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Metal and metalloid concentrations in marine fish marketed in Salvador, BA, northeastern Brazil, and associated human health risks

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ABSTRACT

The research aims to determine the content of arsenic, mercury, cadmium and lead in 13 species of marine fish marketed in Salvador, Bahia, northeastern Brazil and assess the risk to human health associated with the consumption of these fish by using the Target Hazard Quotient (THQ) as a tool. The levels of these metals in fish are of great interest to the public health due to the toxicity of these elements and biomagnification in the food chain. The elements arsenic, cadmium and lead were determined by inductively coupled plasma mass spectrometry (ICP-MS) and mercury by cold vapor atomic absorption spectrometry (CVAAS). In general, intra and interspecific variations in arsenic concentrations were observed (0.01 - 1.85 mg kg^{-1}), cadmium (0.03 to 0.32 mg kg^{-1}), lead (< 0.01 - 0.10 mg kg^{-1}) and mercury (0.001 - 1.85 mg kg^{-1}), with arsenic having the highest levels in the vast majority of fish species, followed by mercury, cadmium and lead. THQ>1 value was observed only for mercury in *Seriola* spp. and *Mycteroperca intertitialis* indicating a potential risk of consumption of these species.

1. Introduction

Fish play an essential role in human nutrition due to their nutritional qualities (Núñez et al., 2018; Antão-Geraldes et al., 2018). Fish meat is a source of high-quality proteins, vitamins and omega 3 polyunsaturated fatty acids, especially EPA and DHA, in addition to having low cholesterol levels (Olmedo et al., 2013; Vieira et al., 2011; Okyere et al., 2015). Its consumption is associated with reduced cholesterol levels, decreased incidence of cardiovascular disease, beneficial effects on the immune system and contribution to neurological development in children (Okyere et al., 2015; Galuch et al., 2018). Over the years, high concentrations of trace metals and metalloids such as As, Hg, Pb and Cd, have been discharged into the aquatic environment through effluents from urban centers, agriculture and industries (Arantes et al., 2016; Bilandžić et al., 2011). High levels of these chemical elements in the environment significantly influence water quality (Filippini et al., 2018) and are considered to be the most important source of contamination in marine ecosystems (Ayotunde et al., 2012), due to the non-degradability of heavy metals and their accumulative power in living organisms, especially in fish (Okyere et al., 2015; Trevizani et al., 2019).

Fish is also one of the foods that mostly contribute to metal intake by humans, as these compounds accumulate in fish tissues and may reach toxic concentrations through trophic biomagnification (Olmedo et al., 2013; Núñez et al., 2018). This is considered potentially responsible for causing damage or reduction of central and nervous functions, lower metabolic energy levels, problems in the blood and vital organs and a series of other health risks (Ayotunde et al., 2012; Bilandžić et al., 2011).

Understanding the dangers related to the accumulation of potentially toxic metals in the human body, several countries have developed ways to inspect the levels of these elements in fish. In Brazil, the National Health Surveillance Agency (ANVISA) is responsible for establishing the maximum permitted limits for each contaminant contained in food items. Regarding the metals and metalloids analyzed in this study, the maximum tolerable limits (MTL), published in Resolution 42, dated August 29, 2013 (BRAZIL, 2013), are 1.00 mg kg^{-1} for arsenic; 0.30 mg kg^{-1} for lead; 0.05 to 0.30 mg kg^{-1} for cadmium, depending on the species (0.10 mg Kg^{-1} for *Katsuwonus* spp., *Eugerres* spp., *Anguilla* spp., *Mugil* spp., *Caranx* spp., *Scomberomorus* spp., *Sardinella* spp.,

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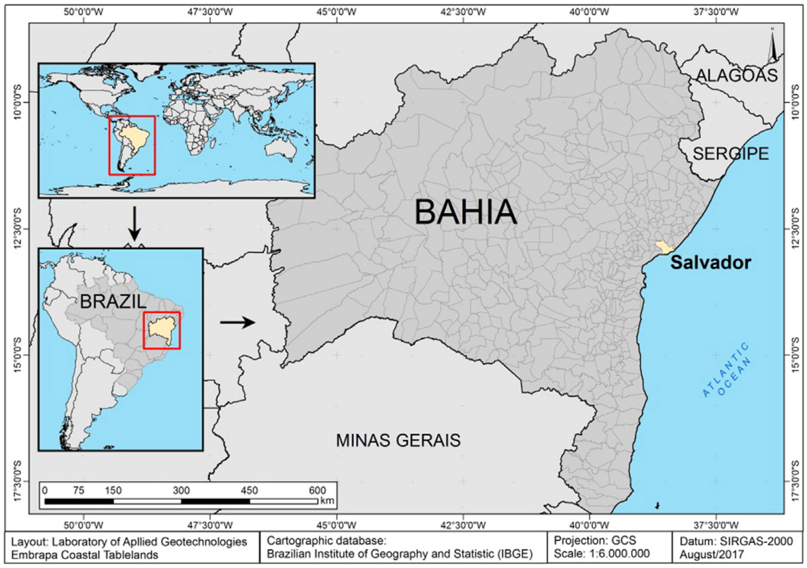

Fig. 1. Study area. Location of the Municipality of Salvador in the State of Bahia, Brazil.

Table 1
Classification, habitat and dietary category of fish species purchased in markets in the city of Salvador/BA, Northeast, Brazil.

Vulgar name	Family	Scientific name	Habitat	Dietary category
Amberjack	Carangidae	*Seriola* spp.	Demersal	Carnivorous
Tuna fish	Scombridae	*Thunnus* spp.	Pelagic	Piscivorous
Catfish	Ariidae	*Bagre* spp.	Demersal	Benthic invertebrates
Lookdown	Carangidae	*Selene vomer*	Coastal water	Carnivorous
Dolphinfish	Coryphaenidae	*Coryphaena hippurus*	Pelagic	Fish and invertebrates
Acoupa weakfish	Sciaenidae	*Cynoscion acoupa*	Demersal	Fish and invertebrates
Grouper	Serranidae	*Mycteroperca interstitialis*	Reef-associated	Piscivorous
Whitemouth croaker	Sciaenidae	*Micropogonias furnieri*	Neritic	Invertebrates
Snook	Centropomidae	*Centropomus undecimalis*	Neritic	Piscivorous
Mullet	Mugilidae	*Mugil curema*	Pelagic	Planktivous
Flounder	Paralichthyidae	*Paralichthys orbignyanus*	Coastal Lagoon	Carnivorous
King mackerel	Scombridae	*Scomberomorus cavala*	Pelagic	Fish and invertebrates
Snapper fish	Lutjanidae	*Lutjanus* spp.	Demersal	Omnivorous

Thunnus spp. and *Paralichthys* spp.; 0.20 mg Kg^{-1} for *Euthynnus* spp. and 0.30 mg kg^{-1} for *Trichiurus* spp.) and 0.50 mg kg^{-1} for mercury concerning non-carnivorous species, and 1.00 mg kg^{-1} for carnivorous species (BRAZIL, 2013). Just as with other regulatory agencies, the MTL defined by ANVISA is not sufficient to evaluate and/or control the health risks associated with fish consumption. Since the calculation does not consider the frequency of exposure or the rate of ingestion of toxic elements, it is necessary to use other ratios to better assess the problem. The Target Hazard Quotient (THQ), developed by the United States Environmental Protection Agency (USEPA, 1989), represents the relationship between exposure to a certain toxin and the reference dose, and is often used as a parameter for the assessment of risks related to fish consumption (Ramos-Miras et al., 2019).

