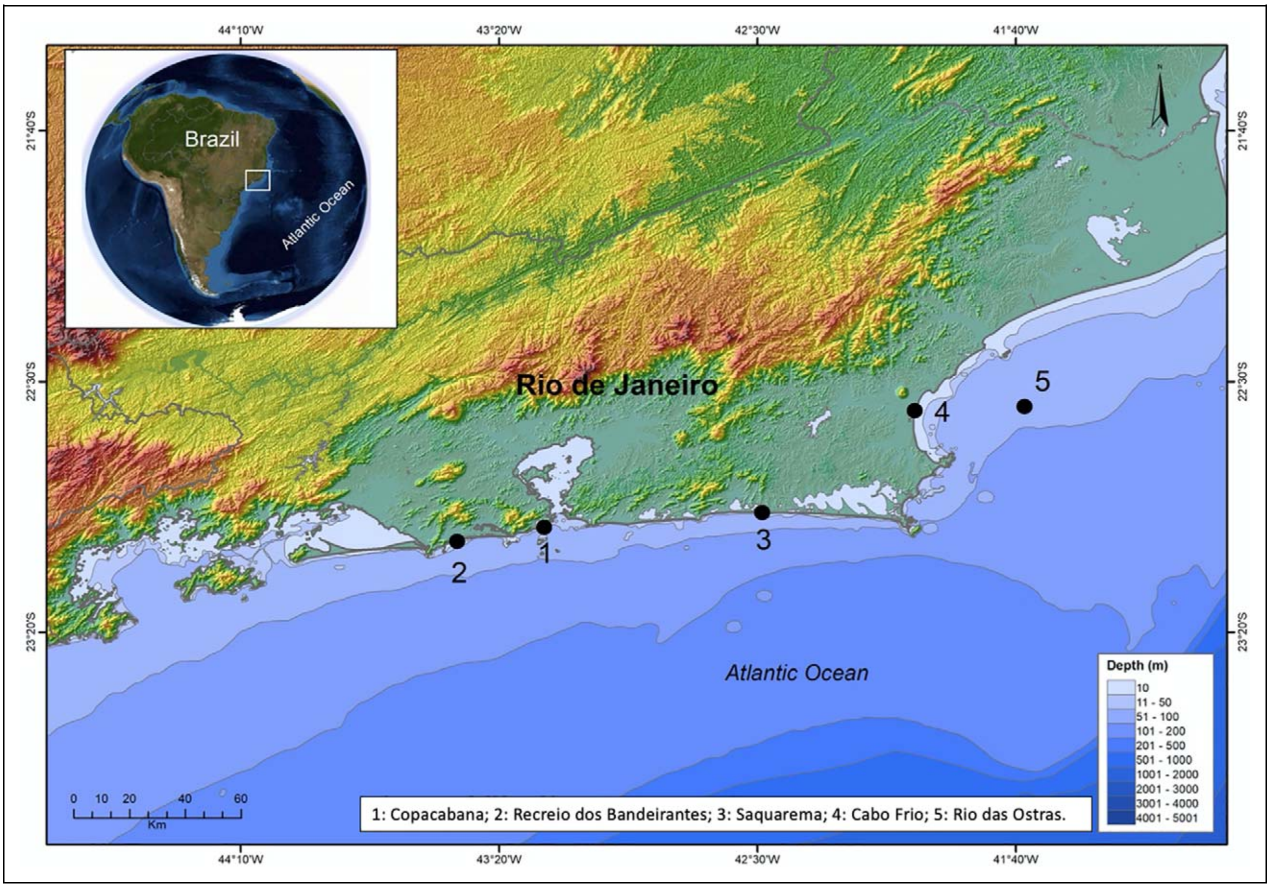


**Figure 1.** Map indicating the sharpnose shark sampling sites in Rio de Janeiro, southeastern Brazil. DOI: https://doi.org/10.1525/elementa.022.f1

**Table 1.** ICP-MS instrumental operating conditions applied for the determination of mercury in sharpnose sharks. DOI: https://doi.org/10.1525/elementa.022.t1

| ICP-MS Parameter | Condition |
|---|---|
| RF power | 1,100 W |
| Plasma gas flow rate | 17.0 L $min^{-1}$ |
| Auxiliary gas flow rate | 1.2 L $min^{-1}$ |
| Carrier gas flow rate | 0.98 L $min^{-1}$ |
| Sampling and skimmer cones | Pt |
| Dwell time | 30 ms per isotope |

sell their catches. This selling location is due to the fact that artisanal fisheries in Brazil are small-scale, display low autonomy, and do not carry refrigerated containers or have ice holds, which leads to port-adjacent fisheries activities by the fleets (Silva Junior et al., 2008; Prestrelo et al., 2019). The fish were then immediately put on ice and taken to the laboratory, with small variations in time spans and under the same storage conditions, thus not affecting the results.

