





# Mercury in piscivorous and detritivorous fish from the Teles Pires River basin, Southern Amazon

Mercúrio em peixes piscívoros e detritívoros da bacia do rio Teles Pires, Amazônia Meridional

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## ABSTRACT

Mercury accumulation in fish can differ among species living in the same water body, and these differences can be related to their life cycle, feeding habits, and position in the trophic chain. The objective of this study was to evaluate the concentrations of total mercury (THg) in the muscle, liver, kidneys, and gills of the detritivorous fish *Prochilodus nigricans*, and the piscivorous fish *Cichla mirianae*, *Hydrolycus armatus*, and *Hydrolycus tatauaia* from the Teles Pires River basin. Samplings were conducted in September 2015 (dry season) and May 2017 (recession season). The THg concentrations in the liver of *P. nigricans* from the Teles Pires basin ( $1,05 \pm 1,24$  mg.kg<sup>-1</sup>) and *C. mirianae* from the Peixoto River ( $0,46 \pm 0,26$  mg.kg<sup>-1</sup>) were significantly higher than in the other tissues analyzed; for the other species, the concentrations in the liver and kidneys were similar. THg concentrations in the muscle were significantly higher ( $p=0,0002$ ) in piscivorous fish (*H. armatus* 0,30 mg.kg<sup>-1</sup>; *H. tatauaia* 0,18 mg.kg<sup>-1</sup>; *C. mirianae* from the Teles Pires River 0,17 mg.kg<sup>-1</sup> and from the Peixoto River 0,21 mg.kg<sup>-1</sup>) than in the detritivorous fish (*P. nigricans* from the Teles Pires basin 0,05 mg.kg<sup>-1</sup>), which was expected due to the biomagnification of mercury. The vast majority of specimens had THg concentrations in the muscle below that recommended for human consumption by the World Health Organization but, considering that the region has a high consumption of fish, the daily mercury intake exceeds the limit. The detritivorous fish *P. nigricans* presented the lowest THg concentrations in the muscle; therefore, it can be preferentially consumed by the general population, especially by sensitive groups (lactating women, infants, and children), and frequent consumers such as indigenous and riverine populations.

**Keywords:** Tapajós River basin; bioaccumulation; biomagnification; contamination; environmental risk.

## RESUMO

O acúmulo de mercúrio em peixes pode ser diferente entre as espécies que vivem no mesmo corpo hídrico, e tais diferenças podem estar relacionadas ao ciclo de vida, hábitos alimentares e sua posição na cadeia trófica. O objetivo deste estudo foi avaliar as concentrações de mercúrio total (THg) no músculo, fígado, rins e brânquias do peixe detritívoro *Prochilodus nigricans*, e dos piscívoros: *Cichla mirianae*, *Hydrolycus armatus* e *Hydrolycus tatauaia* da bacia do Teles Pires. As coletas foram realizadas em setembro de 2015 (seca) e maio de 2017 (vazante). As concentrações de THg no *P. nigricans* da bacia do Teles Pires ( $1,05 \pm 1,24$  mg.kg<sup>-1</sup>) e *C. mirianae* do rio Peixoto ( $0,46 \pm 0,26$  mg.kg<sup>-1</sup>) foram significativamente maiores no fígado do que nos demais tecidos analisados; para as demais espécies, as concentrações no fígado e rins foram similares. As concentrações de THg no músculo foram significativamente mais elevadas ( $p=0,0002$ ) nos piscívoros (*H. armatus* 0,30 mg.kg<sup>-1</sup>; *H. tatauaia* 0,18 mg.kg<sup>-1</sup>; *C. mirianae* no rio Teles Pires 0,17 mg.kg<sup>-1</sup> e no rio Peixoto 0,21 mg.kg<sup>-1</sup>) do que no detritívoro (*P. nigricans* na bacia do Teles Pires 0,05 mg.kg<sup>-1</sup>), o que era esperado pela biomagnificação do mercúrio. A grande maioria dos exemplares apresentaram concentrações de THg no músculo abaixo do permitido para consumo humano pela Organização Mundial da Saúde, mas considerando que a região tem um alto consumo de pescado, a ingestão diária de mercúrio ultrapassa o limite. O peixe detritívoro *P. nigricans* apresentou as menores concentrações de THg no músculo, portanto, pode ser preferencialmente consumido pela população em geral, especialmente por grupos sensíveis (lactantes, lactentes e crianças) e consumidores frequentes como indígenas e ribeirinhos.

**Palavras-chave:** bacia do rio Tapajós; bioacumulação; biomagnificação; contaminação; risco ambiental.

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## Introduction

Trace metals accumulate in the various tissues of the fish body in different concentrations. These differences result from the affinities of the metals with certain fish tissues, forms of absorption, accumulation and excretion rates (Basta et al., 2023). The accumulation of mercury (Hg) in fish can differ between species living in the same body of water, which can be related to feeding habits and the entire life cycle of the species. Aquatic organisms at the top of the food chain generally have higher Hg concentrations than those at lower trophic levels, even in the same habitat (Basta et al., 2023). In aquatic environments, sediment naturally accumulates a high Hg load, especially adsorbed to finer particles such as silt and clay (Bettoso et al., 2023). In this way, detritivorous fish feeding on sediment can accumulate high concentrations of Hg, sometimes even higher than carnivorous fish, who are exposed to it by eating insects and other fish (Tesser et al., 2021).

Bioaccumulation and biomagnification are processes that interact and can generate differences in concentrations between the tissues of the same organism as well as in fish of different species that inhabit the same body of water and belong to the same trophic guild. Studies in the Amazon basin have shown high Hg concentrations in carnivorous and piscivorous fish (Alcala-Orozco et al., 2020). Hg bioaccumulation in aquatic organisms generally varies according to the structure of the food chain and the size/age of the fish. Plankton would be at the bottom, followed by small fish, and piscivorous fish in which the highest concentrations of Hg are expected (Evers et al., 2024). In order to understand the risk of Hg to ichthyofauna and their consumers, studies are needed on Hg in fish and the environment in which they live (Silva and Lima, 2020; Bettoso et al., 2023). Studies investigating Hg concentrations in fish in the Tapajós River basin (of which Teles Pires is one) reveal the highest concentrations of total mercury (THg) in carnivores, with some studies showing THg concentrations above that recommended by the World Health Organization (WHO) for human consumption (Matos et al., 2021; Oliveira et al., 2022). Hg contamination affects various organs and systems in the human body, including the nervous system, lungs, heart, liver, kidneys, and skin, in addition to causing damage to fetal health (WHO, 2021).