The State of Sergipe, Brazil, has 163 km of coastline, with more than 25 thousand ha of mangroves (Almeida and Barbieri, 2008), the largest nursery of olive turtle (*Lepidochelys olivacea*) in Brazil (Oliveira et al., 2014), in addition to a representative fishing production, about two thousand tons per year (Carvalho Filho, 2019). Areas of high environmental relevance to the coast of the Northeast, Brazil. Monitoring the quality of the fish is important in order to, over the years, generate a diagnosis of anthropic influences, such as, for example, the oil spill will last for years, with immeasurable effects on the coral reefs and abundant swamp areas in the regions which could damage coastal biodiversity, generation of jobs and income mainly related to tourism and fishing (Paula et al., 2018).

In this context, this study was developed in order to measure and qualify the concentrations of arsenic, cadmium, lead and mercury in 13 marine fish species marketed in the city of Salvador, Northeastern Brazil, based on the MTL, while also analyzing the risks associated with fish consumption through the THQ.

2. Material and methods

2.1. Sample collection and preservation

The 13 analyzed fish species were selected based on information from the Fisheries and Aquaculture Statistical Bulletin (BRAZIL, 2011), as well as their availability and frequency of purchase by the population. Whole fish were identified according to Menezes and Figueiredo (1985). For fish fillets, species identification was based on the conformation of myomers and myoses in the muscles. The samples were randomly purchased from open markets, fish markets and/or supermarkets in Salvador, Bahia, northeastern Brazil (Fig. 1) in 2017, totaling 39 samples comprising three samples per species. The classification, habitat and dietary categories of the assessed fish species are displayed in Table 1.

The samples were processed at the Environmental Impact Laboratory of the Embrapa Coastal Tablelands, Aracaju, Sergipe,

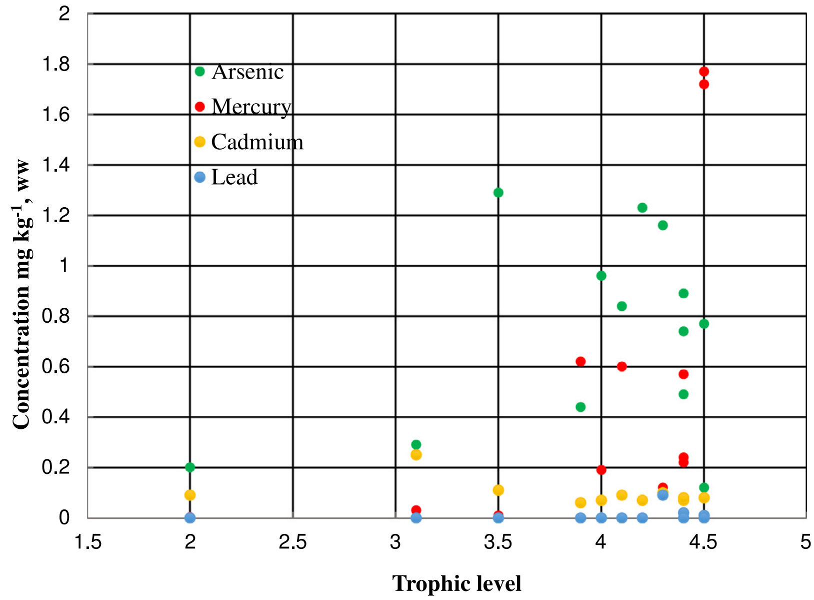

Fig. 2. Relationship between metal concentration (present work) and trophic levels in fish. The trophic level for each species was determined using FishBase information (http://www.fishbase.org/search.php).

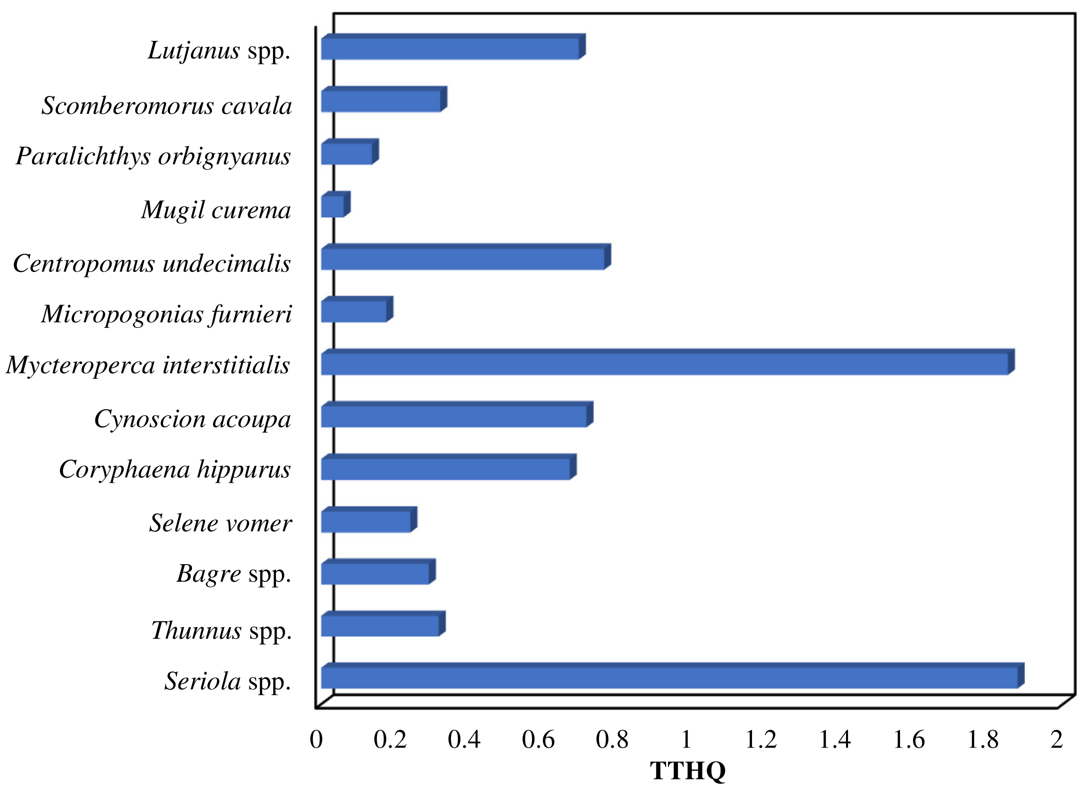

Fig. 3. Total Target Hazard Quotient (TTHQ) for the concentrations of Cd, As, Pb and Hg in fish samples sold in the city of Salvador, Northeast, Brazil.

