BOLETIM DE CONJUNTURA

BOCA

Ano VI | Volume 19 | Nº 57 | Boa Vista | 2024

http://www.ioles.com.br/boca

ISSN: 2675-1488

https://doi.org/10.5281/zenodo.14254327

BOLETIM DE CONJUNTURA

www.ioles.com.br/boca

TRACE METALS DEPOSITION IN OTOLITHS OF *PSECTROGASTER AMAZONICA* AS ENVIRONMENTAL INDICATORS IN THE MIDDLE TOCANTINS RIVER

Matheus Bilio Alves[1]

Thiago Machado da Silva Acioly[2]

Alana Lislea de Sousa[3]

Diego Carvalho Viana[4]

Abstract

The growing expansion of aquaculture and the increasing pollution of water bodies have raised concerns about the accumulation of trace metals in aquatic organisms, particularly fish. In this context, otoliths, structures located in the inner ear of fish, stand out as important tools for environmental monitoring, being used to assess the presence of pollutants and identify species based on the morphology of these structures. This study investigated the morphometry of otoliths and the deposition of trace metals (Calcium, Aluminum, Vanadium, Potassium, Copper, Iron, Magnesium, Indium, Gold, Tin, Mercury, and Manganese) in Psectrogaster amazonica from the middle Tocantins River, Maranhão, Brazil. Otoliths from 37 specimens were analyzed using inductively coupled plasma optical emission spectrometry (SHIMADZU, ICPE-9000, Kyoto, Japan) to quantify trace metals. Morphometric data were used to calculate shape indices such as roundness, circularity, rectangularity, ellipticity, and form factor, correlating these indices with pollutant presence. Among the elements analyzed, Aluminum (0.823 mg/L), Vanadium (0.382 mg/L), and Copper (0.035 mg/L) showed average concentrations exceeding certain national and international standards. The aluminum results are particularly concerning, as previous studies along the banks of the Tocantins River have reported elevated levels of this metal in water and sediments. These findings suggest that P. amazonica can serve as an environmental biomarker, contributing to understanding pollution levels in aquatic environments and highlighting the importance of otoliths in ecological assessments.

Keywords: Environmental Indicators; Metals; Tocantins River.

Resumo

A crescente expansão da piscicultura e o aumento da poluição das águas têm gerado preocupações sobre o acúmulo de metais traço em organismos aquáticos, especialmente em peixes. Nesse contexto, os otólitos, estruturas presentes no ouvido interno dos peixes, destacam-se como ferramentas importantes para o monitoramento ambiental, sendo utilizados tanto na avaliação da presença de poluentes quanto na identificação de espécies com base na morfometria dessas estruturas. Este estudo investigou a morfometria dos otólitos e a deposição de metais traço (Cálcio, Alumínio, Vanádio, Potássio, Cobre, Ferro, Magnésio, Índio, Ouro, Estanho, Mercúrio e Manganês) em *Psectrogaster amazonica* no médio Rio Tocantins, Maranhão, Brasil. Foram analisados otólitos de 37 exemplares da espécie utilizando espectrometria de emissão óptica com plasma indutivamente acoplado (SHIMADZU, ICPE-9000, Kyoto, Japan) para quantificação dos metais traço. Os dados morfométricos permitiram calcular índices de forma, como redondeza, circularidade, retangularidade, elipticidade e fator de forma, correlacionando-os com a presença de poluentes. Dentre os elementos analisados, Alumínio (0,823 mg/L), Vanádio (0,382 mg/L) e Cobre (0,035 mg/L) apresentaram concentrações médias superiores a certos padrões nacionais e internacionais. Os resultados de alumínio são especialmente preocupantes, uma vez que estudos anteriores nas margens do Rio Tocantins indicam níveis elevados desse metal em água e sedimentos. Os resultados indicam que *P. amazonica* pode atuar como um biomarcador ambiental, contribuindo para o entendimento dos níveis de poluição em ambientes aquáticos e evidenciando a importância dos otólitos em avaliações ecológicas.

Palavras chave: Indicadores Ambientais; Metais; Rio Tocantins.

---

[1] Bachelor in Veterinary Medicine at the State University of the Tocantina Region of Maranhão (UEMASUL). E-mail: matheusimalves@gmail.com

[2] Ph. D. candidate in Animal Science at the State University of Maranhão (UEMA). E-mail: tmsacioly@gmail.com

[3] Professor at the State University of Maranhão (UEMA). Doctor of Sciences. E-mail: alislea@hotmail.com

[4] Professor at the State University of the Tocantina Region of Maranhão (UEMASUL). Doctor of Sciences. E-mail: diego_carvalho_@hotmail.com

INTRODUCTION

The increasing concern about water pollution and the bioaccumulation of trace metals in aquatic organisms, particularly fish, highlights the importance of using biomarkers such as otoliths. These calcified structures in fish's inner ear have been extensively studied due to their potential to record environmental information and reflect the quality of the ecosystems where fish live.