Individuals were identified at the species level and grouped by size classes (embryos, juveniles, or adults), according to Andrade et al. (2008), sexed and measured, with the total length determined using a measuring tape to the nearest centimeter. Subsequently, all specimens were dissected, and the organs of interest (muscle, liver, and brain) were removed and stored at –20 °C in sterile 15-mL polypropylene screw-capped tubes until analysis.

### 2.3. Hg determinations

For Hg determinations, approximately 100 mg of each sample was weighed in sterile 15-ml polypropylene screw-capped tubes and acid-digested with 1.0 ml of bidistilled $HNO_3$ (Merck, Rio de Janeiro) overnight. The following day, the samples were heated at 100 °C for approximately 4 h in the closed tubes, avoiding loss of volatile elements (USP, 2013). After cooling, the samples were made up with ultrapure water (resistivity > 18.0 MΩ cm) obtained from a Merck Millipore water purifying system (Darmstadt, Germany) and analyzed by inductively coupled plasma mass spectrometry on a NexIon 300X ICP-MS equipment (PerkinElmer, Norwalk, CT, USA). Hg concentrations were determined through external multielemental calibration using $^{102}$Rh as the internal standard. Analytical curve correlation coefficients were above 0.995. The instrumental ICP-MS operating conditions are given in **Table 1**.

Method accuracy was verified by the parallel analysis of a certified reference material (CRM) ERM®-BB422 (fish muscle, European Commission) in triplicate. Certified reference material recovery values were considered adequate,

**Table 2.** Total length (cm) for *Rhizoprionodon lalandii* and *R. porosus* specimens from southeastern Rio de Janeiro, Brazil. DOI: https://doi.org/10.1525/elementa.022.t2

| | *Rhizoprionodon lalandii* | | | | | | | *R. porosus* | |
|---|---|---|---|---|---|---|---|---|---|
| | Juveniles | | Adults | | | Embryos | | Juveniles | |
| | Rio das Ostras | | Copacabana | Saquarema | Recreio | Recreio | | Cabo Frio | |
| Variables | Males ($n$ = 2) | Females ($n$ = 5) | Gravid females ($n$ = 6) | Nongravid females ($n$ = 3) | Gravid female ($n$ = 1) | Embryos ($n$ = 3) | | Males ($n$ = 12) | Females ($n$ = 10) |
| Mean ± *SD* | 40.3 ± 2.69 | 41.2 ± 3.32 | 77.0 ± 17.8 | 60.0 ± 2.00 | na[a] | 25.4 ± 2.51 | | 49.3 ± 3.07 | 47.6 ± 2.32 |
| Minimum | 38.4 | 38.2 | 64.5 | 58.0 | na | 23.0 | | 44.0 | 43.0 |
| Maximum | 42.2 | 46.1 | 112 | 62.0 | na | 28.0 | | 56.0 | 52.0 |

This gravid female was sampled with her head and tail cut off.

[a]Not available.



at 113% (Certified value: 0.601 ± 0.030, determined value 0.680 ± 0.032), as per Eurachem standards (Eurachem, 1998; Ishak et al., 2015).

### *2.4. Statistical analyses and calculation of dietary Hg intake*

Data normality was first assessed by the Shapiro–Wilk test. As the data were non-normally distributed, a Kruskal–Wallis test and the Mann–Whitney test were applied to assess differences in Hg concentrations between age groups and tissues. Spearman's correlation test was applied to investigate potential correlations between Hg concentrations among tissues, applying the correlation strength standards proposed by Bryman and Cramer (2011), where 0.00 < $r$ < 0.19 is classified as very weak; 0.20 < $r$ < 0.39 as weak; 0.40 < $r$ < 0.69 as moderate; 0.70 < $r$ < 0.89 as strong; and 0.90 < $r$ < 1.0 as very strong. Data analyses were carried out using the Prism® V. 8 software. Statistical significance was accepted when $P < 0.05$.

To estimate dietary mercury intake for *Rhizoprionodon lalandii* and *R. porosus* muscle, the mean value from each sampling site was multiplied by the average weekly fish consumption in the metropolitan Rio de Janeiro area (355 g) and divided by the mean body weight of a Brazilian adult (estimated at 70 kg).