Currently, the main economic activities in the Teles Pires River basin are cattle ranching, agriculture, gold mining, and hydroelectric power plants (Guimarães, 2020; Matos et al., 2020; Lucanus et al., 2021). In the 1970s, with the onset of colonization in the Northern region of Mato Grosso state, Brazil, there was intense logging activity, deforestation, fires, erosion, and gold mining (Matos et al., 2021). Mining activity declined after the 1990s due to the depletion of easily accessible gold deposits (Guimarães, 2020). However, gold's value has been overvalued since 2011, fueling a new gold rush in the Amazon, less massive and more discreet, using cyanidation alone or in combination with amalgam (Guimarães, 2020). Thus, logging, mining, agriculture, and farming activities are currently adding their impacts to those of hydroelectric plants (Matos et al., 2020; Matos et al., 2021; Freitas et al.,

2022). Hydroelectric reservoirs provide a favorable environment for mercury methylation (MeHg), as they have anoxic, slightly acidic waters with high concentrations of dissolved organic matter and intense microbial activity (Costa and Costa, 2023; Evers et al., 2024).

To understand THg accumulation in fish with different feeding habits, we analyzed four species of bony fish belonging to three trophic levels in the Teles Pires River basin: *Prochilodus nigricans* (2nd trophic level, primary consumer), *Hydrolycus armatus* and *Hydrolycus tatauaia* (3rd trophic level, secondary consumer), and *Cichla mirianae* (4th trophic level, tertiary consumer). The species *H. armatus* (Jardine and Schomburgk, 1841) and *H. tatauaia* (Toledo-Piza, Menezes and Santos, 1999), which belong to the Cynodontidae family, and are popularly known as “peixe-cachorra”, make reproductive migrations and have a piscivorous feeding habit (Dary et al., 2017; Barbosa et al., 2018). *C. mirianae* (Kullander and Ferreira, 2006), the “tucunare”, belongs to the Cichlidae family, is territorial, and has a piscivorous eating habit (Gomiero and Braga, 2004). *P. nigricans* (Agassiz, 1829), the “curimba”, belongs to the Prochilodontidae family, performs reproductive migrations, and has a detritivorous feeding habit (Yossa and Araújo-Lima, 1998). The general objective of this study was to evaluate THg concentrations in the gills, muscle, liver, and kidneys of detritivorous and piscivorous fish from two rivers in the Teles Pires River basin and to produce a risk index of methylmercury (MeHg) contamination for the population that consumes these fish species.

## Materials and Methods

### Study area

The samples were collected in the Teles Pires River basin, in the Northern region of the state of Mato Grosso, Brazil. The Teles Pires River basin covers 141,524 km<sup>2</sup> and is located in Southern Amazonia, being one of the rivers that form the Tapajós River basin. Comprising the Amazon and Cerrado domains, the Teles Pires River basin has vegetation characteristic of both biomes as well as transitional areas (Kraeski et al., 2022).

The region of the two sampling points in the Teles Pires River basin has different economic activities (Figure 1) and, as a result, each River is subject to different anthropogenic sources of Hg. One sampling point was the Teles Pires River in the stretch near the municipality of Sinop, where agriculture, livestock farming, and dumping of tannery effluents are common practices (Figures 1E and 1F). More recently, after the collections in this study, the Sinop hydroelectric power plant (HPP) was built, which has been in operation since 2019 (Matos et al., 2020) with an installed capacity of 401.88 MW and a reservoir of 342 km<sup>2</sup>; only 30% of the vegetation in this reservoir was removed (Fearnside, 2019). The other sampling point was the Peixoto River, located in the municipality of Peixoto de Azevedo, a tributary of the Teles Pires River, where gold mining, agriculture (soybeans, corn, and rice), and cattle ranching are the main economic activities (Figures 1C and 1D). In this region, gravitational concentration/amalgamation processing with Hg prevails, and in the period 2009–2018, there was a 232% increase in the area ex-

exploited by gold mining (Almeida, 2019). The discharge of sediment from mining tailings into streams and rivers can vary from 1–2 tons/g of gold produced (Lobo et al., 2017). Peixoto de Azevedo accounts for around 60% of the state of Mato Grosso's gold production and, in 2021, the state produced 14,634 kg of raw gold (ANM, 2021). According to the document “Organized Crime and Illegally Mined Gold in Latin America” (GIATOC, 2016), which refers to illegal gold production, we should add 15% to this amount. The consumption of Hg by small-scale gold mining (characteristic of the Peixoto de Azevedo region) is three times greater than the gold produced, 85% of which is recovered and 15% is lost to the environment (Castilhos and Domingos, 2018).

### Fish collection and biometrics

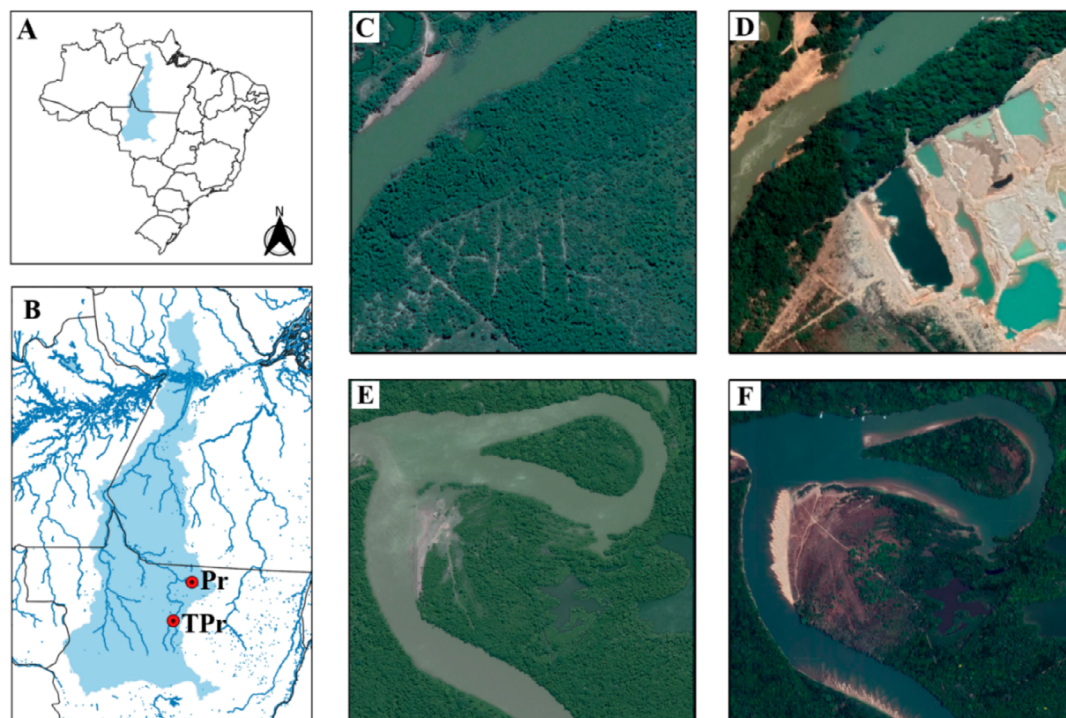
The fish were collected in September 2015 (recession season) and May 2017 (ebb season). They were caught with authorization (n° 18924<sup>1</sup>) from Chico Mendes Institute for Biodiversity Conservation (ICMBio). We used rods with reels and artificial bait to catch the fish. After being caught, the fish were euthanized with Eugenol® in accordance with the laws of the American Veterinary Medical Association (AVMA, 2020), packed in plastic bags, and submerged in ice. We collected 65 fish specimens belonging to four species, one detritivore *P. nigricans* (n=23), and three piscivores: *C. mirianae* (n=20), *H. armatus* (n=10), and *H. tatauaia* (n=12) (Table 1).

