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Metal concentrations (Fe and Zn) in *Macrobrachium amazonicum* from Brazilian Amazon rivers

Concentrações de metais (Fe e Zn) em *Macrobrachium amazonicum* de rios da Amazônia brasileira

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ABSTRACT

The objective of this work was to analyze the contamination by iron (Fe) and zinc (Zn) in the shrimp Macrobrachcium amazonicum in the region of the mouth of the Amazon River in the State of Amapá, taking into account aquaculture and fishing activities. 90 shrimp were used, 45 females and 45 males of *M. amazonicum*. The animals (30) were found in three different points, being Mazagão Velho in Mazagão, a community on Ilha de Santana, a district in the Brazilian municipality of Santana and Arquipélago do Bailique. Fe and Zn determinations were performed by conventional flame atomic absorption spectrophotometry. The greatest accumulation of Zn in the organs and the lowest concentration of Zn in the muscle and exoskeleton of M. amazonicum was observed. Fe had the highest accumulation in organs, followed by exoskeleton and muscle. Zn accumulation in male organs was greater than in muscle and exoskeleton while Fe concentrations were greater in organs, followed by exoskeleton and muscle. In females, Zn accumulation was greater in our organs, with no difference between muscle and exoskeleton. According to the present study, to maintain levels of food security, the consumption of freshwater shrimp caught and farmed in Mazagão, Bailique Archipelago and community of Ilha de Santana should be limited to a daily shrimp intake dose of 96. 52 g for Fe and 226.81 g for Zn.

Keywords: aquaculture, amazon shrimp, minerals, fishing, food security.

RESUMO

O objetivo desse trabalho foi analisar a contaminação por ferro (Fe) e zinco (Zn) no camarão *Macrobrachcium amazonicum* na região da foz do Rio Amazonas no Estado do Amapá, tendo em vista as atividades de aquicultura e pesca. Foram utilizados 90 camarões, sendo 45 fêmeas e 45 machos de *M. amazonicum*. Os animais (30) foram coletados em três pontos diferentes, sendo Mazagão Velho em Mazagão, comunidade da Ilha de Santana um distrito do município brasileiro de Santana e Arquipélago do Bailique. As determinações de Fe e Zn foram realizadas por espectrofotometria de absorção



atômica de chama convencional. Foi observado o maior acúmulo de Zn nos órgãos e menor concentrações de Zn no músculo e no exoesqueleto de *M. amazonicum*. O Fe teve maior acúmulo nos órgãos seguido de exoesqueleto e músculo. A acumulação de Zn nos órgãos dos machos foi maior do que no músculo e exoesqueleto enquanto as concentrações de Fe foram maiores nos órgãos, seguido do exoesqueleto e músculo. Nas fêmeas, a acumulação de Zn foi maior nos órgãos, não havendo diferença entre músculo e exoesqueleto. Conforme o presente estudo, para manter níveis de segurança alimentar, o consumo de camarões de água doce capturados e cultivados em Mazagão, Arquipélago do Bailique e comunidade de Ilha de Santana devem ser limitados a uma dose diária de ingestão de camarão de 96,52 g para Fe e de 226,81 g para Zn.

Palavras-chave: aquicultura, camarão-da-amazônia, minerais, pesca, segurança alimentar.

1 INTRODUCTION

Shrimp is highly appreciated worldwide, it has a high protein content and a low percentage of fat (Duray et al., 2022; Golder et al., 2022; Liu et al., 2022; Tanjung et al., 2022). The health, safety, and overall quality of shrimp cultivation profoundly hinge on various factors, including water quality, feed composition, and the mineral content in shrimp (Kawan et al., 2019; Amir et al., 2021; Bull et al., 2021; Vieira et al., 2021).

When cultivating shrimp, it is important to take into account the ability of crustaceans to accumulate both non-essential metals such as cadmium (Cd) and lead (Pb) and essential minerals like iron (Fe) and zinc (Zn). This accumulation is an integral part of the homeostatic processes within shrimp (Silva et al., 2016; Mostafiz et al., 2020; Arisekar et al., 2022). Essential minerals are vital for the metabolic functions of shrimp, ensuring their overall well-being (Maia et al., 2022).