Studying otolith composition and morphometry is crucial given the need for environmental monitoring in regions heavily impacted by human activities, such as the Tocantins River basin. Understanding pollution levels and their impacts on aquatic organisms not only contributes to conservation strategies but also aids in water resource management and the health of communities that depend on these ecosystems.

This study aimed to investigate the morphometry and chemical composition of the otoliths of ***Psectrogaster amazonica***, correlating their morphometric characteristics with trace metal deposition. The research seeks to evaluate the potential of this species as an environmental biomarker in the Middle Tocantins River, providing novel data on pollution in the region.

The lack of detailed studies on the presence of trace metals in fish otoliths in the Tocantins region hinders the understanding of the impact of local pollution. This raises the question of how metal bioaccumulation in *P. amazonica* can be used to assess environmental quality and inform mitigation strategies.

The study is grounded in the concept that otoliths, being biologically inert structures, permanently record a fish's exposure to trace elements in aquatic environments. Previous literature emphasizes the relevance of otoliths as indicators of environmental changes and as tools for population discrimination and pollution monitoring.

The methodology involved collecting otoliths from 37 specimens of *P. amazonica* in the study region, followed by morphometric and chemical analyses. Inductively Coupled Plasma Optical Emission Spectrometry (ICPE-9000) was used to quantify trace metals, while shape indices were calculated based on images obtained and analyzed using ImageJ Plus software.

The article is structured into distinct sections to ensure a comprehensive exploration of the subject matter. The introduction provides the context, objectives, and relevance of the study, highlighting the importance of otoliths as biomarkers in assessing environmental quality. The theoretical framework delves into the concepts of trace metal contamination, otolith chemistry, and morphometry, supported by relevant literature. The materials and methods section details the study area, specimen collection, and analytical techniques used for morphometric and chemical analysis of the otoliths. The results and discussion present the findings, including morphometric characteristics and trace metal concentrations, with comparisons to existing studies. Finally, the conclusion lists the main contributions of the research and suggests directions for future studies to advance environmental monitoring and conservation efforts.

TRACE METAL CONTAMINATION

Aquatic ecosystems are under constant threat from contamination by a variety of anthropogenic pollutants originating from industrial, urban, agricultural, and mining activities. Among these, trace metals are particularly harmful due to their widespread distribution and potential to accumulate at high concentrations, posing serious risks to human health, aquatic organisms, and the environment (KUMKAR *et al*., 2023; ONYENA *et al*., 2024). This issue is especially critical in developing nations, where inadequate regulation exacerbates contamination, turning it into a global concern affecting rivers worldwide (CHEN; DING, 2023; GHANI *et al*., 2023).

Trace metals, characterized by their presence in low concentrations across environmental compartments, are commonly found in soil, sediments, plants, and animals (ALVARIÑO *et al*., 2023). The most toxicologically significant metals in aquatic environments—mercury (Hg), arsenic (As), chromium (Cr), lead (Pb), cadmium (Cd), nickel (Ni), and zinc (Zn)—pose severe risks to organisms when present above critical thresholds. Nevertheless, technological advances allow for accurate quantification of these metals, even at trace levels (MALLIK *et al*., 2023).

One of the most concerning aspects of these metal contamination is their bioaccumulative nature, which leads to amplified toxic effects in aquatic organisms (CAVALCANTE *et al*., 2021). Fish are particularly valuable bioindicators for monitoring polluted environments, as their close interaction with the aquatic habitat provides insights into ecosystem health. Research has increasingly focused on the bioaccumulation of pollutants in fish, given their role as environmental biomarkers reflecting water quality (LOMARTIRE *et al*., 2021). Notably, trace and heavy metals accumulate in fish otoliths, structures that record the environmental conditions experienced by the fish throughout its life (VRDOLJAK *et al*., 2020). The elemental composition of otoliths serves as a permanent archive of environmental conditions. These structures grow incrementally, depositing layers that incorporate trace metal signatures from the water, providing a chronological record of exposure.

In the middle Tocantins River, *Psectrogaster amazonica*—known locally as "branquinha"—is a native fish species widely consumed and traded in Imperatriz, Maranhão. This species is a vital resource for local riverside communities that rely on the Tocantins River for subsistence (SILVA, 2022).

Belonging to the family Curimatidae, order Characiformes, *P. amazonica* is a detritivorous fish that feeds on microorganisms, organic matter, and algae. It reaches up to 20 centimeters in length, has a silver coloration with dark spots, and plays a critical role as an environmental biomarker (MORAES, 2021; Pereira, 2019).

Otoliths, primarily composed of calcium carbonate, are located in the inner ear of fish and are essential for orientation, balance, and hearing. Each fish has two otoliths—right and left—immersed in endolymph, a fluid capable of absorbing environmental pollutants, including trace metals (REIS-SANTOS *et al*., 2023). These structures are categorized into three types—sagitta, lapillus, and asteriscus—stored in compartments called sacculus, utriculus, and lagena, respectively (REIS-JÚNIOR *et al*., 2023). The morphometric analysis of otoliths is crucial for studies on population dynamics, species identification, and ecological research. These analyses provide species-specific information that aids in differentiating taxa and assessing population structures, contributing to sustainable fisheries management and conservation efforts (SANTOS *et al*., 2024; NEVES *et al*., 2023).