Standard length, total weight, and liver weight were measured for each specimen. Sex (macroscopic analysis) and hepatosomatic in-

dex (HSI) were calculated according to Vazzoler (1981). Using stainless steel surgical instruments, the gills, liver, kidneys, and a portion of the dorsolateral muscle were removed from the region above the lateral line of the fish. All these tissues were stored at -20°C until they were analyzed to determine the concentration of THg. Testimonial specimens of all the fish species were deposited in the museum of the *Amazônia National Institute of Research* (INPA, *Instituto Nacional de Pesquisas da Amazônia*)—*C. mirianae* INPA-52812 to 52815—, and in the *Coleção de Peixes do Acervo Biológico da Amazônia Meridional*, ABAM-I *P. nigricans* 702, *H. tatauaia* 709, *H. armatus* 956—of the Federal University of Mato Grosso (UFMT), Sinop Campus.

### Analysis of total mercury

The fish tissues were chemically solubilized as described by Zhou and Wong (2000). The mass of the samples varied according to the size of the organ/tissue (gill/liver 1.0–3.5 g/muscle 1.0–4.5 g/kidneys 0.5–1.5 g). The samples were solubilized with 8 mL of nitric acid: sulphuric acid (HNO<sub>3</sub>:H<sub>2</sub>SO<sub>4</sub>) 2:1 v/v, at room temperature for 3 hours, and then at 60°C for 5 hours. Next, 5 mL of 30% hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>) was added at 65°C and the samples remained at this temperature until they changed from translucent to crystalline (around 1.5 hours). After cooling, the samples were volumized to 25 mL of distilled H<sub>2</sub>O and the THg concentrations were determined using a Cold Vapor Atomic Absorption Spectrometer (Spectrometer model AA 140, Varian) using 10% stannous chloride (SnCl<sub>2</sub>) as a reducing reagent (Akagi and Nishimura, 1991).



**Figure 1** – Map of Brazil highlighting the Tapajós River basin (A). Map of the Tapajós River basin highlighting the rivers sampled: Peixoto River (Pr) and Teles Pires River (TPr) in the Northern region of the state of Mato Grosso (B). Collection area in the Peixoto River (10°12'55.1"S and 54°57'41.1"O) in 2015 (C) and 2023 (D). Collection area in the Teles Pires River (11°44'59.2"S and 55°42'25.1"O) in 2015 (E) and in 2023 (F).

To take the sample readings, a calibration curve was constructed with a standard THg solution (SpecSol brand) of 0, 10, 20, and 30 parts per billion (ppb), and the calibration curve was considered correct when the coefficient of determination ( $r^2$ ) was close to 0.99.

### Quality of analysis

Certified commercial samples and spikes—a term commonly used for fresh fish muscle samples with a known concentration of Hg added—were analyzed to verify the accuracy of the analytical method (Silva et al., 2021). We analyzed the TORT-2 certified sample (lobster hepatopancreas from the National Research Council of Canada) for which the average recovery was 82% ( $n=2$ ; 71 and 92%). The spikes were made from fish muscle samples, which were oven-dried at 40°C until reaching constant weight, macerated in a gral, contaminated with a 10 mg/L THg solution (SpecSol brand), again oven-dried, macerated, weighed, and then subjected to the same digestion process as the other samples. These spike samples ( $n=4$ ) showed an average recovery of 91 standard deviation  $\pm 6\%$ . The results of THg concentrations in the samples in this study represented the analytical results uncorrected for the recoveries observed in the TORT-2 certified samples or in the spikes. The blanks were analyzed periodically to check the effectiveness of the cleaning procedures ( $n=10$ ). Data precision was assessed by reading samples (in duplicate  $n=18$ ), and the average coefficient of variation was  $16.41 \pm 8.82$ , which was considered acceptable in this study. The detection limit, which corresponds to the average concentration of the blanks plus three times the standard deviation of these blanks (Miller and Miller, 1994), was 0.063 µg/L.

### Estimated risk to human health

In general, more than 90% of the Hg in fish muscle is MeHg (Dragan et al., 2023). Therefore, we used the THg concentration value to assess the risk to human health in the consumption of the fish species analyzed, taking as a reference the maximum dose for oral consumption of MeHg recommended by the Food and Agriculture Organization of the United Nations and the World Health Organization (WHO, 2008). The levels of exposure to MeHg in humans resulting from the ingestion of fish muscle, which is the tissue usually consumed, were calculated according to the human health risk assessment determined by the United States Environmental Protection Agency (USEPA, 2000).