Of these essential minerals, zinc (Zn) plays a multifaceted role in the growth, reproductive processes, protein synthesis, energy production, and gene regulation in shrimp (Truong et al., 2020; Yuan et al., 2020; Hassan et al., 2021). Furthermore, zinc is implicated in various critical functions, including antioxidant properties and immune responses, by catalyzing numerous enzyme-mediated metabolic processes (Muralisankar et al., 2014; Yuan et al., 2020; Shi et al., 2021).

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Iron stands as a vital element for all living organisms. Shrimp, in particular, require this mineral for numerous essential functions such as bolstering innate immunity against pathogens, supporting growth, enabling reproduction, conferring resistance to stress factors, and participating in crucial biological processes like electron transport and DNA synthesis (Hsu et al., 1999; Mostafiz et al., 2020; Tang et al., 2020). However, the presence of inorganic forms of Zn and Fe in shrimp diets can introduce contamination to the aquaculture environment and its surroundings, with the potential to become toxic to aquatic organisms, especially when present in high concentrations (Frías-Espericueta et al., 2003; Wu et al., 2011; Albuquerque et al., 2020; Jiao et al., 2022).

Shrimp farming significantly contributes to socioeconomic development. According to the Food and Agriculture Organization of the United Nations (FAO, 2022), the global production of species belonging to the *Macrobrachium* genus increased by approximately 22% between 2006 and 2016, generating profits totaling approximately US\$4.2 million, based on the production of 506.6 thousand tons (Azad et al., 2021; Ray et al., 2021; Golder et al., 2022). In Brazil and globally, the most cultivated species of freshwater shrimp is the exotic *Macrobrachium rosenbergii*. Nonetheless, the cultivation of this species raises concerns about sustainability, as it introduces an exotic organism that may potentially harm the biodiversity of the native Brazilian aquatic fauna (Silva et al., 2017; Lopes et al., 2020; Tan et al., 2022).

Macrobrachium amazonicum, on the other hand, is a native species that exhibits favorable attributes for captive production. These include high fecundity, a relatively short production cycle, the capacity to endure high densities (up to 80 shrimp per square meter), robustness, and meat that is both firmer and more flavorful than the exotic *M. rosenbergii* (Araújo et al., 2017; Taddei et al., 2017; Heldt et al., 2019; Brazão et al., 2022; Perroca et al., 2022). *M. amazonicum* enjoys a widespread distribution in the lower Amazon River regions of Amapá and Pará and does not pose a risk of ecosystem invasion, being native to these areas (Bentes et al., 2011; Da Silva et al., 2016).

However, in the state of Amapá, mining is one of the leading contributors to the local economy. Unfortunately, the waste generated from mineral extraction is often discharged directly into the region's rivers and streams without any form of treatment.



This, in turn, causes soil and water pollution, detrimental to the health of shrimp and the well-being of humans who consume them as a food source (Lima et al., 2015).

The locations selected for the collection of *M. amazonicum* specimens are strategically placed to represent different facets of environmental influence. Mazagão Velho, situated within the Mazagão municipality, is a rural area historically tied to mining activities since the 1990s (Souza et al., 2019). The community of Ilha de Santana, located in the port area, is significantly impacted by mining operations, serving as a disposal site for mining byproducts (Azevedo et al., 2019). The Bailique Archipelago, found at the mouth of the Amazon River, is exposed to the coastal environment, which includes environmental degradation and pollution stemming from mining waste (Guabiraba et al., 2016).

Given the contextual backdrop and considerations mentioned above, the primary objective of this study is to assess the contamination levels of Fe and Zn in the freshwater shrimp *M. amazonicum* within the region of the mouth of the Amazon River in the State of Amapá. This assessment is conducted carefully regarding aquaculture and fishing activities, highlighting the significance of monitoring the mineral contents in these shrimps.

2 METHODS

2.1 SAMPLE COLLECTION

The study involved a total of 90 *M. amazonicum* shrimp. This sample consisted of 30 shrimp from each of the following three distinct locations:

1. Mazagão: Situated in a rural and sparsely populated region, Mazagão served as the first collection site. The geographical coordinates for this location are approximately 0°13'33.99" S latitude and 51°25'58.05 "W longitude.