The middle Tocantins River has faced increasing environmental pressures due to agricultural expansion, deforestation, and the introduction of non-native species (ACIOLY *et al*., 2024). These activities have intensified environmental degradation, altering water quality and natural habitats. Consequently, trace metals from pesticides, industrial waste, and fertilizers accumulate in aquatic ecosystems, threatening biodiversity and posing health risks to communities relying on the river for sustenance (MADADI *et al*., 2021).

Fish otoliths absorb various substances present in the aquatic environment, including trace metals, which become integrated into their structure as the fish grows (SPICH; FEY, 2022). This makes otoliths valuable for assessing the environmental conditions experienced by the fish. However, despite their potential as tools for environmental monitoring, studies investigating trace metal concentrations in the otoliths of *P. amazonica* are still lacking for the Tocantina region of Maranhão.

OTOLITHS CONCEPTS

Otoliths are calcified structures primarily composed of calcium carbonate, located within the inner ear of fish, housed in the neurocranium (PONTUAL *et al*., 2024). They play a crucial role in maintaining balance, detecting sound, and sensing linear and angular acceleration, which relates to head movement about the body. Each inner ear contains three otolithic organs: the sacculus, utriculus, and lagena (D'IGLIO *et al*., 2022). Within these organs lie three otoliths: the sagitta, lapillus, and asteriscus.

Each fish possesses a pair of otoliths, which are surrounded by the otolithic membrane, a structure representing the space between the otolith and the macula sacculi (WANG *et al*., 2021).

The formation of otoliths occurs through a biomineralization process involving the continuous deposition of calcium carbonate ($CaCO_3$) crystals, predominantly in the form of aragonite, with lesser amounts of vaterite and calcite (WAGNER *et al*., 2021). These deposits form organized layers, increasing incrementally with the fish's age, with one layer typically forming per day (YEDIER *et al*., 2022). This layered structure allows researchers to determine the age of fish—a technique initially described by Reibisch (1899) and later refined into a precise methodology (CAMPANA *et al*., 1992).

Otoliths not only fulfill critical physiological roles, such as balance and sound detection but also act as permanent archives of environmental conditions (CURTHOYS, 2020). The growth rings within these structures reflect changes in water chemistry, providing a chronological record of the fish's habitat (BOYLE, 2021). This dual functionality—biological and environmental—renders otoliths exceptional tools for multidisciplinary research in fields like ecology, fisheries science, and environmental monitoring.

The chemical composition of otoliths further enhances their utility in environmental studies. While primarily composed of carbon (C) and calcium (Ca), otoliths also incorporate trace amounts of elements such as sodium (Na), potassium (K), strontium (Sr), sulfur (S), nitrogen (N), phosphorus (P), and chlorine (Cl) (MARTINO *et al*., 2021). These elemental compositions vary in response to environmental factors, including salinity, temperature, pH, and nutrient availability, as well as intrinsic factors such as species, age, life stage, and genetics (ANDRADE *et al*., 2020). For instance, a study conducted in Uruguay and Argentina found higher concentrations of Sr and Ca in marine fish compared to freshwater species (AVIGLIANO *et al*., 2021).

Trace elements are incorporated into otoliths primarily through the surrounding water and the fish's diet. Dissolved ions in the water, such as potassium, strontium, and sodium, are integrated into the otolith's mineral matrix during daily deposition processes (FEY *et al*., 2021). This incorporation occurs via concentration gradient mechanisms between the water and the fish's body fluids, particularly the endolymph within the otolithic membrane (LOEPPKY *et al*., 2021). Additionally, dietary intake contributes to the deposition of trace elements after digestion (HUANG *et al*., 2021).

Otoliths are highly valuable in environmental analysis due to their capacity to absorb trace elements, including copper (Cu), mercury (Hg), iron (Fe), aluminum (Al), and others (KHAWAR *et al*., 2024). These structures provide a historical and ongoing record of aquatic environments, enabling continuous monitoring of water quality and pollution levels (JOHNSON *et al*., 2020). Furthermore, the elemental composition of otoliths can reveal migratory patterns and habitat use, offering critical data for understanding species movement and assessing aquatic ecosystem health (WINDOM *et al*., 2024).

In Brazil, the use of otoliths for monitoring environmental pollution has gained increasing attention, especially in regions impacted by mining, agriculture, and industrial activities (HOFF *et al*., 2022). Despite this progress, many freshwater species remain underexplored, underscoring the need for further research. Expanding studies on these species is essential for informing environmental management policies, mitigating pollution impacts, and preserving aquatic biodiversity (SANTOS *et al*., 2022).

OTOLITH SHAPE

The morphometry of otoliths is a widely utilized tool for fish stock discrimination and the management of species populations. However, the factors that determine otolith shape remain only partially understood (VIGNON *et al*., 2010). Given that otoliths do not undergo structural changes after their complete formation, they serve as permanent records of the fish's life history. This unique feature offers valuable opportunities to investigate which factors influence their morphology across different species (KIKUCHI *et al*., 2021).