## 3. Results and discussion

### *3.1. Biometric sharpnose shark data*

The total lengths for all sharpnose sharks sampled in the present study at different sampling sites throughout the state of Rio de Janeiro are provided in **Table 2**. This morphometric parameter is applied to determine fish maturity classes (juvenile, adult) through literature comparisons (Frisk et al., 2001; Mas et al., 2014), as fish age is typically positively correlated to fish length (Trudel and Rasmussen, 1997; Van Walleghem et al., 2007). Due to slow Hg elimination rates, Hg accumulation is observed with increasing fish age, as fish consume more food through time (Castro et al., 2002; Chumchal and Hambright, 2009; Piraino and Taylor, 2009). Consequently, the effects of fish length on Hg contamination is one of the most important relationships to assess due to the central role that this biometric parameter plays in both understanding contaminant dynamics and in determining natural resource policies (Sackett et al., 2013).

Twenty-two *R. porosus* individuals were sampled only at Cabo Frio, while all other sampled individuals were *R. lalandii* specimens, including six gravid females from Copacabana, one gravid female containing three embryos from Recreio dos Bandeirantes, seven juveniles from Rio das Ostras, and three nongravid females from Saquarema.

Sharpnose shark life cycles are restricted characteristically to coastal environments. The higher frequency of *R. lalandii* juveniles observed herein at the end of spring and in summer has been reported previously for southeastern Brazil (Motta et al., 2007; Andrade et al., 2008; Viana et al., 2017), while adults have been reported as being more common from April to July (calendar-wise defined by sampling dates, as autumn/spring) with neonates beginning to appear in June and dominating in August and September (spring/summer), the birth season for this species. A high frequency for *R. porosus* juveniles was also observed herein in summer, seemingly following the same trend in the reproductive cycle as *R. lalandii*, as observed in Guaratiba, located near Recreio (Ferreira, 1988). Mature specimens come nearer to the coast for parturition and copula in winter in southeastern Brazil, corroborated herein by the presence of five gravid females in Copacabana during this season, although the sixth specimen, the largest individual (112 cm), was captured in spring and the Recreio specimen in autumn.

Significant differences ($P$ < 0.05) in total length assessed by the Kruskal–Wallis test were observed only between adult gravid and nongravid females from Copacabana and Saquarema, respectively (only one gravid female was sampled from Recreio, preventing statistical comparisons), with Copacabana specimens being larger. A gravid female measuring 112 cm in total length from Copacabana is the first record for an individual of this size, while all other sampled specimens were within the maximum documented size for this species, of about 80 cm (Motta et al., 2007; Andrade et al., 2008; Gomes et al., 2010).

**Table 3.** Muscle, liver, and brain Hg concentrations (mg kg$^{-1}$ w.w.) expressed as mean ± standard deviation (with the range of values) for *Rhizoprionodon lalandii* and *R. porosus* specimens from southeastern Rio de Janeiro, Brazil. DOI: https://doi.org/10.1525/elementa.022.t3

| | *Rhizoprionodon lalandii* | | | | | | *R. porosus* | |
|---|---|---|---|---|---|---|---|---|
| | Juveniles | | Adults | | | Embryos | Juveniles | |
| | Rio das Ostras | | Copacabana | Saquarema | Recreio | Recreio | Cabo Frio | |
| Organ | Males (*n* = 2) | Females (*n* = 5) | Gravid females (*n* = 6) | Non-gravid females (*n* = 3) | Gravid female (*n* = 1) | Embryos (*n* = 3) | Males (*n* = 12) | Females (*n* = 10) |
| Muscle | 0.12 ± 0.040 | 0.15 ± 0.070 | 5.0 ± 5.9 | 0.21 ± 0.040 | 1.3 (1.3) | 0.22 ± 0.030 | 0.17 ± 0.10 | 0.18 ± 0.080 |
| | (0.090–0.14) | (0.090–0.26) | (0.44–16) | (0.17–0.24) | | (0.20–0.25) | (0.080–0.38) | (0.070–0.31) |
| Liver | 0.030 ± 0.010 | 0.040 ± 0.010 | 0.47 ± 0.48 | 0.07 ± 0.00 | 3.5 (3.5) | 0.040 ± 0.010 | 0.090 ± 0.010 | 0.15 ± 0.12 |
| | (0.020–0.030) | (0.030–0.060) | (0.070–1.1) | (0.070–0.070) | | (0.040–0.050) | (0.090–0.10) | (0.050–0.28) |
| Brain | 0.02 ± 0.4 | 0.025 ± 0.013 | 1.65 ± 3.05 | 0.054 ± 0.017 | 0.22 (0.22) | 0.046 ± 0.0066 | ns[a] (ns) | ns (ns) |
| | (0.017 ± 0.025) | (0.010–0.040) | (0.030–6.2) | (0.039–0.072) | | (0.038–0.050) | | |

[a]Not sampled.