2. Santana Island: This collection site is located in the port region, characterized by higher levels of human activity. The coordinates for Santana Island are approximately $0^{\circ}4'9.03''$ S latitude and $51^{\circ}9'51.66''$ W longitude.



3. Bailique Archipelago: Positioned at the mouth of the Amazon River, the Bailique Archipelago is close to the coastal region and influenced by the surrounding saline environment. The coordinates for this location are approximately 0°52'56.66" N latitude and 50°6'22.22" W longitude.

Figure 1. The collection sites for *M. amazonicum* are visually represented in Figure 1, with corresponding geographical coordinates provided for each location.



Source: Adapted from Google Earth (2023).

Shrimp collection was conducted using a trap device known as a "matapi". Following collection, the captured shrimp specimens were placed in an appropriately labeled container. To ensure preservation and maintain their freshness, the shrimp specimens were stored on ice. Subsequently, the preserved shrimp specimens were transferred to the aquaculture laboratory at Embrapa-AP. In the laboratory, the shrimp specimens underwent a series of processes including weighing, sex determination, and measurement of their length.

2.2 SAMPLE PREPARATION

Once collected, the shrimp specimens were subjected to a systematic sample preparation process as follows:



1. The shrimp bodies were dissected into distinct parts, categorizing them into muscle, organs (including gonads, heart, hepatopancreas, and gills), and exos-keleton.

2. Each of these components resulted in a total of 45 samples per gender and collection point, amounting to 135 samples in total.

3. Each part of the shrimp body was meticulously weighed using a precision scale and then stored in 15 mL Falcon tubes. These tubes were subsequently placed in a conventional freezer for 24 hours at -18°C.

4. After the freezing period, the samples were dried through freeze-drying for a duration of 10 h. This freeze-drying process was facilitated by utilizing a Terroni benchtop freeze-dryer, specifically the Interprise I model.

5. Following the freeze-drying step, the samples were weighed once more to determine their moisture content. Subsequently, these dried samples were crushed in an analytical mill, specifically the KIA analytical mill, until they achieved a homogeneous powdered form.

2.3 SAMPLE DIGESTION

The following digestion process was implemented for the samples based on the methodology of Souza et al. (2005):

1. The freeze-dried components of the shrimp were placed into a digestion tube. This was followed by the addition of 4.0 mL of concentrated HNO_3 (70%) to each sample.

2. The samples were left to rest for a full night within the digestion tubes before being placed in a digester block the following morning.

3. The digester block was gradually heated until it reached a temperature of 120°C, maintaining this temperature for a duration of 1 h.

4. After the designated heating period, the tubes were extracted, and, once cooled, 4.0 mL of H_2O_2 (30%) were introduced to the samples.

5. The samples were then reintroduced to the digester block and subjected to further heating at 120°C until the solution turned colorless.



6. After cooling and acclimatization to room temperature, the samples were transferred to Falcon tubes. To ensure consistency in volume, distilled water was added to each sample until reaching a total volume of 10 mL.



2.4 METAL DETERMINATIONS

Concentrations of Fe and Zn in the shrimp samples were determined using conventional flame atomic absorption spectrophotometry. Specifically, a Thermo Scientific atomic absorption spectrophotometer, model ICE3300, was employed.

1. The equipment was configured to read the metals of interest, and calibration was performed using calibration curves generated from commercial standard solutions. This included an Iron standard solution (1000 mg/L) and a Standard Zinc Solution (1000 mg/L).

2. The three distinct parts of the shrimp's body, namely muscles, organs (including gonads, heart, hepatopancreas, and gills), and exoskeleton, were individually analyzed for metal concentrations.