We examined the different rates (classes) of fish consumption, ranging from riverine and indigenous people who consume large quantities of fish to those who do not use fish as their main source of protein (class 1: indigenous and riverine people consuming 0.340 kg/day; class 2: regular consumers of 0.142 kg/day; class 3: sporadic consumers of 0.030 kg/day; and class 4: residents from the state of Mato Grosso consuming 0.009 kg/day). For each of these rates, we calculated the average daily intake (IMD) of THg by humans, considering the fish specimens with the lowest and highest concentrations of THg in muscle tissue (amplitude) according to Equation 1:

$$\text{IMD (mg/kg/d)} = (C \cdot \text{TI} \cdot \text{FE} \cdot \text{DE}) / (\text{PC} \cdot \text{TV}) \quad (1)$$

Where:

C = total mercury concentration in the fish muscle (mg/kg wet weight);

**Table 1 – Characteristics and relationship between weight and standard length of fish collected in September 2015 (dry season) and May 2017 (recession season), in the Teles Pires River basin, Brazilian Amazon.**

River	Species	Trophic level	Seasonal season	n	Sex			Standard		Weight (g)		W versus SL	
								length (mm)					
					F	M	I	Average±	Minimum–	Average±	Minimum–	Equation	r²
								SD	maximum	SD	maximum		
Teles Pires River	<i>Cichla mirianae</i> (sedentary)	Piscivore - Tertiary consumer	Dry	11	7	4	0	279±28.5	230–335	601±203.26	265–1015	W=1,273 <sup>06</sup> *SL <sup>3.406</sup>	0.97
	<i>Hydrolycus armatus</i> (migratory)	Piscivore - Secondary consumer	Dry	10	5	5	0	659±87.2	540–830	5415±2625.91	2650–11000	W=5,139 <sup>06</sup> *SL <sup>3.294</sup>	0.95
Peixoto River	<i>Cichla mirianae</i> (sedentary)	Piscivore - Tertiary consumer	Dry	9	5	3	1	273±78.6	160–410	756±639.97	100–2095	W=3,044 <sup>05</sup> *SL <sup>3.006</sup>	0.98
	<i>Hydrolycus tatauaia</i> (migratory)	Piscivore - Secondary consumer	Dry	12	5	7	0	295±25.0	255–340	441±124.13	310–720	W=8,860 <sup>07</sup> *SL <sup>3.512</sup>	0.86
Peixoto + Teles Pires rivers*	<i>Prochilodus nigricans</i> (migratory)	Detritivore - Primary consumer	Dry and recession	23	12	11	0	242±44.9	180–355	516±316.11	190–1500	W=4,366 <sup>05</sup> *SL <sup>2.938</sup>	0.93

\*Since *Prochilodus nigricans* is a migratory species, the results from individuals of this species collected at the two collection points (Teles Pires and Peixoto) were evaluated together and are presented here as “Peixoto + Teles Pires rivers”. The data for *P. nigricans* in each river are shown in Table 3. SD: standard deviation; W: weight; SL: standard length;  $r^2$ : coefficient of determination.



TI = average intake rate: 0.340 kg/day for indigenous and riverine people (Fany, 2011); 0.142 kg/day for adult habitual fish consumers (USEPA, 2000); 0.030 kg/day for adult sporadic consumers (USEPA, 2000); and 0.009 kg/day for consumers in Mato Grosso (IBGE, 2011);

FE = exposure frequency (365 days/year);

DE = lifetime exposure duration (70 years used);

PC = individual's body weight (60 kg used); and

TV = average lifespan (70 years x 365 days/year used).

The risk assessment gives us an idea of how many times above or below the recommended dose the population in question is being exposed to, and this was quantified by calculating the risk index (IR) according to the Equation 2:

$$IR = IMD / RfD \quad (2)$$

Where:

IMD = average daily intake, calculated as described above; and

RfD = reference dose of MeHg orally (mg/kg/day), based on the maximum level of MeHg intake recommended as acceptable for an adult human being with a body weight of 60 kg.

The RfD used in this study was 0.0001 mg/kg/day, which is suggested for sensitive groups such as pregnant and lactating women, babies, and children (WHO, 2008). IR values <1.0 indicate that the population's exposure is less than the maximum recommended dose, and if the IR is  $\geq 1.0$ , the population's exposure is equal to or greater than this dose and therefore, adverse health effects may occur.

## Data analysis

In this study, *C. miriana* and *P. nigricans* were collected at both collection points; *C. miriana* is territorial, which probably reflects local contamination. Thus, after verifying the non-normality of data distribution using the Shapiro-Wilk test, the THg concentrations in this species were compared between sites using the U-test (Wilcoxon/Mann-Whitney). On the other hand, *P. nigricans* is a migratory species. A study analyzing aspects of *P. nigricans* migration in the Napo River basin (Ecuador) pointed out that this species can migrate distances of more than 500 km (Silva and Stewart, 2017). Once in our study, the distance between the two collection points (Teles Pires River and Peixoto River) is approximately 250 km, we consider that the collected specimens of *P. nigricans* possibly belong to the same population, and therefore, the THg concentrations in this species from the two collection points were analyzed together. However, the specimens of *P. nigricans* from the Peixoto River were collected in September 2015 (dry) and the specimens from the Teles Pires River were collected in May 2017 (recession). After verifying the non-normality in the distribution of data through the Shapiro-Wilk test, the THg concentrations in this species were also compared between locations using the U test (Wilcoxon/Mann-Whitney) since changes in THg concentrations in the sediment due to seasonality can affect detritivorous fish.

To verify whether there was bioaccumulation of THg in the fish, we analyzed the correlations between THg concentrations in the muscle and the standard length and total weight of the fish separately for each species and collection site (a method similar to that used by Paiva et al. (2022), as shown in Figure 2). Standard length and total weight were employed because they are measures that correspond to the age of the fish. We verified non-normality in the distribution of data using the Shapiro-Wilk test.