Brazil. A mass of 100 to 150 g of the musculature of the fish's laterodorsal region and were frozen (-80 °C) for subsequent freeze-drying (Liotop model L101). The samples were kept at low temperature (-15 °C) until crushing and sieving through (250 µm mesh). To avoid contamination, all material was washed with 10% v v^{-1} nitric acid and rinsed with ultra-pure water (18.2 MΩ cm).

2.2. Standard reagents and solutions

All solutions were prepared with analytical grade reagents and ultra-pure water (18.2 M Ω cm) obtained from a Millipore Simplicity purifier (Molsheim, France). All materials used in sample preparation and analysis were previously immersed in an HNO_3 acid bath 10% v v^{-1} for 24h and rinsed with ultra-pure water

Table 2
Mean concentrations of metals (mg kg^{-1} w. w.) in 13 species of marine fish (n=3) commercialized in the city of Salvador/BA, Northeast, Brazil.

Species		Cd	As	Pb	Hg
Amberjack	*Seriola* spp.	0.08 ± 0.03 (0.04 – 0.11)	0.12 ± 0.001 (0.12 – 0.12)	0.10 ± 0.12 (0.01 – 0.19)	1.77 ± 0.08 (1.71 – 1.85)
Tuna fish	*Thunnus* spp.	0.07 ± 0.01 (0.06 – 0.09)	0.89 ± 0.56 (0.49 – 1.09)	<0.01[a]	0.22 ± 0.02 (0.21 – 0.24)
Catfish	*Bagre* spp.	0.07 ± 0.06 (0.10 – 0.01)	0.96 ± 0.13 (0.86 – 1.06)	<0.01[a]	0.19 ± 0.03 (0.16 – 0.22)
Lookdown	*Selene vomer*	0.10 ± 0.04 (0.04 – 0.08)	1.16 ± 0.72 (0.41 – 1.85)	0.09 ± 0.03 (0.06 – 0.11)	0.12 ± 0.001 (0.12 – 0.12)
Dolphinfish	*Coryphaena hippurus*	0.07 ± 0.02 (0.03 – 0.10)	0.74 ± 0.62 (0.29 – 1.18)	0.02 ± 0.001 (0.02 – 0.03)	0.57 ± 0.13 (0.41 – 0.69)
Acoupa weakfish	*Cynoscion acoupa*	0.09 ± 0.01 (0.07 – 0.10)	0.84 ± 0.28 (0.54 – 1.12)	<0.01[a]	0.60 ± 0.19 (0.46 – 0.73)
Grouper	*Mycteroperca interstitialis*	0.08 ± 0.01 (0.07 – 0.08)	0.77 ± 0.28 (0.57 – 0.77)	<0.01[a]	1.72 ± 0.03 (1.70 – 1.74)
Whitemout croaker	*Micropogonias furnieri*	0.25 ± 0.07 (0.19 – 0.32)	0.29 ± 0.02 (0.28 – 0.31)	<0.01[a]	0.03 ± 0.001 (0.03 – 0.04)
Snook	*Centropomus undecimalis*	0.07 ± 0.02 (0.05 – 0.08)	1.23 ± 0.47 (0.89 – 1.56)	<0.01[a]	0.70 ± 0.01 (0.70 – 0.71)
Mullet	*Mugil curema*	0.09 ± 0.02 0.07 – 0.11	0.20 ± 0.06 0.14 – 0.26	< 0.01	0.003 ± 0.001 (0.001 – 0.004)
Flounder	*Paralichthys orbignyanus*	0.11 ± 0.05 (0.05 – 0.15)	1.29 ± 0.29 (0.96 – 1.48)	< 0.01	0.01 ± 0.001 (0.01 – 0.02)
King mackerel	*Scomberomorus cavala*	0.08 ± 0.01 (0.07 – 0.08)	0.49 ± 0.07 (0.37 – 0.75)	< 0.01	0.24 ± 0.07 (0.19 – 0.29)
Snapper fish	*Lutjanus* spp.	0.06 ± 0.01 (0.06 – 0.07)	0.44 ± 0.06 (0.39 – 0.48)	< 0.01	0.62 ± 0.17 (0.43 – 0.75)

Mean ± standard deviation; variation interval in parentheses.
[a] < 0.01 — less than the Lead detection limit.

(18.2 MΩ cm) (Souza et al., 2019a). For ICP-MS analysis, a 100 mg L^{-1} (SpecSol®, Jacareí, Brazil) multi-element solution containing As, Cd, Pb was used to prepare an intermediate solution at a concentration of 500 µg L^{-1}, which was then used to prepare the calibration curve solutions in the concentration range of 0.25 up to 50 µg L^{-1} (Souza et al., 2019b). For Hg analysis, a 1000 mg L^{-1} (SpecSol®, Jacareí, Brazil) stock solutions were used to prepare the calibration standards.

2.3. Sample digestion

For sample As, Cd, Hg and Pb extraction about 0.5 g of freeze-dried muscle (dry weight) were mixed with 10 mL of HNO_3 (7.0 mol L^{-1}) and 2.0 mL of H_2O_2 (30% v v^{-1}) in reaction teflon flask, according to methodology adapted from Jarić et al. (2011). Subsequently, the samples were taken to a closed microwave-assisted digestion system (Anton Paar, model Multiwave 3000), with maximum power of 1400 W and the steps described in Silva et al. (2019). After cooling to room temperature, the digested samples were brought to the volume of 100 mL with ultra-pure water (18.2 MΩ cm) and stocked at 4 °C until analysis.

For mercury analysis, 15 mL of a 1:1 v v^{-1} solution (H_2SO_4:HNO_3) was added to a cold finger reactor and heated at 60 °C for 2h in a sand bath, for 0.5 g of sample. After cooling, 3 ml of H_2O_2 30% w w^{-1} were added., 15 mL of $KMnO_4$ 5% w v^{-1} and 52 mL of ultra-pure water and the digested samples were brought to the volume of 85 mL. The excess oxidant was then neutralized with 12% w v^{-1} hydroxylamine hydrochloride A 5 mL aliquot of the digested sample, added with 1 mL of $SnCl_2$ (20% w v^{-1}), was used for analysis of total mercury. (Hatch and Ott, 1968; Hight and Cheng, 2006).