3. The final concentrations of Fe and Zn, expressed in mg/kg on a dry basis, were determined using the equation:

 $[Cf] = (C \times V \times f) / m$

Where:

- C = concentration in mg/mL obtained using the metal calibration curve
- V = total volume of the extract (10 mL)
- f = dilution factor of the original extract, if applicable
- m = mass of the freeze-dried sample

2.5 DETERMINATION OF CONSUMABLE QUANTITY

To determine the quantity of *M. amazonicum* shrimp captured in the regions of Bailique, Mazagão, and Ilha de Santana that is safe for daily human consumption, the following equation was employed:

1,000g ÷ (mineral concentration) / (maximum intake limit)



2.6 STATISTICAL ANALYSIS

For statistical analysis, the recommended procedures of Sampaio (2010) were followed. These included the use of Infostat Version 9.0 software and the application of the Shapiro-Wilk test for normality assessment. The Duncan test was utilized for normal variables, and the Kruskal-Wallis test was applied for non-normal variables, both at a significance level of 5%.

3 RESULTS AND DISCUSSIONS

Table 1 provides the biometric data for *M. amazonicum* specimens (length and weight measurements), organized by gender and collection sites.

Table 1. Biometrics	of M. amazon	<i>icum</i> collected in Brazili	ian Amazon Rivers.
Location	Sex	Total Weight (g)	Length (mm)
Bailique	Male	6.24 ± 1.40^{b}	$9.48\pm0.93^{\text{a}}$
	Female	5.45 ± 0.92^{ab}	9.34 ± 068^a
Ilha de Santana	Male	5.33 ± 1.39^{ab}	$9.51\pm0.87^{\rm a}$
	Female	$5.05 \pm 1.21^{\text{a}}$	$9.16\pm0.70^{\rm a}$
Mazagão	Male	$7.41\pm0.97^{\circ}$	10.48 ± 0.60^{b}
	Female	$6.01\pm0.85^{\text{b}}$	$9.67\pm0.55^{\mathtt{a}}$
Bailique	Total	5.84 ± 1.23^{b}	$9.41\pm0.81^{\texttt{a}}$
Ilha de Santana	Total	$5.19 \pm 1.29^{\mathrm{a}}$	$9.34\pm0.79^{\mathtt{a}}$
Mazagão	Total	$6.71 \pm 1.14^{\circ}$	10.08 ± 0.70^{b}
	Male	$6.33 \pm 1.51b$	$9.82\pm0.92b$
Total	Female	$5.50 \pm 1.06^{\text{a}}$	$9.39\pm0.67^{\mathtt{a}}$
	Total	5.91 ± 1.29	9.61 ± 0.80

Different letters in the same column demonstrate significant differences using Duncan's parametric test (p<0.05). Source: Prepared by the authors (2023).

The average length and total weight values of the evaluated freshwater shrimp across all locations surpass those reported by Coelho et al. (2017) in the western region of Pará State. The authors documented measurements of 6.37 mm/1.89 g for males and 6.96 mm/2.57 g for females. Similarly, lower mean length and total weight measurements were observed in a study conducted in the State of Amazonas by Pereira et al. (2019), where measurements were 6.83 cm/2.62 g for males and 7.66 cm/3.68 g for females.



Among the three collection sites, the highest shrimp catch was noted in Mazagão, where the specimens exhibited greater weight and length when compared to those from Bailique and Santana Island. This variation can be attributed to an array of biotic and abiotic factors that influence the development of these organisms, including water quality, food availability, animal density, and overall health.

The results presented in Table 3 illustrate a higher accumulation of Zn in females compared to males, while no significant gender-based difference was observed for Fe concentrations. In the Bailique Archipelago, Zn accumulation was greater in females, whereas males showed higher concentrations of Fe.

For animals collected from Santana Island and Mazagão, there was no noticeable disparity in Fe and Zn concentrations between genders. Collectively, shrimp from Bailique displayed higher Zn accumulation in comparison to their counterparts from Santana Island and Mazagão. However, the presence of Fe was notably lower in shrimp from Mazagão, despite their larger size and weight, as indicated in Table 1. In contrast, animals from Santana Island and Bailique exhibited greater Fe accumulation. Furthermore, Zn accumulation in male organs exceeded that in muscle and the exoskeleton. Conversely, for Fe, the highest concentrations were observed in the organs, followed by the exoskeleton and muscle.