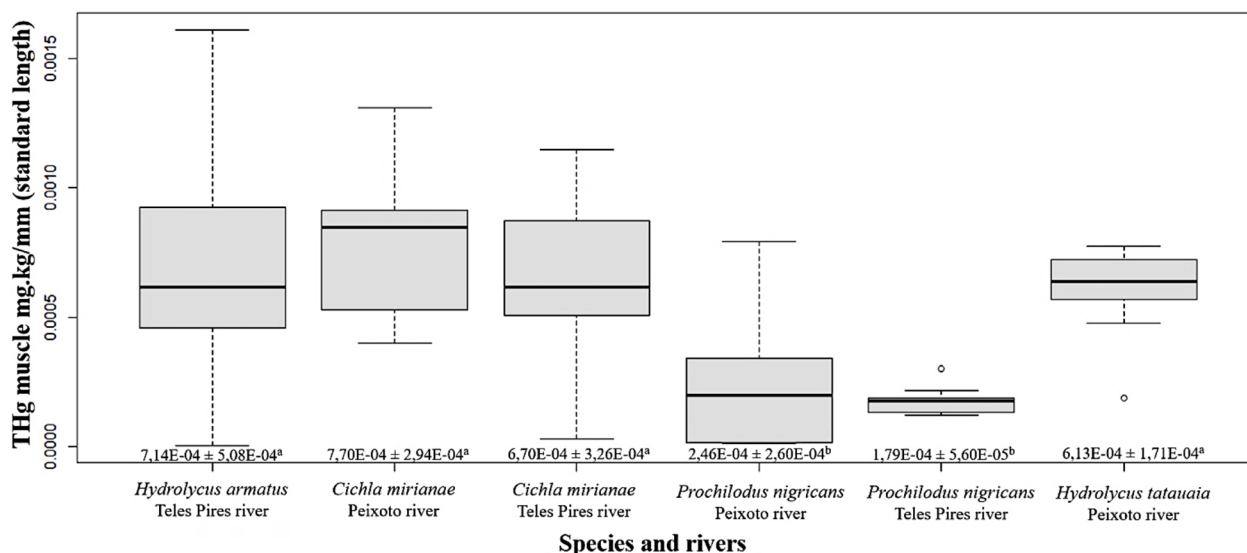


Figure 2 – Total mercury standardized by standard length (mm) (mean ± standard deviation) in fish species collected in September 2015 (dry season) and May 2017 (recession season), at two points in the Teles Pires River basin, Brazilian Amazon. Differences between species: equal letters (a, b, c, d) indicate no statistical difference, while different letters indicate statistical difference (Kruskal-Wallis/Dunn test; 5% significance level).

Thus, the nonparametric Kruskal-Wallis test was used to examine the differences in THg concentration between the tissues of the fish. Spearman's correlation was applied to verify whether there was a correlation between THg concentrations in the liver of the fish of each species and the HSI. Considering that total weight and standard length are positively correlated (Table 1), only standard length was used to correlate size with THg concentration in the muscle. All analyses were performed using R Statistical Software v. 1.0. 4.4.2 (R Core Team (2024), <https://cran.r-c3sl.ufpr.br/>).

## Results

A total of 65 fish samples belonging to the four species were collected (Table 1). The *C. miriana* specimens collected from both locations had similar sizes (Table 2). The *C. miriana* specimens from the Teles Pires River presented THg concentrations in the tissues, similar to those from the Peixoto River, with the exception of the kidneys (Table 2). The *P. nigricans* specimens from the Teles Pires River (recession season/2017) were larger than those from the Peixoto River (dry/2015), but presented similar THg concentrations in the tissues, except for the kidneys (Table 3). In general, THg concentrations were significantly higher in the liver than in the other tissues evaluated, and the gills presented the lowest concentrations (Table 4). THg concentrations in muscle, which is the part generally consumed, are below the safe values ( $0.50 \text{ mg.kg}^{-1}$ ) for consumption recommended by the WHO (2008), except in two specimens of *H. armatus* from the Teles Pires River (Table 4). Comparing THg concentrations in the liver, we observed that they were significantly higher in *P. nigricans* and *H. armatus* (Table 4). THg concentrations in the muscle were significantly higher in piscivorous species, especially in *C. miriana* (both rivers) which belong to the top of the chain (4th trophic level, tertiary consumer) (Table 4; Figure 2).

The HSI ranged from 0.60 in *C. miriana* in the Teles Pires River to 0.36 in *P. nigricans* in the Teles Pires River basin (Table 5). Only the detritivorous species *P. nigricans* showed a correlation between THg concentrations in the liver and the HSI (Table 5). In all species and locations, the THg concentration in the muscle of fish was not related to their total weight, but showed a strong relationship with the standard length of the individuals (Table 5).

The IR values for the consumption of the fish species analyzed in this study are presented in Table 6, according to the range of MeHg concentrations estimated for the four different fish consumption rates. Table 6 shows the IR values based on the oral reference dose of  $0.0001 \text{ mg.kg.day}$  of MeHg indicated for the sensitive groups (lactating women, infants, and children). The results of this study show the IR range  $>1$  (for all species) for consumers in class 1 and class 2. For consumers in class 3, the IR range  $<1$  only for the detritivore species *P. nigricans*. For consumers in class 4, the IR range  $>1$  only for the piscivore species *H. armatus*.

## Discussion

### Mercury in tissues

This study is the first record of Hg analysis in different tissues of fish from the Teles Pires River that addresses different trophic levels before the formation of the Sinop HPP reservoir, which belongs to a hydroelectric complex of four plants in a cascade system. The use of different fish tissues to monitor Hg in water bodies helps to assess the state of preservation and the natural biogeochemical characteristics of aquatic ecosystems (Adam, 2002). Considering that the liver is an important organ for the purification of contaminants from organisms, we hypothesized that the liver of fish would present the highest concentrations of THg when compared to other tissues. Corroborating our hypothesis, THg concentrations were generally significantly higher in the liver and kidneys, except for *H. tatauaia* (Peixoto River), which presented similar concentrations in the liver and muscle. No significant relationship was found between the concentration of THg in the liver and the HSI for piscivores collected in the Teles Pires River basin, suggesting that Hg in the organism would not be causing liver changes.

Fish can absorb trace metals from the ingestion of food and non-food particles, and from diffusion through the gills and skin (Zaynab et al., 2022). Once absorbed by the body, Hg enters the bloodstream and can be transported to different tissues in the body, transformed by the liver, or eliminated through the biliary system or kidneys (Zaynab et al., 2022). High concentrations of THg in the liver or kidneys of fish were observed in freshwater fish in Russia (Moiseenko and Gashkina, 2020);

**Table 2 – Comparisons of standard length, weight, and total mercury concentrations of *Cichla miriana* specimens collected in September 2015 (dry season) at two locations in the Teles Pires River basin, Brazilian Amazon.**

Rivers	n	Standard length (mm)		Weight (g)		Total mercury (ppb wet weight) (mean±SD)			
		Average±	Minimum	Average±	Minimum	Gills	Liver	Muscle	Kidneys
		SD	maximum	SD	maximum				
Teles Pires River	11	279±28.50	230–335	601±203.26	265–1015	90±50 <sup>a</sup>	470±290 <sup>a</sup>	170±110 <sup>a</sup>	490±570 <sup>a</sup>
Peixoto River	9	273±78.6	160–410	756±639.97	100–2095	50±20 <sup>a</sup>	460±260 <sup>a</sup>	210±90 <sup>a</sup>	190±110 <sup>b</sup>
		t=0.205; p=0.841		t=-0.692; p=0.505		p=0.09; W=72	p=1; W=49	p=0.66; W=39	p=0.009; W=83

Differences between locations: the same letters within the same column (a, b, c, d) indicate no statistical difference, while different letters indicate statistical difference (Wilcoxon/Mann-Whitney; 5% significance level). W: Mann-Whitney test result; ppb: parts per billion; SD: standard deviation; t: test value; p: p-value.

in the carnivorous *Plagioscion squamosissimus* in the Madeira River basin in Brazil (Costa et al., 2022); in the *Danio rerio* in Portugal (Vieira et al., 2021); and in carnivorous fish during the flood season in Western Amazonia (Silva et al., 2019). The gills are also extremely irrigated by blood, like liver and kidneys; however, they do not have a detoxification function like the liver and kidneys. In the present study, the gills of all species presented lower THg concentrations in all environments evaluated.