2.4. Chemical analysis and quality assurance

A inductively coupled plasma spectrometer mass (ICP-MS, Thermo, Germany) was used for the quantitative determination of ^{75}As, ^{111}Cd and ^{208}Pb. The instrumental parameters of the ICP-MS were as follows: radiofrequency power of 1.3 kW, plasma gas flow of 13 L min^{-1}, auxiliary gas flow of 0.7 L min^{-1}, nebulizer gas flow of 0.87 L min^{-1}, peak jump scan mode, dwell time of 10 ms and number of readings per repetition equal to three. Reagent blanks were also processed in the same way and read for each batch of 10 samples. For the analyses, stable isotopes ^{72}Ge, ^{103}Rh and ^{205}Tl were added, as internal standards, at a concentration of 50 µg L^{-1} to both calibration curves and samples solutions. Total mercury was determined on a cold vapor atomic absorption spectrometer (CVAAS, with Zeeman correction, Russia).

The analytical trueness evaluation was performed using the certified reference material DORM-3 (fish protein certified reference material for trace metals — NRCC) based on the recovery values of the analytes.

2.5. Risk assessment

Mean concentrations of arsenic, cadmium, lead, and mercury were used for the estimation of the Target Hazard Quotient (THQ). The formula $THQ = [(EF x ED x FIR x C)/(RFD x BW x ET)] x10^{-3}$ was proposed by the USEPA (1989), where *EF* is exposure frequency (365 days year $^{-1}$); *ED* is exposure duration (70 years), equivalent to the average human-life estimate; *FIR* is food intake rate (fish = 36 g $person^{-1} day^{-1}$, as suggested by USEPA (1989); *C* is the metal concentration in the fish (µg g^{-1}); *RFD* is the oral reference dose (As = 0.3 × 10^{-3} µg g^{-1} day^{-1}, Cd = 1 x 10^{-3} µg g^{-1} day^{-1}, Pb = 4 x 10^{-3} µg g^{-1} day^{-1}, Hg 0.5 × 10^{-3} µg g^{-1} day^{-1}) (Storelli, 2008); *BW* is the mean adult body weight (70 kg) and *ET* is the noncancerous exposure time (365 days $year^{-1}$ x ED).

Table 3
THQ values for fish consumption, for cadmium, arsenic, lead, and mercury, in Salvador, Brazil.

Species	Cadmium	Arsenic[a]	Lead	Mercury
Seriola spp.	0.04	0.01	0.01	1.82
Thunnus spp.	0.04	0.05	0.0	0.23
Bagre spp.	0.04	0.05	0.0	0.2
Selene vomer	0.05	0.06	0.01	0.12
Coryphaena hippurus	0.04	0.04	0.0	0.59
Cynoscion acoupa	0.05	0.04	0.0	0.62
Mycteroperca interstitialis	0.04	0.04	0.0	1.77
Micropogonias furnieri	0.13	0.01	0.0	0.03
Centropomus undecimalis	0.04	0.06	0.0	0.72
Mugil curema	0.05	0.01	0.0	0.0
Paralichthys orbignyanus	0.06	0.07	0.0	0.01
Scomberomorus cavala	0.04	0.03	0.0	0.25
Lutjanus spp.	0.03	0.02	0.0	0.64

Values above 1.00 indicate risk.
[a]The inorganic arsenic content constitutes 3% of the total As in fish, according to (Copat et al., 2018); FSA, 2014 and (Sirot et al., 2009).

Whereas the fish preparation process for human consumption does not change the concentration or contaminant toxicity, and oral ingestion is the same as the dose absorbed by the human body (USEPA, 1989). It is inferred that when the calculated THQ is <1 does not have appreciable risk for the pollutant and when THQ is >1 there is an imminent risk of continuing to consume fish (Storelli, 2008).

3. Results and discussion

The quality of the obtained results was confirmed by the DORM-3 analysis. The results were expressed as means ± 95% confidence interval (n = 3). The limits of detection (LOD) and limit of quantification (LOQ) were determined, 10 blank solutions were prepared and analyzed, from the parameters of the analytical curves. The LOQ values obtained for ICP-MS were 0.09 μg g^{-1} for As, 0.03 μg g^{-1} for Cd and 0.02 μg g^{-1} for Pb. When CVAAS was employed, the LOQ for Hg determination was 3 μg kg^{-1}. The agreement with the certified values ranged from 89% (As) to 103% (Hg) (Silva et al., 2019). The precision was expressed as relative standard deviation (RSD%, n = 3), which was better than 10% (Thompson et al., 2002). The values showed satisfactory precision and accuracy for the applied method (Costa et al., 2013). The metal concentrations in the fish samples are given in mg kg^{-1} on a wet basis (Table 2) for comparison with values published on the same basis by the Brazilian Legislation.

3.1. Cadmium

The cadmium concentration range varied from 0.03 to 0.32 mg kg^{-1}, with the lowest value found in *Coryphaena hippurus* and the greatest in *M. furnieri*. The species *Thunnus* spp. (0.07 ±0.01 mg kg^{-1}), *M. curema* (0.09 ±0.02 mg kg^{-1}) and *S. cavala* (0.08 ±0.01 mg kg^{-1}), for which the established MTL is of 0.1 mg kg^{-1}, defined by ANVISA, showed average concentrations below the maximum tolerable. The other species (ten species) exceeded the maximum tolerable limit (MTL> 0.05 mg kg^{-1}, defined by ANVISA). Silva et al. (2019) also reported cadmium concentrations above the MTL for *Bagre* spp. and *M. curema* (0.19 mg kg^{-1}).

The mean levels of Cd for all the fish in this study was of 0.09 ± 0.04 mg kg^{-1}, similar to those obtained in other studies, of 0.09 ± 0.01 mg kg^{-1} in the Adriatic Sea (Storelli and Barone, 2013); 0.09 ± 0.02 mg kg^{-1} for fish marketed in Andalusia, Spain (Olmedo et al., 2013), 0.08 ± 0.02 mg kg^{-1} for commercialized fished in Aracaju, Northeastern Brazil (Silva et al., 2019) and 0.07 ± 0.05 mg kg^{-1} for those marketed in southern Italy (Barone et al., 2015). Lower levels, 0.003 ± 0.003 mg kg^{-1} of cadmium compared to the values detected herein were reported off the coast of Tuscany, Italy (Bonsignore et al., 2018), of 0.006 ± 0.003 mg kg^{-1} in the Persian Gulf (Keshavarzi et al., 2018), 0.004 ± 0.002 mg kg^{-1} in Kelantan, Malaysia (Salam et al., 2019), 0.009 ± 0.004 mg kg^{-1} in the Black Sea off Bulgaria (Makedonski et al., 2017), 0.024 ± 0.010 mg kg^{-1} in Galicia, Spain (Núñez et al., 2018) and 0.023 ± 0.015 mg kg^{-1} in the South China Sea (Gu et al., 2017). On the other hand,Elnabris et al. (2013) determined cadmium concentrations below the detection limit of 0.002 mg kg^{-1} in five of six evaluated species. The analyzed fish reached higher cadmium concentrations than in most of the presented studies. However, high levels, of 0.13 ± 0.03 mg kg^{-1}, were reported by Basim and Khoshnood (2016) in the Caspian Sea, Iran, who attributed the result to unmanaged transport activities, river runoff, untreated sewage discharge by coastal settlements, and dumping of toxic and industrial wastes at sea.