Recognizing the concentrations of heavy metals, micronutrients, and microminerals in crustaceans holds significance for both ecosystem management and ensuring the safety of these creatures for human consumption. To gauge the potential health risks posed by heavy metal contamination, the Food and Agriculture Organization (FAO) and the World Health Organization (WHO) have established maximum recommended limits for each heavy metal, as delineated in Table 2 (FAO & WHO, 1994).

Table 2. Minimum and maximum daily intake of Fe and Zn for adult humans.						
Recommended	Mínimum	Source	Maximum	Source		
Daily Intake						
Iron	14 mg/kg	BRASIL, 2005	100 mg/kg	FAO/OMS, 1994		
Zinc	7 mg/kg	BRASIL, 2005	100 mg/kg	FAO/OMS, 1994		
Source: Propered by the outport (2022)						

Source: Prepared by the authors (2023).



Iron and zinc are two indispensable micronutrients crucial for the well-being of both humans and animals. Their appropriate concentrations in the body are vital in physiological and metabolic processes, including oxygen transportation, immune system functionality, and DNA synthesis.

Iron stands as a fundamental constituent of hemoglobin, the protein found within red blood cells responsible for conveying oxygen to the body's cells. Meanwhile, Zn plays a critical role in a range of metabolic functions, encompassing enzyme activity and immune responses. Any alterations in the intake levels of these micronutrients can yield detrimental outcomes for both humans and animals (Zimmermann & Hurrell, 2007).

In light of the calculations regarding the maximum permissible consumption of M. amazonicum shrimp, taking into account the concentrations of Fe and Zn, it is advised that individuals limit their daily intake of shrimp to 96.52 g for Fe and 226.81 g for Zn. This will ensure adherence to the established daily intake limits in the regions under study.

Although Fe and Zn concentrations in the muscle tissue are comparatively lower than in other parts of the shrimp, prudence should be exercised when consuming shrimp caught in rivers proximate to sites impacted by mining activities. In such cases, it is advisable to avoid consuming the organs and exoskeletons of the shrimp, as these components may be intended for industrial purposes. The exoskeleton, for instance, can be utilized for extracting chitosan and astaxanthin, while the organs may be processed into animal feed.

Groups		Metals		
		Zn (mg/kg)	Fe (mg/kg)	
Sex	Male	$695.95 \pm 626.96^{\rm a}$	3117.7 ± 3019.61	
	Fêmea	779.19 ± 775.15^{b}	3339.79 ± 3283.1	
	Total	737.57 ± 701.06	3228.74 ± 3151.50	
Local				
	Male	$690.57 \pm 530.06^{\text{b}}$	2992.26 ± 2440.53	
Bailique	Female	$925.1 \pm 462.22^{\circ}$	4490.22 ± 4338.08	
	Male	$784.06 \pm 714.08^{\text{b}}$	4173.41 ± 4028.05	
Ilha de Santana	Female	843.2 ± 747.35^{ab}	2271.22 ±2101.9 ^a	
	Male	605.73 ± 559.48^{a}	1930.02 ± 2356.2	



Mazagão	Female	649.25 ± 576.38^{ab}	$2445.76 \pm \!$
Bailique	Total	807.83 ± 508.36^{b}	4241.29 ± 4195.05^{b}
Ilha de Santana	Total	$814.3\pm810^{\rm a}$	3272.31 ± 2081.37^{b}
Mazagão	Total	$627.49 \pm 621.12^{\rm a}$	4241.29 ± 4195.05^{a}
Tissue			
Muscle	Male	$292.03 \pm 138.22^{\rm a}$	889.86 ± 429.99^{a}
Organs	Male	$1360.45\pm800.71^{\circ}$	$6822.62 \pm 4160.53^{\text{d}}$
Carapace	Male	366.31 ± 184.71^{ab}	$2073.09 \pm 1924.04^{\circ}$
Muscle	Female	$579.84 \pm 495.7^{\rm b}$	1175.73 ± 875.81^{ab}
Organs	Female	$1409.85 \pm 625.75^{\circ}$	6906 ± 4902.97^{d}
Carapace	Female	412.06 ± 273.99^{ab}	$127035 \pm 429.99^{\text{bc}}$
Muscle	Total	440.89 ± 394.65^{a}	$1036.12\pm 705.6^{\rm a}$
Organs	Total	1384.88 ± 715.97^{b}	$6664.31 \pm 4520.92^{\circ}$
Carapace	Total	$384.18 \pm 235.35^{\rm a}$	1671.72 ± 1478^{b}

Different letters in the same column demonstrate significant differences using the Kruskal-Wallis nonparametric test (p<0.05). **Source:** Prepared by the authors (2023).