Otolith shape is influenced by a combination of biotic and abiotic factors, genotypic characteristics, and the life cycle stages of the fish, such as juvenile, adult, and senile phases (MAHÉ *et al*., 2021). Among the biotic factors, water quality parameters—such as pH, temperature, and salinity—and the fish's genotype are particularly significant. Regarding abiotic influences, habitat characteristics, including freshwater or saltwater environments, turbulence, and water depth, as well as food availability in terms of type and quantity, are critical (D'IGLIO *et al*., 2021). For example, a study on *Amphiprion akindynos* and *Pomacentrus amboinensis* demonstrated that food deprivation can affect otolith development (GAGLIANO *et al*., 2004).

Abnormalities in otoliths can arise due to genetic predispositions, often exacerbated by environmental factors. For instance, trace metals like mercury and lead can induce asymmetries in otolith structures, impairing the fish's balance (KOEBERLE *et al*., 2019). A study conducted in the Mediterranean Sea, Tunisia, involving 240 specimens of *Pagellus erythrinus*, observed otolith asymmetries, potentially linked to genetic factors or physiological stress caused by local water pollution (MEJRI *et al*., 2020). Similarly, research on Pampus candidus in Iraq revealed otolith mass asymmetry in fish exposed to polluted waters (JAWAD *et al*., 2024).

To analyze otolith shape indices, the structures are carefully positioned in Petri dishes under a microscope with reflected light and dark background, ensuring the acoustic sulcus faces upwards (RAHNMAMA *et al.*, 2023). High-resolution images are captured using digital cameras, allowing precise measurements of morphological parameters (QIAO *et al.*, 2022). Specialized image processing software is employed to determine metrics such as length, height, width, area, and perimeter. These data are then used to calculate shape indices, enabling comparisons between otolith morphology and standard geometric shapes (JAWAD *et al.*, 2022).

Otolith shape analysis has proven essential for fish stock discrimination, offering critical insights for sustainable fish population management (AGIADI *et al.*, 2022). Identifying morphological variations between populations aids in the establishment of protected areas, the definition of fishing quotas, and the monitoring of aquatic ecosystem health (ORDOÑEZ *et al.*, 2022). Furthermore, otolith morphometry helps to elucidate migration patterns and habitat connectivity, supporting the development of conservation strategies tailored to local fishery resources (POLITIKOS *et al.*, 2021).

Recent studies indicate that otolith morphometry can vary significantly even among individuals of the same species, particularly when they inhabit distinct environments (GUT *et al.*, 2020). Evidence suggests that water depth influences otolith size and density, with fish from deeper waters exhibiting larger and denser otoliths (MORALES *et al.*, 2023). Additionally, morphometric analyses provide insights into evolutionary adaptations to environmental changes, offering valuable data for ecological studies and predictions regarding fish population responses to ecosystem alterations (NYGAARD *et al.*, 2021).

The study of otolith shape also contributes to evolutionary biology, as these structures reflect long-term adaptations to ecological niches (SMOLINSKI *et al.*, 2019). Variations in otolith morphology among populations often indicate evolutionary responses to selective pressures such as predation, feeding strategies, or habitat complexity (VASCONCELOS *et al.*, 2024). For instance, comparative analyses between demersal and pelagic fish species reveal significant differences in otolith compactness and edge irregularity, corresponding to their distinct ecological roles (MESA *et al.*, 2020).

OTOLITH CHEMISTRY

Since the earliest studies on the chemical composition of otoliths, microchemical analysis has advanced significantly, especially since the 1990s, thanks to technological breakthroughs in biogeochemistry (HÜSSY *et al.*, 2021). Spectrometry techniques, such as ICP-MS (Inductively Coupled Plasma Mass Spectrometry) and XRF (X-Ray Fluorescence), have revolutionized this field by providing high-precision identification of trace elements, offering detailed insights into aquatic habitats (AVIGLIANO *et al*., 2021).

These methodologies are pivotal for detecting and quantifying chemical elements in various samples, with applications spanning geoscience, biology, and ecology (LORENC *et al*., 2022). ICP-MS, in particular, is widely recognized for its sensitivity in detecting heavy metals, such as lead, mercury, and copper, even at trace concentrations, making it an ideal tool for analyzing biological samples like otoliths (MACIEL *et al*., 2021).

The importance of spectrometry in ecological studies can be traced back to pioneering research, such as Odum's (1951) analysis of trace element incorporation in gastropod tissues, which laid the groundwork for subsequent studies in otolith microchemistry. Building on this foundation, Kalish (1989) explored the influence of temperature, season, and fish age on the chemical composition of otoliths in *Arripis trutta*, further advancing the understanding of environmental variables affecting otolith formation.

While other structures, such as scales, vertebrae, opercula, and cleithra, can accumulate trace elements, otoliths stand out due to their inert nature (VRDOLJAK *et al*., 2020). Once chemical elements are deposited in otoliths, they remain unaltered, providing a permanent record of environmental conditions throughout a fish's life (WOOD *et al*., 2022). This characteristic makes otoliths a critical tool for ecological and environmental studies, particularly in monitoring the deposition of potentially toxic trace metals (REIS-SANTOS *et al*., 2022).