Also in the present study, the detritivorous species *P. nigricans* presented the lowest Hg concentrations in the muscle and higher concentrations in the liver compared to the piscivorous species, besides a marginally significant correlation between THg concentrations in the liver and the HSI ( $r=-42$ ;  $p=0.04$ ). The higher concentrations in the liver probably occur due to the fact that detritivorous fish feed on sediment, a site of major Hg deposition, and they can thus accumulate large concentrations of Hg, sometimes even higher than carnivorous fish (Weber et al., 2013). The liver concentrates the Hg absorbed by the digestive tract and has a high capacity for bioaccumulation; in the enterohepatic circulation, Hg can be eliminated through bile, reabsorbed in the gastrointestinal tract, reaching the liver again, thus explaining high Hg concentrations in this organ (De Marco et al., 2023). Low Hg concentrations in the muscle are expected for detritivorous fish (Silva et al., 2019; Silva and Lima, 2020), as was the case in the present study.

The THg levels presented in Table 4 for *P. nigricans* were analyzed together (Peixoto+Teles Pires rivers) since they are migratory and probably belong to the same population (but they were also presented separately in Table 3). In the Peixoto River, *P. nigricans* specimens were collected in September 2015 (dry season), and in the Teles Pires River, in May 2017 (dry season) as seen in Table 3. During the rainy season, there is an increase in surface erosion and the transport of Hg associated with fine soil particles to aquatic ecosystems (Roulet et al., 1998). Hg has a high affinity for suspended particles, accumulating in the sediment of water bodies, where Hg methylation processes occur (Guimarães, 2020). During the flood season, there is greater access to MeHg from the environment and available food items, and the recession season reflects this environmental condition (Silva et al., 2019).

**Table 3 – Comparisons of standard length, weight, and total mercury concentrations of *Prochilodus nigricans* specimens collected in September 2015 (dry season) and May 2017 (recession season) at two sites in the Teles Pires River basin, Brazilian Amazon.**

Rivers	Seasonal season	n	Standard length (mm)		Weight (g)		Total mercury (mg.kg <sup>-1</sup> wet weight) (mean±SD)			
			Average±	Minimum–	Average±	Minimum–	Gills	Liver	Muscle	Kidneys
			SD	maximum	SD	maximum				
Teles Pires River	Recession	9	290±27.30	270–355	813±301.0	480–1500	0.006±0.004 <sup>a</sup>	0.731±0.594 <sup>a</sup>	0.051±0.013 <sup>a</sup>	0.085±0.029 <sup>b</sup>
Peixoto River	Dry	14	211±19.47	180–250	326±116.1	190–560	0.031±0.055 <sup>a</sup>	1.234±1.417 <sup>a</sup>	0.050±0.053 <sup>a</sup>	0.275±0.309 <sup>a</sup>
			t=7.443; p<0.001		t=4.637; p=0.001		p=0.540; W=50	p=0.169; W=38	p=0.76; W=61	p=0.001; W=11

Differences between sites: the same letters within the same column (a, b, c, d) indicate no statistical difference, while different letters indicate statistical difference (Wilcoxon/Mann-Whitney; 5% significance level). W: Mann-Whitney test result; ppb: parts per billion; SD: standard deviation; t: test value; p: p-value.

Detritivorous fish, such as *P. nigricans*, can accumulate high concentrations of Hg when feeding on sediments rich in MeHg (Weber et al., 2013). However, THg concentrations in the tissues of *P. nigricans* collected during the recession season (Teles Pires River) showed lower levels in the kidneys than those collected during the dry season (Peixoto River) ( $W=11$ ; kidneys  $p=0.001$ , Wilcoxon/Mann-Whitney). In fish, the kidneys and liver are the main organs responsible for the elimination and detoxification of Hg (Zaynab et al., 2022). A study with a detritivorous fish in a lake polluted with Hg found the highest concentrations of MeHg and inorganic Hg in the kidneys, suggesting preferential accumulation in this organ before its excretion through the urinary tract (Jackson, 2018). In the kidneys, the highest concentration is usually inorganic Hg, since MeHg would be eliminated more easily. This excretion mechanism could involve the systematic elimination of Hg species (Jackson, 2018). In the present study, the recession and the changes imposed by the rainy season on Hg dynamics did not cause a pronounced increase in THg levels in the kidneys and liver of *P. nigricans* collected in the Teles Pires River. Thus, although *P. nigricans* belongs to a low trophic level (detritivore), it is probably directly exposed to high Hg loads in the sediment in the Peixoto River region.

**Table 4 – Total mercury concentrations in specimens collected in September 2015 (dry season) and May 2017 (recession season), in the Teles Pires River basin, Brazilian Amazon.**

Rivers and species	Total Mercury (ppb wet weight) (mean±standard deviation)			
	Gills	Liver	Muscle	Kidneys
<b>Teles Pires River</b>				
<i>Cichla mirianae</i>	90±50 <sup>c</sup>	470±290 <sup>a,y</sup>	170±110 <sup>b,x</sup>	490±570 <sup>a</sup>
<i>Hydrolycus armatus</i>	60±40 <sup>b</sup>	810±660 <sup>a,x,y</sup>	300±320 <sup>b,x</sup>	310±290 <sup>ab</sup>
<b>Peixoto River</b>				
<i>Cichla mirianae</i>	50±20 <sup>c</sup>	460±260 <sup>a,y</sup>	210±90 <sup>b,x</sup>	190±110 <sup>b</sup>
<i>Hydrolycus tatauaia</i>	60±30 <sup>c</sup>	170±30 <sup>a,z</sup>	180±50 <sup>a,x</sup>	120±50 <sup>b</sup>
<b>Peixoto+Teles Pires rivers</b>				
<i>Prochilodus nigricans</i>	20±47 <sup>d</sup>	1050±1240 <sup>a,x</sup>	50±40 <sup>c,y</sup>	200±270 <sup>b</sup>

Differences between tissues of the same species/location: equal letters within the same row (a, b, c, d) indicate no statistical difference, while different letters indicate statistical difference. Differences between liver and muscle of the same species/locations: equal letters within the same column (x, y, z) indicate no statistical difference, while different letters indicate statistical difference.