In a study involving the marine shrimp *Penaeus merguiensis*, Yap et al. (2019) reported the concentration of Zn in muscle (55.5 μ g/g) and exoskeleton (42.9 μ g/g). Anandkumar et al. (2017) revealed that the average Zn concentration in the muscle of the crustacean *Harpiosquilla harpax* ranged from 62.43 to 203.00 μ g/g, with the highest concentration being 203.00 μ g/g. For *Acetes indicus* shrimp, the observed concentration of Zn was 62.42 μ g/g. Notably, the average concentration of Zn in the muscles of freshwater shrimp in this present study was 292.03 ± 138.22 for males and 579.84 ± 495.7 for females, surpassing the results of 114.56 μ g/g found for marine shrimps of the *Penaeus* genus by Anandkumar et al. (2017).

Furthermore, the Zn concentration in shrimp muscles in the current study exceeded the values reported in Malaysia (68-186 μ g/g) for the crustacean *Penaeus indicus*, as indicated by Patimah & Dainal (1993). Similarly, Ismail et al. (1995) reported Zn concentrations in the same species (5.00-16.00 μ g/g of Zn). Everaarts & Nieuwenhuize (1995) found concentrations of 106.00 μ g/g of Zn in shrimps of *Litopenaeus vannamei* on the Kenyan Coast. Vázquez et al. (2001) identified concentrations of 107.00 μ g/g of Zn for the crustacean *Penaeus monodon* in the Gulf of Mexico. Off the Cochin Coast in India (44.80 - 88.70 μ g/g), Borrel et al. (2016) reported



values of 18.80 to 55.14 μ g/g. From the coast of Indonesia, Soegianto et al. (1999) recorded concentrations of 2.13 μ g/g.

However, the total value of Zn in shrimp muscles in Amapá, which was $440.89 \pm 394.65 \text{ mg/kg}$, was lower than the total value (1184.00 µg/g) reported by Guhathakurta & Kaviraj (2000) in their study with *Litopenaeus vannamei* in Sunderbans, India. The results obtained by Firat et al. (2008) showed significantly higher values in the hepatopancreas (804.8±103.1 Zn and 684.9±153.2 Fe) compared to the muscle in male marine shrimp *Penaeus semisulcatus*. Shrimps, being benthic organisms, tend to accumulate micronutrient particles in their bodies through absorption via the gills, sediments, and contaminated water.

In studies conducted by Sanchez-Chardi et al. (2007), Zn and Fe were found in large quantities in the organ tissues of *Penaeus semisulcatus*. In gill tissues analyzed by El Gendy et al. (2015), the concentration of Zn was higher than in the exoskeleton and greater than in the muscle.

In contrast to Penaeid shrimps, Carid crustaceans exhibit Zn regulations in body concentrations in proportion to the bioavailability of dissolved Zn, combining Zn excretion with Zn absorption (Nuñez-Nogueira et al., 2005).

The higher presence of Zn in shrimp organs is likely linked to the elevated bioavailability of this element in the aquatic environment. Additionally, it is associated with its role as an enzymatic regulator in several enzymes in the hepatopancreas and glands located in the animal's cephalothorax (Bryan & Langston, 1992). Following the findings of Lemos & Dantas (2023), Zn displayed an average concentration of 52 mg/kg in the muscle of the shrimp *M. amazonicum*.

Due to its strong affinity for metallothionein, Zn maintains a low excretion rate and tends to accumulate non-toxically inside the cell, reducing its elimination. It is important to highlight that females have a greater metabolic demand for Zn during their reproductive phase compared to males during the same period (Amaral et al., 2005; Nascimento et al., 2016).