Otolith microchemistry has also been extensively employed to investigate fish migratory patterns. For example, a study in the Mekong River, Asia, utilized strontium/calcium (Sr:Ca) and barium/calcium (Ba:Ca) ratios to reveal that Pangasius mekongensis and Pangasius krempfi migrate up to 1400 km from marine to freshwater environments for spawning (VU *et al*., 2022). Similarly, research in the St. Lawrence River, Canada, demonstrated the extensive migratory range of the invasive species Tinca tinca, reaching distances of up to 250 km (MORISSETTE *et al*., 2021).

Emerging studies have highlighted the potential of otolith chemistry for assessing the bioavailability of contaminants and their ecological impacts (SPILSBURY *et al*., 2022). Trace elements such as cadmium (Cd), lead (Pb), and arsenic (As) detected in otoliths not only act as markers of environmental pollution but also provide valuable information on bioaccumulation and the trophic transfer of these toxicants within aquatic food webs (KOROSTELEV *et al*., 2021). Understanding these dynamics is crucial for designing mitigation strategies for polluted ecosystems.

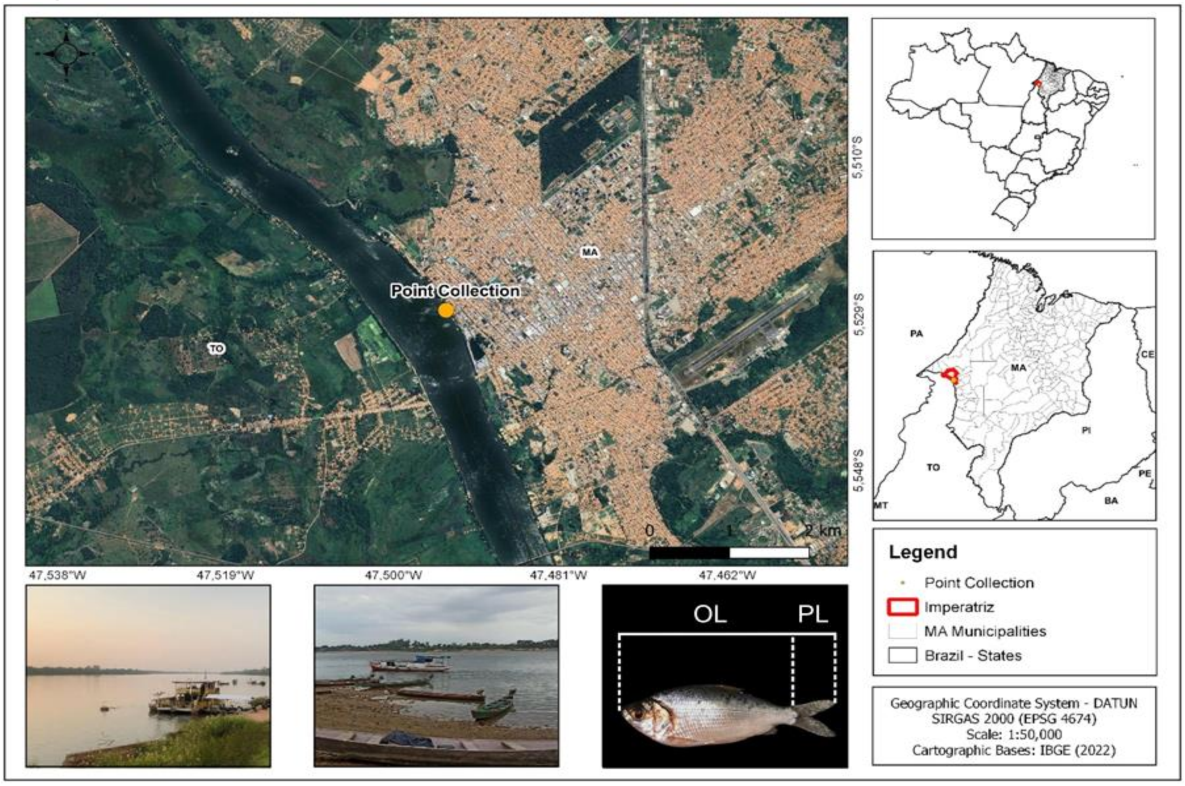
Despite these advancements, the application of otolith microchemistry remains geographically and taxonomically limited, particularly in regions like South America (HERMANN *et al*., 2021). For instance, although the Amazon Basin presents significant research potential, only 3 out of 45 studies on otolith microchemistry in South America have focused on species from this region (AVIGLIANO *et al*., 2016). Combining spectrometry techniques with isotopic analysis could enhance the understanding of environmental processes affecting fish populations, providing essential data for conservation strategies in such biodiverse areas (PHILLIPS *et al*., 2020).

MATERIALS AND METHODS

Study area, sampling, and ethical aspects

The study was conducted in the middle Tocantins River, within the area of influence of Imperatriz, Maranhão, Brazil (Figure 1). This region is a strategic transportation hub and gateway to the Amazon, often called the "portal to the Amazon. The area is well-urbanized with significant human activity, with thriving agribusiness and industry driving growth and regional advancement.