### 3.2. Muscle, liver, and brain sharpnose shark Hg concentrations

Hg concentrations for *R. lalandii* and *R. porosus* muscle, liver, and brain samples from Cabo Frio, Rio das Ostras, Copacabana, Saquarema, and Recreio are provided in **Table 3**. Data are expressed as mean ± standard deviation, with the range of values, in units of mg kg$^{-1}$ wet weight (w.w.). The data that support the findings of this study are available as supplemental material.

Many studies indicate an exponential relationship between length (and, consequently, age) and muscle Hg concentrations, suggesting rapid Hg uptake rates due to the low metabolic rate and long life span of these animals (Lacerda et al., 2000; García-Hernandez et al., 2007; Rumbold et al., 2014; Ehnert-Russo and Gelsleichter, 2020). No correlation between these parameters was observed herein, which may be due to the fact that most individuals were juveniles, limiting the range of assessed body length, as reported in other studies for silvertip sharks (*Carcharhinus albimarginatus*) from Japan (Endo et al., 2009), or due to the relatively low sample number, as reported for the Smooth Hammerhead Shark, *Sphyrna zygaena*, from the Mexican Pacific Ocean (Escobar-Sánchez et al., 2010). Therefore, this lack of correlation should not be taken as evidence of effective detoxification (Escobar-Sánchez et al., 2010).

Concerning sex, no differences between Hg concentrations for both muscle and liver were observed between male and female juveniles from either Cabo Frio or Rio das Ostras, corroborating literature assessments of Hg concentrations for other *Rhizoprionodon* members (*R. acutus*) from South Africa (McKinney et al., 2016) and the Persian Gulf (Adel et al., 2017), as well as other sharks, such as the blue shark *Prionace glauca* and shortfin mako *Isurus oxyrinchus*, sampled from the NE Atlantic (Biton-Porsmoguer et al., 2018), and nine shark species sampled from the Florida Coast (Hueter et al., 1995). Thus, juvenile males and females were assessed together per location in further statistical analyses.

Mercury is widely known for its bioaccumulation processes along trophic chains (Lavoie et al., 2013). No differences in Hg loads were observed herein for the two assessed species. Although juvenile specimens from Cabo Frio (*R. porosus*) were significantly larger than juveniles from Rio das Ostras (*R. lalandii*), no significant differences were observed for muscle Hg concentrations. Liver concentrations, on the other hand, were significantly higher in *R. porosus* specimens. This finding contrasts with other assessments for this species carried out in Rio de Janeiro, where differences in muscle Hg concentrations were observed, with *R. lalandii* presenting higher mean values than *R. porosus* (Lacerda et al., 2000). *R. lalandii*, however, displays ontogenic variations in feeding habits (Bornatowski et al., 2012), which can directly affect contamination levels among different age groups.

With regard to adult gravid and nongravid females, significantly higher concentrations in muscle tissue, but not liver, were observed in the former. This finding may be due to the gravid status per se, as gravid sharks increase their feeding rates due to increased energy costs of carrying their young (Olsen, 1953; Madigan et al., 2015; Townsend and Gilchrist, 2017). Increased feeding rate increases their lipid reserves, leading to higher mercury accumulation, as this element is highly lipophilic (Clarkson and Magos, 2006). Alternatively, the higher Hg concentrations in the muscle may be due to the larger size of females from Copacabana and the fact that this area is Hg-contaminated. The 112-cm gravid female from Copacabana, in fact, presented the highest Hg concentrations in muscle tissue of all specimens assessed in this study, at 16.40 mg kg$^{-1}$ and 6.22 mg kg$^{-1}$ in the brain (no liver data are available), probably due to both age and, consequently, increased environmental exposure time, as well as higher Hg concentrations observed in GB compared to the Campos Basin, where the Cabo Frio and Rio das Ostras sampling sites are located further north (Lacerda and Gonçalves, 2001).