At concentrations of 15 and 20 mg/kg of Zn in the hepatopancreas of freshwater shrimp *M. nipponense*, reproductive parameters, especially the egg fertilization rate,



increased at higher concentrations (50 mg/kg). Furthermore, there were noticeable delays in the molting and spawning periods, and reproduction was ultimately interrupted (Tavabe, 2023).

In the case of females, Zn accumulation was higher in the organs and no significant difference was observed between Zn concentrations in the muscle and exoskeleton. Several studies have suggested that females tend to accumulate more Zn than males, primarily due to the development of their gonads (Wang & Rainbow, 2008; Nascimento et al., 2016). Organs such as the gonads, hepatopancreas, and gills exhibit a greater capacity for metal absorption compared to muscle tissues, primarily due to their higher metabolic activity. As a result, they tend to accumulate higher concentrations of metals (Langston, 1990; Amundsen et al., 1997).

Zinc is indeed an essential element, playing critical roles related to growth, immune function, cellular metabolism, and the survival of most animals. Elevated levels of these metals may be attributed to their essentiality for the organism (Baboli et al., 2013). However, excessive levels of Zn can lead to metabolic disorders and deficiencies in other metals such as Fe and Cu. Conversely, excessive Fe levels can impede the body's ability to absorb Zn (Fosmire 1990; Cuajungco et al., 2021).

Cresswell et al. (2014) conducted an assessment of Zn concentrations in tissues of *Macrobrachium australiense*, *M. rosenbergii*, and *Macrobrachium latidactylus*. They found an average concentration of 144 ± 33 mg/kg in the organs of these species. In contrast, the present study reported notably higher values, with Zn concentrations of 1360.45 ± 800.71 mg/kg for males and 1409.85 ± 625.75 mg/kg for females.

In the marine shrimp *Penaeus semisulcatus*, Zn concentrations were found to be higher in the hepatopancreas than in the muscle and exoskeleton (Pourang, 2005). Anderson et al. (1997) also reported that in decapod crustaceans, the hepatopancreas is the primary site for the storage of metals and micronutrients, and it plays a vital role in detoxification in these animals, in addition to its central role in metabolism. Lindahl & Moksnes (1993) suggested that the increase in Zn concentration in the hepatopancreas, compared to other shrimp tissues, may be related to the organ's function in excreting substances from the organism.



As per Zhang et al. (2021), the dietary Zn requirement for shrimp of the genus *Macrobrachium* falls within the range of 50 to 81 mg/kg. Meanwhile, Srinivasan et al. (2016) concluded that the daily Fe intake for *M. rosenbergii* can go up to 20 mg/kg. Both authors highlight that exceeding these mineral requirement levels can harm the growth, reproduction, nutrition, and feeding of shrimp due to mineral excess.

Lin et al. (2013) reported that Zn had a significant impact on reducing the cumulative mortality of *L. vannamei* when exposed to *Vibrio harveyi*, suggesting an enhancement of the immune system and, consequently, improved resistance in penaeid shrimp. Additionally, Zn intake can mitigate the toxicity of Cd, underscoring the role of zinc in aquatic environments for the well-being of shrimp (Brzóska & Moniuszko-Jakoniuk, 2001).

In terms of Fe concentration, it was most prominent in the organs, followed by the exoskeleton and muscle. Similar trends in the order of Fe accumulation among shrimp samples have been observed by other researchers (Páez-Osuna & Tron-Mayen, 1996). Notably, differences were noted in Fe accumulation within the exoskeleton, with farmed shrimp exhibiting lower concentrations than those captured in natural environments.

The relatively higher concentration of Fe in shrimp organs in this study can likely be attributed to the bioavailability of this element in the environment, as it is widely distributed in all terrestrial compartments (Soni et al., 2001). In marine environments, for example, the marine shrimp *Farfantepenaeus californiensis* showed significantly higher concentrations of Zn in the hepatopancreas compared to the muscle and exoskeleton (Frías-Espericueta et al., 2016).