In relation to *S. cavalla*, the levels of 0.08 ±0.01 mg kg^{-1} were higher when compared to those detected in other assessments for fish belonging to the Scombridae family, as follows: *Scomberomorus* spp. 0.0038 ± 0.0048 mg kg^{-1} from Bogota, Colombia (López-Barrera and Barragán-Gonzalez, 2016), *Scomber japonicus* 0.01 ± 0.01 mg kg^{-1} from the coast of Almería, Spain (Ramos-Miras et al., 2019) and 0.007 ± 0.003 mg kg^{-1} from Eastern Central Atlantic Ocean (Vieira et al., 2011), *Scomber scombrus* 0.003 ± 0.003 mg kg^{-1} from the Gulf of Catania, Italy (Copat et al., 2018), 0.001 ± 0.001 mg kg^{-1} from the Mediterranean coast off Spain (Olmedo et al., 2013) and 0.03 ±0.01 mg kg^{-1} from the Central Adriatic Sea (Perugini et al., 2014). Likewise, *M. curema* 0.09 ± 0.02 mg kg^{-1} obtained from the commerce of Salvador, Bahia, Brazil contained Cd concentrations higher than 0.009 ± 0.004 mg kg^{-1} found in samples of the same species commercialized in São Paulo, SP, Brazil, (Morgano et al., 2011). However, comparisons of concentrations between fish species obtained from commerce are difficult, because the geographic origins of the fish are unknown (Burger and Gochfeld, 2005).

Cadmium is introduced into the environment through natural (volcanic emissions and weathering of rocks) and anthropogenic processes such as the burning of fossil fuels, the incineration of domestic and industrial waste and the use of certain fertilizers containing this element. Cadmium can be absorbed by fish by passive diffusion in the aquatic environment through the gills and/or through the ingestion of the first links in the food chain, i.e. microorganisms and plankton (EFSA, 2009). In muscle tissue, cadmium is bound to proteins and displays the ability to bioaccumulate due to its very slow elimination rate. It is a highly toxic metal that can be concentrated in the human body for long periods of time. In addition to being classified as a carcinogen, its accumulation is related to impaired renal function, reproductive capacity, liver dysfunction and nervous system (Ullah et al., 2017; Zhong et al., 2018; Rodríguez et al., 2015). The biologic half-life of cadmium in the kidney is estimated up to 38 years and the half-life of cadmium in the liver is up 19 years (ATSDR, 2008). Despite the cadmium content of most species assessed herein being above the MTL set by Brazilian legislation, the THQ, which takes into account daily doses and frequency of ingestion, displayed results of less than 1, configuring no health risk associated with the consumption of these species (Table 3). Corroborating this study, Vieira et al. (2011), also applying the THQ tool in fish sold in the city of Porto, Portugal, reported the absence of adverse effects on human health due to fish cadmium content. Silva et al. (2019) reported 0.03 ≤ THQ ≤ 0.05, for commercial fish in Aracaju, Northeastern Brazil, also configuring no associated health risk.

3.2. Arsenic

The lowest and highest As concentrations were 0.01 mg kg^{-1} in *Mugil curema* and 1.85 mg kg^{-1} in *Selene vomer*, respectively. Similar results were observed for commercial fish in Aracaju, Northeastern Brazil, ranging from 0.07 mg kg^{-1} (*Cynoscion* spp.) to 2.03 mg kg^{-1} (*R. canadum*) (Silva et al., 2019). The average concentration of arsenic in the studied species was of 0.72 $\pm$ 0.39 mg kg^{-1}, similar to the value of 0.73 $\pm$ 0.05 mg kg^{-1} reported by Makedonski et al. (2017) in fish from the Black Sea off Bulgaria, 0.76 $\pm$ 0.78 mg kg^{-1} reported by Raknuzzaman et al. (2016) in fish from Bhola, Bangladesh, and 0.69 $\pm$ 0.31 mg kg^{-1} reported by Silva et al. (2019) in fish from Aracaju, Northeastern Brazil.

The levels of arsenic found in the predatory species *Selene vomer*, *Centropomus undecimalis* and *Paralichthys orbignyanus* exceeded the maximum value determined by the Brazilian legislation of 1.0 mg kg^{-1}, while *Thunnus* spp., *Bagre* spp. and *Cynoscion acoupa* contained concentrations close to the established limit. These high levels may be related to the habitat characteristics (Morgano et al., 2011; Silva et al., 2019) and the carnivorous diet of these species, increasing As concentrations the food chain (Silva et al., 2019); Rodríguez, Ríos & Botero, 2015).

The results were not very variable when compared to the arsenic values reported as ranging from 0.01 to 70.9 mg kg^{-1} in fish from coastal waters in the eastern Adriatic Sea (Bilandžić et al., 2011). High arsenic levels concentrations were detected in fish from the Central Adriatic Sea, with an average of 49.30 $\pm$10.60 mg kg^{-1} for demersal species and 33.03 $\pm$2.27 mg kg^{-1} for pelagics (Perugini et al., 2014). In the Mediterranean Sea, *Diplodus annularis*, a benthic predator, contained high levels of 10.82 $\pm$ 0.02 mg kg^{-1} (dw), probably related to a continuous discharge of metals from industrial activity in the study region (Zohra and Habib, 2016). On the other hand, lower levels (0.18 $\pm$ 0.18 mg kg^{-1}) were observed in fish samples sold in the central market of Granada, Spain (Olmedo et al., 2013).

In the present study, *M. curema* contained arsenic levels of 0.20 $\pm$0.01 mg kg^{-1} below those detected in *Mugil cephalus* from Rosignano, Italy, of 0.42 $\pm$0.03 mg kg^{-1} (Bonsignore et al., 2018) and above 0.1 $\pm$0.1 mg kg^{-1} reported for the Black Sea off Bulgaria (Makedonski et al., 2017).