Figure 1 – Geolocation of the collection point and specimen *Psectrogaster amazonica*



Source: Self elaboration.
Note: OL = Overall length; PL = Partial length.

This site faces potential contamination from high vessel traffic, recreational beach activities, waste disposal, residues from commercial iceboxes, and urban sewage discharge (ACIOLY *et al*., 2024). The same authors provide insights into the distribution of potentially toxic elements in regional sediments, noting elevated levels of Al, with Cr, Ni, Pb, Cu, and Zn exceeding regulatory quality standards. Another study by Acioly *et al*. (2024) assesses levels of potentially toxic and essential elements in the water, finding concentrations of Al, Cu, Fe, Mg, and Se above regulatory thresholds.

The fish specimens, *P. amazonica* (known locally as "branquinha"), were collected in 2023 and handled following rigorous sample preservation protocols. Each sample was carefully stored in pre-cleaned plastic vials, thoroughly rinsed with metal-free water, and kept within a cold chain (refrigerated at 4 °C) until they arrived at the laboratory of the State University of the Tocantina Region of Maranhão (UEMASUL). Sagittal otoliths—the largest of the three otolith pairs, responsible for enabling fish to hear, sense water vibrations, maintain balance, and navigate—were extracted from each specimen. The otoliths were removed with anatomical forceps through a ventral opening in the neurocranium (Figure 1). Following extraction, both left and right sagittal otoliths were cleaned by soaking in a 10% sodium hydroxide solution for 3–5 days and then transferred to individual vials for storage (MACDONALD *et al*., 2019).

The 37 specimens in the study averaged over 40 grams in weight, with total lengths exceeding 14 cm and standard lengths over 11 cm, showing minimal variation in size, which allowed for more precise morphometric analysis. Both left and right otoliths displayed consistent morphologies, including characteristic anatomical features such as the sulcus acusticus, antirostrum, rostrum, dorsal depression, and ventral sulcus (PEREIRA *et al*., 2023). Significant variations in otolith morphology are rare and generally linked to sudden dietary shifts that impact opacity, shape, and structure (WIĘCASZEK *et al*., 2020).

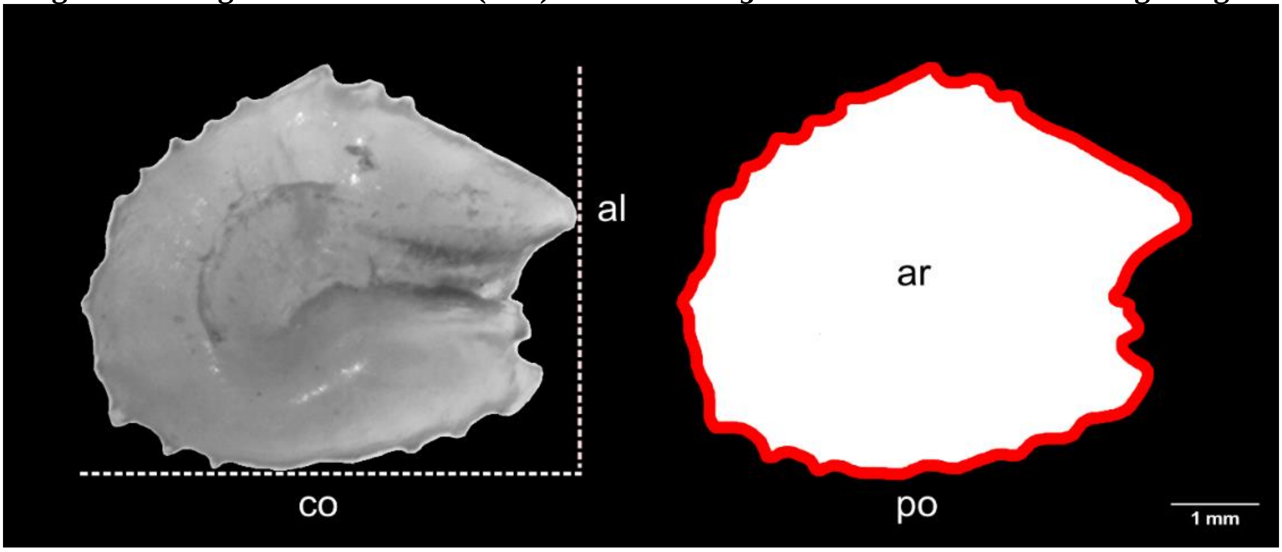
The animals in this study were collected together using fishing nets, making it unlikely to observe drastic changes in otolithic conformation. Field collection and laboratory procedures adhered to ethical standards and were formally reviewed and approved by the Institutional Ethics Committee of the State University of the Tocantin Region of Maranhão (CEUA No: 1025220722) and the State University of Maranhão (CEUA No: 040/2023). Additionally, scientific activities were conducted with authorization (SISBIO No: 87310–1), in compliance with Normative Instruction No. 748/2022, as specified by ICMBio Ordinance.