In this regard, Copacabana and Recreio are located on the coastal zone adjacent to the highly eutrophized and environmentally contaminated GB. Hg contamination in this bay originates from several diffuse sources, including river contamination plumes, atmospheric deposition, landfill runoff, and a chlor-alkali plant on the northwestern side of the bay (Covelli et al., 2012). Oceanic currents and a local upwelling phenomenon in this area, in turn, lead to significant Hg exports to neighboring coastal areas (da Carreira et al., 2012; de Melo et al., 2015; Martins et al., 2016). Thus, Hg contamination at both Copacabana and Recreio may be expected and have been reported previously for several marine biota fish. For example, methylmercury (MeHg), the most toxic form of Hg, has been reported as effectively biomagnifying from the prey, consisting of microplankton, mesoplankton, and fish with different feeding habits, to several pelagic–demersal top predators at GB (Kehrig et al., 2011). On the other hand, MeHg concentrations in this bay may be lower than expected due to high organic matter content, leading to Hg complexing, dilution of mercury inputs, and decreased residence time in the water column, with consequent decreased bioavailability for the local trophic web (Kehrig et al., 2002).

Data, however, are scarce concerning sharks in general and the *Rhizoprionodon* genus in particular. In fact, although both species are regularly consumed by humans in Rio de Janeiro (Andrade et al., 2008), no systematic assessments regarding their potential as bioindicators for environmental Hg exposure, or, indeed, other contaminants, are available. In this regard, Viana et al. (2005) investigated *R. lalandii* and *R. porosus* from Recreio, reporting Hg concentrations ranging from 0.086 to 0.726 mg kg$^{-1}$ w.w. in muscle tissue, with a mean of 0.199 $\pm$ 0.157 mg kg$^{-1}$ w.w. ($n = 20$), although no further data on sex or size were given. In another study, within GB, Lacerda et al. (2000) sampled both *R. lalandii* and *R. porosus*, reporting mean values of 0.018 mg kg$^{-1}$ w.w. ($n = 45$) and 0.0094 mg kg$^{-1}$ w.w. ($n = 12$) in muscle tissue, respectively. No standard deviation or sex information, however, is given. The other assessed areas are less urbanized than Recreio and Copacabana, probably resulting in lower contamination levels (Marins et al., 2004). The results reported herein for muscle in other *Rhizoprionodon* genus members are in the same range as those reported, for example, for *R. acutus* from Djibouti, Africa (0.07 mg kg$^{-1}$ w.w.; Boldrocchi et al., 2019) and off the coast of Korea (0.16 and 0.21 mg kg$^{-1}$ w.w.; Kim et al., 2016), *R. taylori* from Northern Australia (0.03 to 1.2 mg kg$^{-1}$ w.w.; Lylel, 1986), *R. longurio* from the Gulf of California (0.20 to 0.58 mg kg$^{-1}$ w.w.; Frías-Espericueta et al., 2015), and *R. terraenovae* from the Gulf of Mexico (0.63 mg kg$^{-1}$ w.w.; Núñez Nogeira et al., 1998), east-central Florida, USA (0.17 to 0.29 mg kg$^{-1}$ w.w.; Adams and McMichael, 1999), and South Carolina, USA (0.54 mg kg$^{-1}$ w.w.; Smith and Guentzel, 2010). Higher individual Hg concentrations in muscle, however, have been noted for *R. acutus* sampled in northern Australia (maximum of 2.0 mg kg$^{-1}$ w.w.; Lylel, 1986), the Arabian Sea (0.76 mg kg$^{-1}$ w.w.; Al-Raesi et al., 2007), the East Coast of Africa (2.5 mg kg$^{-1}$ w.w.; McKinney et al., 2016), and the Persian Gulf (1.2 mg kg$^{-1}$ w.w.; Adel et al., 2017), for *R. longurio* from Mexico (3.36 mg kg$^{-1}$ w.w.; Hurtado-Banda et al., 2012), and for *R. terraenovae* from east-central Florida (2.3 mg kg$^{-1}$ w.w.; Adams and McMichael, 1999) and Lee County, also in Florida (2.0 mg kg$^{-1}$ w.w.; Rumbold et al., 2014), and the southeastern U.S. coast (3.1 mg kg$^{-1}$ w.w.; Ehnert-Russo and Gelsleichter, 2020). None, however, are as high as the gravid female from Copacabana (16 mg kg$^{-1}$ w.w.), while three of the other five gravid females also presented higher Hg muscle concentrations compared to most of the available literature (3.9, 4.2, and 4.7 mg kg$^{-1}$ w.w.).