**Table 5 – Mean hepatosomatic index, r and p-values of Spearman correlation (5% significance level) between liver total mercury concentration and hepatosomatic index, and p and r<sup>2</sup> values of multiple linear regression of standard weight and length and total mercury in muscle of specimens collected in September 2015 (dry season) and May 2017 (recession season), in the Teles Pires River basin, Brazilian Amazon.**

Rivers and species	HSI	THg-liver versus HSI		Multiple linear regression $\ln(\log(\text{weight})\sim\log(\text{standard length}))+\text{THg concentration in muscle}$		
		R	p	p (weight)	p (standard length)	r <sup>2</sup>
Teles Pires River						
<i>Cichla miriana</i> e	0.60	0.25	0.45	0.85	<0.0001	0.96
<i>Hydrolycus armatus</i>	0.33	0.03	0.94	0.28	<0.0001	0.96
Peixoto River						
<i>Cichla miriana</i> e	0.59	0.13	0.74	0.84	<0.0001	0.98
<i>Hydrolycus tatauaia</i>	0.37	-0.05	0.87	0.95	<0.0001	0.82
Peixoto + Teles Pires rivers						
<i>Prochilodus nigricans</i>	0.36	-0.42	0.04	0.94	<0.0001	0.96

HIS: hepatosomatic index; THg: total mercury; r<sup>2</sup>: coefficient of determination; p: p-value (of Spearman correlation; 5% significance level).

**Table 6 – Range of total mercury concentrations and risk index of adverse health effects calculated based on four different fish consumption rates of specimens collected in September 2015 (dry season) and May 2017 (recession season), in the Teles Pires River basin, Brazilian Amazon.**

Species	THg	Risk Index				Oral RfD MeHg
	Muscle [ ] mg.kg	Class 1 0.340 (kg/day)	Class 2 0.142 (kg/day)	Class 3 0.03 (kg/day)	Class 4 0.009 (kg/day)	(mg/kg/d)
<i>Cichla miriana</i> (Teles Pires River)	0.008–0.368	0.459–20.904	0.192–8.731	0.041–1.845	0.012–0.553	0.0001
<i>Hydrolycus armatus</i> (Teles Pires River)	0.002–0.979	0.155–53.816	0.065–22.476	0.014–4.749	0.004–1.425	0.0001
<i>Cichla miriana</i> (Peixoto River)	0.064–0.340	3.632–19.295	1.517–8.059	0.321–1.703	0.096–0.511	0.0001
<i>Hydrolycus tatauaia</i> (Peixoto River)	0.054–0.255	3.094–14.473	1.292–6.044	0.273–1.277	0.082–0.383	0.0001
<i>Prochilodus nigricans</i> (Peixoto+Teles Pires rivers)	0.002–0.198	0.130–11.231	0.054–4.691	0.012–0.321	0.003–0.297	0.0001

THg: total mercury; RfD: oral reference dose (based on WHO, 2008); MeHg: methylmercury; Class 1: indigenous and riverine; Class 2: regular consumers; Class 3: sporadic consumers; Class 4: consumers from the state of Mato Grosso.

We found a strong positive relationship between THg concentration in fish muscle and the standard length of the individuals analyzed in the present work. In the Amazon basin, several studies have demonstrated the same relationship for detritivorous and piscivorous fish (Silva and Lima, 2020). Hg bioaccumulation is controlled by the absorption, biotransformation, and excretion of the element, which are closely dependent on the metabolic rate of organisms (McCoy et al., 1995). However, due to Hg bioaccumulation, larger fish tend to have higher concentrations accumulated throughout life than smaller fish, a fact found in the present study.

Based on the processes of bioaccumulation and trophic biomagnification, it is expected to find higher concentrations of Hg in the muscle of piscivorous fish than in detritivores (Moiseenko and Gashkina, 2020). This pattern was also found in the present study for piscivorous species (*H. armatus*, *H. tatauaia*, and *C. miriana*), presenting higher concentrations of THg in the muscle than in the detritivorous species

(*P. nigricans*). There are studies in the Amazon basin that present higher concentrations of THg in the muscle of fish than the present study: *Hydrolycus scomberoides* (Basta et al., 2023); *Cichla* sp. (Nyholt et al., 2022; Basta et al., 2023); and *P. nigricans* (Azevedo et al., 2019; Nyholt et al., 2022; Basta et al., 2023) (Table 7). There are also studies with lower concentrations for the detritivorous species *P. nigricans* (Silva and Lima, 2020) and piscivorous *Cichla monoculus* (Hacon et al., 2020) (Table 7). These variations in Hg concentrations in the ichthyofauna can be explained by seasonal fish migrations, dietary variations that are not only temporal but also spatial (Tesser et al., 2021), interactions between guilds and seasonality (Azevedo et al., 2020), and flood pulse (Azevedo et al., 2019). The dynamics of the Tapajós basin, due to the seasonal flooding process, in addition to causing changes in fish feeding, can bring a large supply of nutrients and organic matter to the riverbed and facilitate the development of favorable conditions for Hg methylation (Guimarães, 2020; Pereira et al., 2021).



Table 7 – Total mercury data in the muscle of detritivorous and piscivorous fish from the Brazilian Amazon basin.

Species	Location	THg (mg.kg <sup>-1</sup> )	Reference
<i>Prochilodus nigricans</i>	Teles Pires River basin	0.05±0.04	present study
<i>Cichla miriana</i>	Teles Pires River	0.17±0.11	present study
<i>Hydrolycus armatus</i>	Teles Pires River	0.30±0.32	present study
<i>Cichla miriana</i>	Peixoto River	0.21±0.09	present study
<i>Hydrolycus tatauaia</i>	Peixoto River	0.18±0.05	present study
<i>Prochilodus nigricans</i>	Madeira River basin	0.24±0.07	Azevedo et al. (2019)
<i>Prochilodus nigricans</i>	Madeira River basin	<0.50	Azevedo et al. (2020)
<i>Cichla monoculus</i>	Cassiporé River	0.19	Hacon et al. (2020)
<i>Prochilodus nigricans</i>	Solimões River	0.05	Silva and Lima (2020)
<i>Hydrolycus armatus</i>	Teles Pires River	0.23	Matos et al. (2021)
<i>Cichla</i> spp	Juruá River	~4.00	Nyholt et al. (2022)
<i>Prochilodus nigricans</i>	Juruá River	~0.50	Nyholt et al. (2022)
<i>Prochilodus nigricans</i>	Amazon basin	0.09	Basta et al. (2023)
<i>Cichla</i> spp	Amazon basin	0.21	Basta et al. (2023)
<i>Cichla</i> spp	Amazon basin	0.84	Basta et al. (2023)
<i>Hydrolycus scomberoides</i>	Amazon basin	0.97	Basta et al. (2023)
<i>Prochilodus nigricans</i>	Amazon basin	0.04	Basta et al. (2023)

THg: total mercury; ±: standard deviation.