Otolith preparation, imaging, and shape analysis

The otoliths were mounted on polished glass microscope slides (26 × 76 mm) with the acoustic groove facing upward and the anterolateral region oriented to the left. Observations were conducted under a Stemi 305 (Zeiss, New York, United States of America) stereomicroscope equipped with Proview® (digital image acquisition software. Each otolith image was calibrated using photographs taken alongside a measuring ruler and converted into two-dimensional images in ImageJ Plus software to minimize errors caused by otolith curvature. The methodology involved image calibration and the measurement of length, height, area, and perimeter for 37 pairs of otoliths, resulting in a total of 74 analyzed images.

For shape analysis, the ImageJ Plus software was calibrated using an image of an otolith placed alongside a measuring ruler. After calibration, measurements of height (mm), length (mm), area (mm), and perimeter (mm) were taken for each otolith in the paired images, separated into left and right otoliths for each sample (Figure 2). To enhance measurement accuracy and minimize error, the images were adjusted for brightness, reduced noise from luminosity, and transformed into two-dimensional representations. These adjustments were performed using the same software.

Figure 2 - Length measurements (mm) of the *Psectrogaster amazonica* otolith using ImageJ



Source: Self elaboration.
Note: Otolith height (al); Otolith length (co); Otolith area (ar); and Otolith perimeter (po).

Shape indices, including roundness, circularity, rectangularity, ellipticity, and form factor, were calculated using specific equations (Table 1). Roundness and circularity assess the similarity of the otolith's shape to a perfect circle, while rectangularity describes the relationship between the otolith's height and length. Ellipticity evaluates the proportionality of changes in the axes, and the form factor quantifies surface irregularities (ZISCHKE *et al*., 2016).

Table 1 – Shape indices calculated for the otoliths of *Psectrogaster amazonica* and their equations

Shape índices	Equation	Reference
Roundness	$4AR / \pi CO^2$	Moore *et al*. (2022)
Circularity	PO2 / AR	Saygin *et al*. (2020)
Rectangularity	OA/ (CO x AL)	Souza *et al*. (2020)
Ellipticality	(CO – AL) / (CO + AL)	Assis *et al*. (2020)
Form fator	$4\pi AR / PO$	Więcaszek *et al*. (2020)

Source: Self elaboration.
Note: AR = Otolith area (mm); CO = Otolith length (mm); PO = Otolith perimeter (mm); AL = Otolith height (mm).

Determination of trace metals in otoliths

Before otolith digestion, 10 ml of 5% H_2O_2 was added to the vial containing the otoliths for about 5 minutes to remove any remaining surface tissue. The vials were then rinsed with quartz double-distilled water (Q water) and transferred to 50 ml storage tubes. The otoliths were subsequently dried in a laminar flow hood, weighed, and placed in beakers with 1 ml of 1% HNO_3, 1 ml of nitric acid (50%), and 8 ml of water added following the analytical methodology of the U.S Environmental Protection Agency (US EPA Method 3050B). The resulting solutions were then transferred to clean vials for later analysis. The digestion was then accelerated in a digestion block (ECO 16, VELP Scientifca) for 2 h at 120ºC.

The concentrations of aluminum (Al), antimony (Sb), arsenic (As), barium (Ba), cadmium (Cd), chromium (Cr), mercury (Hg), lead (Pb), and nickel (Ni), along with essential elements like boron (B), selenium (Se), silica (Si), phosphorus (P), copper (Cu), iron (Fe), calcium (Ca), potassium (K), magnesium (Mg), manganese (Mn), molybdenum (Mo), sodium (Na), vanadium (V), cobalt (Co), tin (Sn), and zinc (Zn), were quantified using an Inductively Coupled Plasma Optical Emission Spectrometer (SHIMADZU, ICPE-9000, Kyoto, Japan). Analytical quality control procedures were implemented throughout the study, with blank samples processed alongside test samples to ensure accuracy.

Detection limits were set as follows: 0.01 mg/L for Al, Au, Hg, Ca, Pb, S, and Si; 0.02 mg/L for As, B, Ba, Cu, Cr, Fe, K, Mg, Mn, Na, Ni, Se, Sn, and Zn; and 0.03 mg/L for Cd, Co, In, Mo, P, Sb, and V. Results were compared with the National Environmental Council Resolution No. 357/2005

(CONAMA, 2005), the World Health Organization standards (WHO, 2017), and Environmental Protection Agency guidelines (USEPA, 2000).

Data processing and statistical analysis

All biometric data from the samples, including the number of animals, weight (g), total length (cm), and standard length (cm), were recorded in electronic spreadsheets (Excel® 2016). Morphological and morphometric data of the otoliths were also documented and properly identified. The data were then organized into digital files for statistical analysis. To assess significant differences, Analysis of Variance (ANOVA) was used. Before this, normality was confirmed using the Kruskal-Wallis test. All statistical analyses were conducted using R software (Version 4.4.0).