Liver assessments are not as abundant as for muscle samples but report similar levels for *R. acutus* from Djibouti, Africa (0.17 mg kg$^{-1}$ w.w.; Boldrocchi et al., 2019), *R. longurio* from Mexico (0.001–0.22 mg kg$^{-1}$ w.w.; Hurtado-Banda et al., 2012), and *R. terraenovae* from the Gulf of Mexico (0.16 mg kg$^{-1}$ w.w.; Núñez Nogeira et al., 1998) compared to the *R. lalandii* and *R. porosus* assessed herein from Rio de Janeiro.

Concerning the brain, significant differences for brain Hg concentrations were observed between male and female juveniles from Rio das Ostras and between embryos, juveniles, and adults. The gravid female from Recreio (0.217 mg kg$^{-1}$w.w.) and two of the gravid females from Copacabana (6.2 and 0.32 mg kg$^{-1}$ w.w., respectively) presented higher Hg levels than the other specimens, including adults from Saquarema, which ranged from 0.010 to 0.072 mg kg$^{-1}$ w.w. Hg is considered a potent neurotoxin that can impair cognitive functioning and individual functions, such as swimming and foraging, as well as ecosystem functions (Nam et al., 2014; Rumbold et al., 2014). Field assessments and laboratory exposure studies, concerning both freshwater and marine fish, indicate that the brain is the primary organ affected by Hg exposure and reflects environmental contaminant concentrations, altering both the central nervous system morphology, decreasing the amount of cerebellum, hypothalamus, and optic tectum nerve cells, and modifying predator-avoidance behavior, memory, and swimming patterns that may lead to populational and ecosystemic alterations (Webber and Haines, 2003; Pereira et al., 2014).

The brain Hg concentrations observed in the present study were higher than in *R. terraenovae* samples from the Gulf of Mexico and the Southeastern U.S. Coast, which ranged from 0.04 to 0.61 mg kg$^{-1}$ in males and from 0.00 to 1.7 mg kg$^{-1}$ in females ($n = 11$ in both cases; Núñez Nogeira et al., 1998). The authors, however, indicated that they freeze-dried their samples for Hg determinations but did not state whether their results are given in w.w. or dry weight, hindering direct comparisons to the levels observed herein. In another study on the same species, brain Hg concentrations ranged from 0.005 to 1.1 mg kg$^{-1}$ w.w. (mean of 0.23 $\pm$ 0.22 mg kg$^{-1}$ w.w, $n = 163$) in adults and 0.009 to 0.055 mg kg$^{-1}$ w.w. (0.024 $\pm$ 0.012 mg kg$^{-1}$ w.w., $n = 28$) in juveniles (Ehnert-Russo and Gelsleichter, 2020), within the same range as both *R. lalandii* and *R. porosus* observed herein, excepting the gravid females. As reported by Ehnert-Russo and Gelsleichter (2020), and even though mean brain concentrations reported herein were generally low, with the exception of gravid females, some individuals exceeded threshold values set for certain neurological responses, such as neurochemistry and neurobehavior alterations (0.7 to 1.2 mg $kg^{-1}$ w.w.; Dietz et al., 2013). These threshold values, however, were set for arctic mammals like seals, polar bears, and beluga whales. Altered nervous system enzymatic activities have been reported at Hg concentrations as low as 0.008 mg $kg^{-1}$ w.w., although in muscle tissue, while vital behavioral changes in freshwater fish, such as swimming, foraging, and reproduction, have been described at about 0.135 mg $kg^{-1}$ w.w. also in muscle (Sandheinrich and Wiener, 2011). For marine fish, especially for sharks, thresholds concerning Hg neurotoxicity are, thus, lacking.

Interestingly, Ehnert-Russo and Gelsleichter (2020), when assessing Hg accumulation and distribution in Atlantic sharpnose shark (*R. terraenovae*) specimens collected along the southeastern U.S. coast throughout most of its U.S. geographical range, reported that MeHg concentrations in the brain were low compared to InorgHg concentrations and that higher Hg levels were observed in the forebrain compared to mid- and hindbrain, indicating possible selective effects if brain Hg accumulation were to increase. As some studies have indicated that placental viviparous species may be more susceptible to Hg neurotoxicity, as they tend to have larger brains (Mull et al., 2011), an interesting field of research concerning brain Hg uptake in sharks from species that exhibit different brain morphologies is set (Ehnert-Russo and Gelsleichter, 2020).