Considering that plants are an important sink for atmospheric Hg (Zhou and Obrist, 2021), it is possible that natural accumulation in leaf litter over time could be a significant source of Hg for Amazonian soils (Nyholt et al., 2022). The main anthropogenic sources of Hg in the Amazon are: small-scale artisanal gold mining, deforestation and biomass burning, and hydropower (Crespo-Lopez et al., 2021); silviculture (forest management) that causes disturbance and erosion of soils and sediments (Feinberg et al., 2024); and currently, small hydropower plants and aquaculture that promote the synergism of impacts (Freitas et al., 2022). Given that this seasonal flooding process in the Teles Pires River basin is being drastically altered by the activity of four hydroelectric plants in a cascade system, Hg methylation and bioaccumulation in fish may be enhanced (Lacerda and Malm, 2008), similar to what occurred in the Tucuruí reservoir (Fearnside, 2015). The results of this study provide a database from before the Sinop HPP flooding, with which future studies can be compared to establish rates of increase in Hg bioaccumulation after the Teles Pires River damming.

The Tapajós River basin (in which the Teles Pires River basin is located) is increasingly impacted by the construction of hydroelectric dams (Lees et al., 2016). Some have already been built, and others are planned for the region (Aguiar Cavalcante et al., 2021). We know that the construction of dams directly leads to biota contamination in hydroelectric dams, both in the lake upstream or downstream of the dam (Kasper et al., 2014; Aguiar-Santos et al., 2022; Willacker et al., 2023; Oliveira et al., 2025). Therefore, obtaining information on the concentration in fish of different trophic guilds is a snapshot in time,

which can be very important for studies of the impact of these projects (e.g., study conducted in the Teles Pires basin by Oliveira et al., 2025). Without this past scenario, it becomes very difficult, and sometimes even impossible, to assess the impact of a hydroelectric plant on the bioaccumulation of Hg. Pestana et al. (2019) and De Castro Paiva et al. (2024) addressed this problem, evaluating the impact of hydroelectric plants, and all agreed that having the scenario prior to construction is the ideal way to assess it.

#### Estimation of risk to human health

In the present study, THg concentrations in fish muscle are below the limit recommended by the WHO (2008) for human consumption, except for two specimens of *H. armatus* (0.61 and 0.95 THg mg.kg<sup>-1</sup>). Among the four classes of the consumers simulated, the IR<1 for adult sporadic consumers (30 g.day) only for the detritivorous species *P. nigricans*, and for consumers in the state of Mato Grosso (9 g.day) for all species except for the carnivore *H. armatus*. For indigenous and riverine populations (342 g.d) and adult habitual consumers (142 g.day), all species presented IR>1, indicating that adverse health effects may occur. Three indigenous ethnic groups (Mundurukus, Kayabis, and Apiakás) in the Teles Pires River basin consume fish in at least seven meals per week (Fany, 2011). In this basin, there are also three colonies of professional artisanal fishermen (Z-16 of Sinop and region; Z-21 of Vale do Peixoto, and Z-4 of Nobres and region) with around 400 registered fishermen, whose consumption and sale of fish are the main sources of food and income (Matos et al., 2020).

The limit used in this study ( $0.50 \text{ mg.kg}^{-1}$ ) was calculated based on WHO recommendation for a person weighing approximately 60 kg, who ingests approximately 250 g of fish per week. This amounts to approximately an intake of 0.3 g of Hg per kg of body weight per day. Considering a consumption of 340 g/day (estimated for indigenous and riverine populations; Fréry et al. (2001)), a maximum intake of 0.1 g of MeHg/kg of body weight/day (considering sensitive groups; WHO, 2008) and a person weighing 60 kg, the fish intake could reach a maximum of  $0.02 \text{ mg.kg}^{-1}$ . Of the samples evaluated, only 33% of *P. nigricans*, 10% of *C. mirianae*, and 18% of *H. tatauaia* specimens were within this MeHg consumption limit for sensitive groups. The species with the lowest risk of THg contamination is *P. nigricans*, as it presented the lowest concentrations in the muscle. The good news is that, in a survey conducted in the Amazon basin among 517 interviewees, *P. nigricans* was identified as the most consumed fish (Begossi et al., 2019).

Just as important as the concentration of THg in fish is the amount of fish consumed by the population. Due to the high rates of fish consumption by indigenous people and riverine populations, a risk assessment based solely on the concentration of THg found in fish muscles may underestimate the actual amount of THg ingested through the diet and the health risks caused by chronic exposure. The intake of THg above the limit determined by the WHO by women of childbearing age, pregnant women, or planning a pregnancy, infants, children, and malnourished people is a public health problem, as it can affect the development of the communities involved (Silva and Lima, 2020; Vasconcellos et al., 2022).

## Conclusion

We conclude that the THg concentrations evaluated in this study probably did not interfere with the growth and health of the fish. The gills do not have a detoxification function like the liver and kid-

neys, thus, they presented lower THg concentrations. Almost all individuals had THg concentrations in the muscle below the level recommended for human consumption by the WHO, but considering that the region has a high consumption of fish, the daily intake of Hg exceeds the limit. The detritivorous fish *P. nigricans* presented the lowest THg concentrations in the muscle; therefore, this can be preferentially consumed by the general population and especially by sensitive groups, such as lactating women, infants, and children. Considering that the fish analyzed in this research were collected before the formation of the lake of the Sinop HPP reservoir, the results presented here can serve as a database for monitoring the THg concentration in the ichthyofauna of the Teles Pires River.

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## Authors' contributions:

**Matos, L. S.:** data curation, formal analysis, project administration, writing – original draft, writing – review & editing. **Kasper, D.:** formal analysis, writing – review & editing. **Silva, J. O. S.:** conceptualization, data curation, formal analysis, writing – original draft, writing – review & editing. **Carvalho, L.N.:** funding, acquisition, supervision, writing – original draft, writing – review & editing.

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