The arsenic concentrations observed in *Scomberomorus cavalla* (0.49 $\pm$ 0.07 mg kg^{-1}) were lower in comparison to *Scomber scombrus* (2.507 $\pm$ 0.748 mg kg^{-1}) from the Gulf of Catania and *Scomber japonicus* (0.9384 $\pm$ 0.177 mg kg^{-1}) from the Atlantic Ocean, in northeastern Portugal (Copat et al., 2018). Higher values were recorded for *Micropogonias furnieri* (0.73 $\pm$ 0.34 mg kg^{-1}) and *Bagre* spp. (8.9 $\pm$ 11.3 mg kg^{-1}) in another Brazilian region, in coastal waters off Rio de Janeiro (Gao et al., 2018).

Arsenic is widespread in the aquatic environment due to both natural processes, such as volcanic activities and rock weathering, and anthropogenic activities, especially mining and burning fossil fuels (Kumari et al., 2017; Peshut et al., 2008), the use of pesticides, herbicides and fungicides containing arsenic (Bosch et al., 2016) and copper smelting and glass making (Castro-González and Méndez-Armenta, 2008). This metalloid can be found in fish in different chemical forms, which differ according to their state of oxidation and toxicity (Peshut et al., 2008; Muñoz et al., 2000). Organic arsenic species are present in greater amounts, above 90%, as arsenobetaine, and are considered non-toxic (Lin et al., 2005), while the inorganic arsenic forms As V and As III, are toxic and the main forms responsible for causing risks to human health, such as cardiovascular, liver, kidney and hematological diseases. Moreover, there is evidence of the relationship between arsenic concentrations in the body and the development of cancer (Gao et al., 2018; Muñoz et al., 2000).

Due to the variability of arsenic chemical forms and toxicity, the determination of total arsenic levels is not sufficient to assess the negative effects of inorganic arsenic on humans (Peshut et al., 2008; Li and Wang, 2019), so a distinction between toxic and non-toxic fractions is required (Gao et al., 2018). To this end, the literature estimates that inorganic arsenic represents about 10% of total arsenic in marine fish (Lin et al., 2005). However, the United Kingdom Food Standards Agency (FSA), as well as more recent studies, identify inorganic arsenic as about 3% of the total Copat et al., 2018; EFSA, 2014; Sirot et al., 2009. The THQ data for inorganic arsenic (Inorg. As) was calculated based on the latter information. The calculated THQ values in this study are shown in Table 3.

The THQ range of inorganic arsenic varied from 0.01 to 0.07 and none of the species in this study represents consumer risks, since the $THQ_{Inorg.As}$ was lower than 1. The highest $THQ_{Inorg.As}$ values, despite being below the risk estimate, were found for the carnivorous species *P. orbignayanus* (0.07), *S. vomer* and *C. undecimalis*, at 0.06. In commercial fish from Aracaju, Northeastern Brazil, the THQ ranged from 0.03 to 0.26, higher than the values found in the present study (Silva et al., 2019). Average THQ_{As} levels greater than 1 have been reported in the literature, of 1.37 by Vieira et al. (2011) and of 2.77 by Zohra and Habib (2016) in carnivorous species, demonstrating a trend of increasing arsenic concentrations along the food chain.

3.3. Lead

Lead concentrations in the analyzed fish species were above 0.01 mg kg^{-1} for only three species. The highest average concentration, of 0.10 $\pm$0.12 mg kg^{-1}, was detected in *Seriola* spp., followed by *S. vomer*, at 0.09 $\pm$0.03 mg kg^{-1}, and *Coryphaena hippurus*, at 0.02 $\pm$0.03 mg kg^{-1}. All levels were below that established by the Brazilian legislation of 0.30 mg kg^{-1} (BRAZIL, 2013). Concentrations below the lead limit of detection have also been reported in other studies, i.e. for *Trachurus trachurus* (Aydin and Tokalioğlu, 2015); Storelli et al. 2008), *Sparus aurata* (Aydin and Tokalioğlu, 2015; Elnabris et al., 2013), *Thunnus* spp. (Núñez, García, Alonso & Melgar, 2018) and *Scorpaena porcus* (Bonsignore et al., 2018).

The average lead content for all the fish in this study of 0.07 $\pm$ 0.04 mg kg^{-1} was higher than that reported in the literature for fish, of 0.05 $\pm$ 0.01 mg kg^{-1} in fish from the Black Sea, Bulgaria (Makedonski et al., 2017), 0.01 $\pm$ 0.02 mg kg^{-1} in fish from Granada, Spain (Olmedo et al., 2013), 0.04 $\pm$ 0.01 mg kg^{-1} in fish from the Central Adriatic Sea, Italy (Perugini et al., 2014), 0.031 $\pm$ 0.012 mg kg^{-1}, Bogota, Colombia (López-Barrera and Barragán-Gonzalez, 2016), 0.006 $\pm$ 0.005 mg kg^{-1} in fish from the Northwest Mediterranean Sea (Marengo et al., 2018), 0.05 $\pm$ 0.06 mg kg^{-1} in fish from Alaska (Burger et al., 2014); and 0.09 $\pm$ 0.07 mg kg^{-1} in fish from Aracaju, Northeastern Brazil (Silva et al., 2019). On the other hand, the range of variation in lead levels from 0.01 to 0.19 mg kg^{-1} was smaller than that recorded from 0.01 to 0.55 mg kg^{-1}, in the Caspian Sea, in Iran (Basim and Khoshnood, 2016), 0.03 to 0.25 mg kg^{-1}, in the Adriatic Sea (Storelli & Barone et al. 2013) and $<$ 0.02 to 2.92 mg kg^{-1} in São Paulo, Brazil (Morgano et al., 2011).

Alina et al. (2012), when investigating species from the Malacca Strait, identified Pb levels higher than those reported herein for fish of the same family, of 0.015 $\pm$ 0.007 mg kg^{-1} (*Epinephelus sexfasciotus*), 0.26 $\pm$ 0.04 mg kg^{-1} (*Lutjanus malabaricus*), 0.025 $\pm$ 0.021 mg kg^{-1} (*Scomberamorus mucalatus*) and 0.09 $\pm$ 0.08 mg kg^{-1} (*Rastrelliger kanagurta*). Similarly, Elnabris et al. (2013) determined amounts of 0.552 $\pm$0.479 mg kg^{-1} in *M. furnieri* and 0.172 $\pm$0.092 mg kg^{-1} in *M. curema*, in fish from the Gaza Strip, Palestine, much higher than those reported in this study for the same species, with lead content of less than 0.01 mg kg^{-1}.