This study reveals that Zn accumulates more in shrimp organs, with relatively lower concentrations in the muscle and exoskeleton. Conversely, Fe has the highest accumulation in organs, followed by the exoskeleton and muscle. Schmidt et al. (2009) also noted the common occurrence of Fe accumulation in the gills of penaeid shrimp, particularly in aquatic systems with an abundance of this micronutrient.

The exoskeleton of shrimp is recognized for its role in remediating acid mine drainage effluents, particularly for minerals like Fe (Gamage & Shahidi, 2007). This capability stems from the exoskeleton's composition, which includes chitin with a high



calcium carbonate content, acting as an effective biopolymer adsorbent and an acidneutralizing agent (Keteles & Fleeger, 2001; Núnez-Gomez et al., 2017; Rech et al., 2019). The chitin and chitosan present in the exoskeleton are acknowledged for their excellent metal-binding properties (Lemonnie et al., 2021).

In the study conducted by Lewtas et al. (2014), the gills of the penaeid shrimp *Metapenaeus bennettae* exhibited notably higher Fe concentrations when compared to other tissues. Crustaceans, in particular, store Fe in their hepatopancreas cells, which is then transported through various tissues and proteins. The concentration of minerals in water is reflected in the gills, whereas high mineral levels in sediments or ingested food manifest in the hepatopancreas (Lewtas et al., 2014).

The mineral concentrations in the gills primarily represent the absorption of metals present in the water, while the concentration in the hepatopancreas signifies metal storage (Romeo et al., 1999). Gills, which are responsible for oxygen extraction from the water, are exposed to large volumes of water, thus making them suitable for metal accumulation (Swaileh & Adelung, 1994). In contrast, muscle tissue might be less responsive to metal absorption or excretion compared to these organs.

M. amazonicum has the potential to accumulate both Zn and Fe. The results from this study demonstrate that the highest concentrations of Fe and Zn are indeed present in shrimp organs (P<0.05). Notably, higher Fe concentrations were observed in the gills and hepatopancreas of male shrimp when compared to the muscle and exoskeleton.

However, it is worth noting that Baki et al. (2018) discovered higher concentrations of Fe and Zn in the muscle and exoskeleton of the crustacean *L. vannamei*. Firat et al. (2008) reported that the muscle of *P. monodon* shrimp exhibited lower Fe concentrations when compared to the hepatopancreas. According to Eisler (1981), different crustaceans tend to accumulate trace metals in muscle, organs, and external skeletons at varying concentrations.

The variability in trace metal concentrations observed in the edible muscle tissue of the studied invertebrates is influenced by factors such as absorption, excretion, storage, and the efficiency of the organism in regulating and detoxifying these metals (Bryan, 1971). These physiological and biochemical strategies can vary between species

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(Gerlach, 1981), resulting in high and low concentrations that depend on the specific tissue and invertebrate involved (Rainbow, 1993, 1998), as well as the quality of the environmental water.

4 FINAL CONSIDERATIONS

The findings of this study indicate that the degree of mineral accumulation in freshwater shrimp *M. amazonicum*, collected from Mazagão, Bailique Archipelago, and the community of Ilha de Santana, suggests a daily shrimp intake limit of 96.52 g for Fe and 226.81 g for Zn.

These varying patterns of Fe and Zn accumulation provide insights for distinguishing which parts of the shrimp are suitable for different commercial purposes, because the muscle, being the largest edible part, contains lower concentrations of these minerals compared to the organs and external skeleton. Furthermore, *M. amazonicum* females accumulate more Zn as a reproductive strategy, as they require greater Zn during gonadal development compared to males in the reproductive process.

Shrimp exoskeletons can be used effectively for treating pollution caused by mine waste effluents, particularly for minerals like Fe. The higher concentrations of Fe and Zn in the organs are closely related to the hepatopancreas, an organ with functions encompassing storage, detoxification, and substance excretion.

Regarding organs and exoskeletons, it is recommended to exercise caution when using them for culinary purposes; however, they can be valuable in industrial processes for extracting chitosan and astaxanthin. In this context, the Amazon shrimp emerges as a species of significant ecological and nutritional importance, both for humans and other animals in its food web, particularly considering the presence of the metals analyzed in this study.



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