### *3.3. Preliminary maternal transfer implications*

Maternal Hg off-loading and reproductive and embryonic damage risks have been reported as inducing morphological malformations and reducing offspring survival rates in several fish species (Jezierska et al., 2009), and several shark assessments have been carried out for different species (Adams and McMichael, 1999; Lyons and Lowe, 2013; Frías-Espericueta et al., 2015; van Hees and Ebert, 2017). For example, a hypothesis was raised, concerning three *R. lalandii* embryos presenting kyphosis and lordosis deformations in southeastern Brazil, that these malformation events could be associated directly with the presence of environmental contaminants, including Hg (Dos Santos and Gadig, 2014).

This type of contaminant transfer has been confirmed for *Rhizoprionodon* species in other assessments. For example, Frías-Espiricueta et al. (2015) detected Hg in *R. longurio* maternal umbilical cord, blood, and placenta and in embryo livers. These findings are due to the fact that this genus displays placental viviparity (Gomes et al., 2010), and embryos develop without contact with the external environment. Therefore, the only possible source of Hg contamination is from mother contaminant loads (Gomes et al., 2010; Lyons and Lowe, 2013). The maternal transfer process has been postulated as a detoxification strategy for female placental viviparous sharks (Adams and McMichael, 1999). Placental viviparous sharks, on the other hand, present low Hg transfer to their embryos (Le Bourg et al., 2014). Although only three embryos were assessed herein, and no statistical correlations between mother and embryo could be carried out for Hg concentrations to confirm maternal offloading due to this low sample number, Hg was detected in all brain, liver, and muscle embryo samples at levels comparable to Rio das Ostras juveniles and Copacabana pregnant adults, reinforcing the vulnerability of intrauterine phases to environmental Hg contamination. Muscle tissue concentrations were the highest of the three tissues, while liver and brain concentrations were similar. Contaminant concentrations, however, seem to influence the maternal transfer process in sharks (van Hees and Ebert, 2017) and should also be taken into account. Thus, additional assessments should be carried out with other gravid female–embryo pairs to further maternal offloading knowledge in this species.

### *3.4. Public health concerns*

Hg is listed by the World Health Organization (WHO, 2017) as one of the top 10 contaminants of major public health concern worldwide. Due to its high toxicity and the ability to bioaccumulate and cross the placental barrier in mammals (Chen et al., 2014), maximum permissible levels for Hg-contaminated food items have been implemented in order to limit Hg intake. Maximum permissible levels for this contaminant in fish for human consumption, as set by several regulating agencies, as well as their specific fish product, are provided in **Table 4**.

The Hg concentrations observed herein in muscle tissue are lower than all maximum permissible limits for both predator fish and sharks and within the acceptable range set by the FSA (UK) for all juveniles from all sampling locations. Four of the six gravid females from Copacabana and the one gravid female from Recreio, on the other hand, presented high Hg concentrations due to age and, consequently, environmental exposure time to this contaminant. Nongravid females, however, present lower and similar contamination levels as juveniles and embryos.

Fish ingestion rates vary according to country, region, and even states (Dias et al., 2008). To accurately assess public health concerns, the most representative value (if available) concerning fish intake must be considered. In this regard, the average daily fish intake in Brazil is 24.7 g (173 g $week^{-1}$), while daily intake in the metropolitan Rio de Janeiro area has been estimated as double the country's average, or 50.7 g (355 g $week^{-1}$; BRASIL, 2009). Taking these estimated values into account, along with an estimated mean body weight of 70 kg for a Brazilian adult, dietary mercury intakes for *R. lalandii* and *R. porosus* muscle were calculated for comparison to the provisional tolerable weekly intake (PTWI) for inorganic mercury (InorgHg) and MeHg of 4.0 μg $kg^{-1}$ body weight (bw) $week^{-1}$ and 1.6 μg $kg^{-1}$ bw $week^{-1}$, respectively (FAO/WHO, 2010), as established by the Food and Agriculture Organization of the United Nations (FAO) and the Joint FAO/WHO Expert Committee on Food Additives (JECFA). Because the JECFA recommends that dietary exposure to mercury from fish and shellfish be compared only to the PTWI for MeHg (FAO/WHO, 2010), MeHg contents in fish were calculated by considering that 90% of total mercury in fish is usually found in methylated form (WHO, 1990; Noisel et al., 2011). The resulting PTWI values are provided in **Table 5**.