Only *Seriola* spp. and *S. vomer* presented lead THQ values of some significance, equal to 0.01, while the other assessed species contained concentrations below the limit of detection of < 0.01 mg kg^{-1}, although it is suggested that none of the analyzed species represents consumer risks. Lead THQ values are similar to those reported by Basim and Khoshnood (2016), of 0.01, Keshavarzi et al. (2018), of 0.035 and Zhong et al. (2018), of 0.032. The THQ values were higher than those reported by Copat et al. (2018), of 0.0031, Vieira et al. (2011), of 0.003 and Marengo et al. (2018), of 0.0015, and lower than those observed by Pal and Maiti (2019), of 1.61 and Ullah et al. (2017), of 0.1223. Results similar to the present study were reported by Watanabe et al. (2003), although other risk indicators were used, and indicated that lead offers no appreciable risk to fish consumers.

Lead is naturally found in rocks and soils, where erosion and leaching contribute to its entry into the aquatic environment. It is widely used in battery, paint and glass production and recycling industry, and combustion of lead-added gasoline in the past was considered the predominant source of this metal to the atmosphere (Storch et al., 2003). Atmospheric deposition is the main entry route of lead into the oceans. The divalent form Pb II is absorbed by the gills, enters fish bloodstream and accumulates in tissues, especially in muscle, with fish consumption being an important route of exposure in humans (Nussey et al., 2000).

Lead is considered a non-essential element, accumulating in teeth, bones, liver, kidney, spleen, and other vital organs such as the lungs and brain, and able to cross the blood–brain barrier and the placenta, displaying the potential to cause many harmful effects, such as neurotoxicity, nephrotoxicity and damage to the hematological and cardiovascular systems (García-Lestón et al., 2010). Lead poisoning symptoms include headache, irritability, abdominal pain and several nervous system symptoms (Castro-González and Méndez-Armenta, 2008).

3.4. Mercury

The highest and lowest mercury concentrations were, respectively, 1.85 mg kg^{-1} in *Seriola* spp. and 0.001 mg kg^{-1} in *M. curema*. The general mean Hg concentration was 0.52 ± 0.59 mg kg^{-1}, with levels decreasing in the following order: *Seriola* spp. > *M. interstitialis* > *C. undecimalis* > *Lutjanus* spp. > *C. acoupa* > *C. hippurus* > *S. cavalla* > *Thunnus* spp. > *Bagre* spp. > *S. vomer* > *M. furnieri* > *Paralichthys orbignyanus* > *M. curema*. This order is especially influenced by dietary fish habits, since carnivorous fish contain greater Hg levels than herbivores, planktivores and omnivores, as mercury tends to accumulate along the food chain (Ahmad et al., 2015; Bentley and Soebandrio, 2017).

In *M. curema*, a non-carnivorous species, the levels ranged from 0.001 to 0.004 mg kg^{-1}, with an average of 0.003 ±0.001mg kg^{-1}, well below the maximum limit allowed by the Brazilian legislation of 0.50 mg kg^{-1} (BRAZIL, 2013). However, Hg contents in *Seriola* spp. (1.71 to 1.85 mg kg^{-1}) and *M. interstitialis* (1.70 to 1.74 mg kg^{-1}) were above the MTL of 1.0 mg kg^{-1} specific for carnivorous fish (BRAZIL, 2013). In another study, Silva et al. (2019) found mercury concentrations ranging from 0.005 mg kg^{-1} (*M. curema*) to 1.440 mg kg^{-1} (*C. undecimalis*) in fish marketed in Aracaju, Northeastern Brazil, with an average concentration for planktivorous fish of 0.03 ± 0.01 mg kg^{-1} and for the predator group, of 0.71 ± 0.07 mg kg^{-1} (Silva et al., 2019).

The average Hg content of 0.52 ± 0.59 mg kg^{-1} observed in the present study is in agreement with those reported in other studies, of 0.51 ± 0.39 mg kg^{-1} in fish from the Mediterranean Sea, Italy (Storelli et al. 2008), 0.57 ± 1.24 mg kg^{-1} in fish from the Aleutian Island, Alaska (Burger et al., 2014) and in fish from two regions off the Pacific Coast of Mexico, of 0.55 ± 0.34 mg kg^{-1}, Gulf of California and 0.60 ± 0.31 mg kg^{-1} and Baja California (Cruz-Acevedo et al., 2019). The latter study associates the Hg concentrations determined in these areas to metals and organic compounds transported by the Colorado River; urban settlements and mining located around the coast, and marine currents, which transport high amounts of this element. In the Persian Gulf, the average concentrations, considered high, were of 4.10 ± 1.90 mg kg^{-1} (Keshavarzi et al., 2018) and 3.15 ± 1.98 mg kg^{-1} (Fard et al., 2015), resulting from high amounts of contaminants present in urban and agricultural effluents around the area.

High Hg levels in *Epinephelus itajara* samples collected off the coast of Florida, of 1.12 ± 0.23 mg kg^{-1}, were reported by Malinowski (2019). The *Mycoteroperca interstitialis* samples assessed herein, which belong to the same family as *E. itajara*, also contained high Hg levels, of 1.72 ± 0.03 mg kg^{-1}. These fish, despite belonging to a medium trophic level, contain Hg traces equivalent to those found in top predators, indicating that these values may be correlated not only to dietary habits, but also to large sizes and longevity, both typical of the aforementioned species (Malinowski, 2019).

Regarding habitat, the pelagic species evaluated in this study contained an average Hg content of 0.26 ±0.23 mg kg^{-1} below the levels reported by Perugini et al. (2014), of 0.37 ± 0.01 mg kg^{-1} in samples from the Central Adriatic Sea, Italy, 0.58 ± 0.60 mg kg^{-1} in fish from North Pacific, Hawaii (Kaneko and Ralston, 2007), and 0.51 ± 0.84 mg kg^{-1} in fish from Augusta Bay, Italy (Bonsignore et al., 2018), the latter area contaminated by the discharge of the largest petrochemical plant in Europe. On the other hand, low levels, 0.07 ± 0.00 mg kg^{-1}, have been indicated by Anual et al. (2018) in fish from the western peninsula of Malaysia, 0.002 ± 0.001 mg kg^{-1}, Malacca strait (Alina et al., 2012) and 0.05 ± 0.03 mg kg^{-1} in samples from San Francisco Bay, California (Greenfield and Jahn, 2010).

Concerning the demersal fish in this study, the average level of 0.55 ± 0.64 mg kg^{-1} was similar to the value of 0.53 ± 0.07 mg kg^{-1} reported by Perugini et al. (2014), less than 0.72 ± 0.66 mg kg^{-1} reported by Bonsignore et al. (2018), and higher than the other studies, of 0.12 ± 0.07 mg kg^{-1} (Anual et al., 2018) and 0.004 ± 0.002 mg kg^{-1} (Alina et al., 2012). Based on these data, a general trend of increasing Hg concentration with depth is noted (Bonsignore et al., 2018), where bottom fish (demersals) are more exposed to contamination sources, since they live in direct contact with sediments, unlike pelagics that inhabit upper areas (Storelli, 2008; Anual et al., 2018).