**Table 4.** Maximum permissible levels for Hg in fish food items worldwide. DOI: https://doi.org/10.1525/elementa.022.t4

| Regulating Agency[a] | Maximum Permissible Limit (mg $kg^{-1}$ w.w.) | Specificity | References |
|---|---|---|---|
| ANVISA | 1.0 | Predator fish | ANVISA (2013) |
| FAO | 1.0 | Shark | FAO/WHO (2017) |
| FDA | 1.0 | Predator fish | FDA (2014) |
| UKDH | 0.022–0.30 | Shark | Woods (1999) |
| FSANZ | 1.5 | Predator fish | Food Standards Australia New Zealand (2016) |
| CHINA | 1.0 | Predator fish | Clever and Jie (2014) |
| EU | 1.0 | Predator fish | European Commission (2008) |

FAO = Food and Agriculture Organization of the United Nations; FDA = U.S. Food and Drug Administration; UKDH = UK Department of Health; FSANZ = Food Standards Australia New Zealand; USDA = U.S. Department of Agriculture; EU = Commission of European Communities.

[a]ANVISA indicates the Brazilian National Health Surveillance Agency.



**Table 5.** Dietary provisional tolerable weekly intake (PTWI) (μg $kg^{-1}$ bw) for inorganic mercury (InorgHg) and methylmercury (MeHg) for *Rhizoprionodon lalandii* and *R. porosus* sampled from Rio de Janeiro, Brazil. DOI: https://doi.org/10.1525/elementa.022.t5

| Sampling Site | Hg Species | Estimated Level of Exposure (μg $kg^{-1}$ bw $week^{-1}$) |
|---|---|---|
| Saquarema | InorgHg | 0.52 |
| | MeHg | 0.46 |
| Recreio | InorgHg | 3.29 |
| | MeHg | 2.96 |
| Rio das Ostras | InorgHg | 0.34 |
| | MeHg | 0.31 |
| Copacabana | InorgHg | 25.4 |
| | MeHg | 22.9 |
| Cabo Frio | InorgHg | 0.42 |
| | MeHg | 0.38 |

The mean values for InorgHg and MeHg from the six gravid females from Copacabana exceeded InorgHg (4.0 μg $kg^{-1}$ bw $week^{-1}$) and MeHg (1.6 μg $kg^{-1}$ bw $week^{-1}$) PTWI, as did the MeHg value for the only gravid female from Recreio, further corroborating the public health concerns mentioned earlier for these locations, especially as these animals are routinely consumed in high volumes in the metropolitan region of Rio de Janeiro.

## 4. Conclusions

The Hg results for muscle, liver, and brain in *Rhizoprionodon lalandii* and *R. porosus* from the coast of Rio de Janeiro indicate low Hg bioaccumulation for juveniles and nongravid females, while gravid females presented higher Hg concentrations. Hg thresholds for marine fish and sharks in particular, however, have not yet been set, and studies in this regard are still scarce for these animals. Thus, neurotoxicity and other biochemical and behavioral alterations may still be of concern. The largest gravid female at Copacabana presented the highest Hg concentrations, followed by gravid females from Recreio, indicating more significant Hg bioaccumulation with age, although the large number of variables and the relatively small number of fish specimens do not allow comparative assessments. Significant public health concerns regarding *R. lalandii* consumption from Copacabana, for both InorgHg and MeHg, and from Recreio for MeHg are, thus, evident, noting that the assessed specimens at these sampling areas were all adults. Mitigation actions in this regard include the treatment of sewage discharges directly into local water bodies and awareness campaigns among artisanal fishers concerning high Hg concentrations in sharks and other top predators.

## Data Accessibility Statement

The data that support the findings of this study are available as supplemental material.

## Supplemental files

The supplemental files for this article can be found as follows:

- Data_S1.xlsx

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The authors declare no conflicts of interest.

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- Data curation, visualization, investigation, formal analysis, writing—original draft: CAL, IQW, NLFA
- Validation, visualization, investigation, formal analysis: LHSSP
- Validation, visualization, formal analysis, writing—original draft: FM
- Validation, data curation, formal analysis: RCCR
- Validation, resources, funding acquisition, supervision, writing—original draft: TDSP
- Resources, funding acquisition, writing—original draft: LNS
- Conceptualization, resources, funding acquisition, project administration, supervision, writing—original draft and reviewing: SS, MV
- Conceptualization, resources, investigation, validation, data curation, formal analysis; project administration, supervision, writing—original draft and reviewing: RAHD

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