Natural Hg contributions to the environment include volcanic activity and forest fires, while the most significant anthropogenic activities comprise mining, burning fossil fuels and the smelting of Pb, Cu and Zn (Boening, 2000). Mercury contamination is much more serious than other metals, due to its high ability to dissipate via the atmosphere, considered one of the main Hg dispersal routes into the environment (Travnikov and Ryaboshapko, 2002), leading to transport to remote areas across the planet. Several mercury forms occur naturally, with metallic mercury, mercury sulfide, mercury chloride and methylmercury being more common (Acquavita and Bettoso, 2018). Some natural processes, termed methylation and mediated by bacteria, can transform inorganic mercury into organic forms, with methylmercury (MeHg) as the most important, which can biomagnify along the trophic chain (Malvandi and Alahabadi, 2019). In carnivorous fish, MeHg, represents about 90% of total mercury content (Li and Wang, 2019; Malinowski, 2019) and considered the main chemical species in terms of toxic human health effects (Jewett and Duffy, 2007). Mercury accumulation in solution by single-celled organisms and phytoplankton is mainly affected by passive diffusion of neutral forms and the active transport of ionized forms across cell membranes (Moye et al., 2002). Phytoplankton bioaccumulation is the initial transferal process of this metal along the aquatic food chain (Silva et al., 2019). In successive links, dietary habits are a decisive component in Hg content amplification mechanisms in animal tissues. Fish consumption is the main Hg absorption route by humans (Anual et al., 2018; Bonsignore et al., 2018). Human exposure to this element can cause neurological, gastrointestinal, renal, dermatological, cardiovascular and immune disorders, and lead to issues concerning fetal formation (Ahmad et al., 2015; Jewett and Duffy, 2007; Zahir et al., 2005).

The trophic level indicates the position of an organism in the food web based in the diversity of prey items analysis in the stomachs, for fishes this value generally ranges from 2.0 (Herbivores/Planktivore) to 4.7 (Carnivore/ Piscivore top predators) (Pauly et al., 2021; FAO, 2008–2021). Although many *factors influence concentrations of metals in fishes, it can be observed a strong non-linear correlation between* biomagnification and trophic position (Buck et al., 2019). The tendency showed in the present work (Fig. 2) corroborates with other studies in literature, the highest concentrations of metals occurring in species of higher trophic level, so the metal content of herbivorous fish is generally lower than that of carnivorous and piscivore such like tunas and mackerel (Salazar-Camacho et al., 2020; Miao et al., 2020; Storelli et al., 2020).

The relation demonstrated in Fig. 2 is important to evaluate the risk and benefits of fish consumption, mainly because humans have a preference for large-sized fishes which have trophic levels higher than 4 resulting in more exposure to toxic metals (highlighting mercury and arsenic) (Pauly et al., 2021; Storelli et al., 2020).

The average THQ_{Hg} of all 13 listed species was 0.522, among which only two species, *Seriola* spp. (1.88) and *M. interstitialis* (1.77) presented THQ $>$ 1, representing potential human health risk. Silva et al. (2019) reported THQ_{Hg} in fish marketed in Aracaju, Northeastern Brazil ranging from 0.00 to 1.28, with values above 1 representing health risks.

3.5. Total target hazard quotient

Despite the individual concentration of some metals in a given fish species not leading to human health hazards, when these contaminants are present in the same samples and are consumed together, their sum may lead to synergistic effects and potential consumer risks. In order to assess this effect, the total Target Hazard Quotient (TTHQ), which comprises the simple sum of the THQ of each metal is calculated, as $TTHQ = THQ_{As}+THQ_{Cd}+THQ_{Pb}+THQ_{Hg}$, proposed by Chien et al. (2002). With this, the highest and lowest TTHQ values in the present study were 1.88 in *Thunnus* spp. and 0.06 in *M. curema*, respectively.

Only two species presented total Target Hazard Quotient values greater than 1, in *M. interstitialis* (1.85) and *Thunnus* spp., mentioned previously (Fig. 3). Not surprisingly, these fish presented THQ_{Hg} values greater than 1, contributing significantly to the high Target Hazard Quotient value. In a study carried out on fish commercialized in Aracaju, Northeastern Brazil, amberjack, dolphinfish and snook fish all displayed a TTHQ $>$ 1, presenting potential risks to human health (Silva et al., 2019).

In general, intra and interspecific variations were observed concerning arsenic, cadmium, lead and mercury concentrations in the fish evaluated herein. As pointed out by Burger and Gochfeld (2005), the comparison and interpretation of elemental levels between different fish species obtained through the purchase of marketed fish, as in the case of this research, becomes difficult, as the geographical origins of the fish are unknown, although consumers are indeed exposed to certain contaminants when they buy fish.

4. Conclusions

Arsenic was present in the highest levels in most fish species assessed herein, followed by mercury, cadmium and lead. Average arsenic levels above the MTL were detected in *S. vomer*, *C. undedimalis* and *P. orbignyanus*, while average mercury concentrations above the MTL were found in *Seriola* spp. and *M. interstilialis*. It is paramount that the legislation stipulate maximum inorganic arsenic limits in different fish species.

With regard to cadmium, only three species, *Thunnus* spp., *M. curema* and *S. cavala*, presented average levels below the specific MTL. In the case of lead, the concentrations found in the evaluated fish do not represent consumer health risks.

Only mercury presented a THQ $>$ 1, in *Seriola* spp. and *M. interstitialis*, which were also the only species with a TTHQ greater than 1. Therefore, it is important to assess the consumption of these species, given the potential risk they pose to human health. In general, although some specimens exceed the MTL and considering the THQ and TTHQ values, the consumption of eleven of the thirteen studied fish do not suggest imminent risks to human health.

The assessment of potential consumption risks associated with levels of potentially toxic metals in fish are essential in order for health and sanitary surveillance bodies to develop guidelines and warnings for the general population on the safe consumption of the target species, particularly for vulnerable groups, such as pregnant or lactating women and children.

CRediT authorship contribution statement

Carlos A. da Silva: Responsible for acquiring the resource for the research, Participated in the entire methodological process for the construction of the paper. **Carlos A.B. Garcia:** Responsible for supporting the analyzes, Revising the manuscript. **Hortência L.P. de Santana:** Collaborated in every experimental part. **Gabriela C. de Pontes:** Collaborated in every experimental part. **Julio C. Wasserman:** Responsible for supporting the analyzes, Revising the manuscript. **Silvânio S.L. da Costa:** Responsible for supporting the analyzes, Revising the manuscript, Ensuring that the descriptions are accurate and agreed by all authors.

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.

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