RESULTS AND DISCUSSIONS

Otolith shape

The average weight, length, width, height, and perimeter of the *Psectrogaster amazonica* otoliths showed little variation (Table 2). Regarding the weight of the otoliths, the left otolith was slightly heavier than the right. These data are consistent with those observed by Nguyen and Dinh (2020), who found that fish of the species *Glossogobius sparsipapillus* had heavier left otoliths. However, in another study by Bhakta *et al*. (2020) with 184 specimens of *Otolithoides pama*, no differences were observed in the weight of the left and right otoliths. Therefore, further studies on *P. amazonica* are needed.

Table 2 – Biometric information from *Psectrogaster amazonica* otoliths

	WRO (g)	WLO (g)	LRO (mm)	LLO (mm)	WIRO (mm)	WILO (mm)	ARO (mm²)	ALO (mm²)	PRO (mm)	PLO (mm)
Range	0.0107-0.0190	0.0108-0144	4.9-6.2	4.9-6.4	4.0-5.0	4-4.8	1.5-2.1	1.6-2	1.5-2.1	1.5-2.0
Average	0.0145	0.0183	5.56	5.63	4.44	4.46	1.82	1.80	1.72	1.72
Standard Deviation	0.0021	0.0210	0.30	0.35	0.24	0.21	0.21	0.12	0.12	0.11

Source: Self elaboration.

Note: WRO = weight right otolith; WLO = weight left otolith; LRO = length right otolith; LLO = length left otolith; WIRO = width right otolith; WILO = width left otolith; ARO = area right otolith; ALO = area left otolith; PRO = perimeter right otolith; PLO = perimeter left otolith.

There were no significant differences between left and right otoliths in terms of length, which corroborates with Bhuiya *et al*. (2022), who did not observe any length variation between left and right otoliths in their study of *Otolithoides pama*. Otolith morphometric data aid in species identification, predator behavior, dietary preferences, and insights into age, growth, and habitat use, supporting conservation and fisheries management. Species identification is performed by comparing the age, size of the fish, and otolith morphology (ANKITA; KHAN, 2022). The otolith biometric data of *P. amazonica* is valuable for the Middle Tocantins River region, supporting species identification and ecological assessment at this biome interface.

The shape indices obtained represent novel data for *P. amazonica* in the study region (Table 3). These indices are used to identify species through the analysis of otolith shapes using mathematical calculations, helping to assess the population of animals present in the wild (OSMAN *et al*., 2021). All the shape indices of *P. amazonica* differed from *Sphyraena sphyraena* in a study conducted by Yedier (2021) in Turkey, in the Mediterranean Sea. They also differed from all indices of species such as *Trachurus mediterraneus*, *Merlangius merlangus*, and *Rhodeus amarus* (SAYGIN *et al*., 2020; BOSTANCI *et al*., 2024). Therefore, it is observed that the species analyzed in the present study has otolithic characteristics that ensure its differentiation from other species.

Table 3 – Shape indices of *Psectrogaster amazonica* otoliths from the Middle Tocantins River

Index	Average	Range
Roundness	0.093	0.080 - 0.114
Circularity	0.018	0.015-0.021
Rectangularity	0.078	0.077-0.083
Ellipticality	0.14	0.14-0.15
Form factor	0.092	0.091-0.095

Source: Self elaboration.

Microchemical composition of otoliths

Among the elements analyzed in this study, Al (0.823 mg/L), V (0.382 mg/L), and Cu (0.035 mg/L) displayed average concentrations exceeding certain national and international standards (Table 4). The iron (Fe) levels were within normal limits, with alterations reported above 2.000.000 mg/L (TASLIMA *et al*., 2022). Mercury (Hg) levels (0.027 mg/L) were also within normal limits, aligning with findings from studies like Butler *et al*. (2022), which examined Hg concentrations in Lates calcarifer and noted an increase in mercury with age. This is encouraging, as mercury contamination in the Amazon basin has historically been high due to artisanal gold mining since the last century.

Table 4 – Comparison of otolith microchemical concentrations in *Psectrogaster amazonica* with national and international standards

Elements	Average (mg/L)	National	International	
		CONAMA (mg/L, in the water)	WHO (mg/L, in the water)	USEPA (mg/L, in the water)
Calcium (Ca)	126.2 ± 39.89	-	-	-
Aluminum (Al)	*0.823 ± 0.248	0.1	0.1–0.2	-
Vanadium (V)	*0.382 ± 0.173	0.1	-	-
Potassium (K)	0.368 ± 0.156	-	-	-
Copper (Cu)	*0.035 ± 0.013	0.009	0.025	0.013
Iron (Fe)	0.034 ± 0.010	0.3	0.3	-
Magnesium (Mg)	0.233 ± 0.121	0.1	0.1	-
Indian (In)	0.179 ± 0.23	-	-	-
Gold (Au)	0.105 ± 0.015	-	-	-
Tin (Sn)	0.043 ± 0.030	-	-	-
Mercury (Hg)	0.027 ± 0.014	0.002	0.006	0.002
Manganese (Mn)	0.018 ± 0.019	0.5	-	-

Source: Self elaboration. Adapted from CONAMA (2005); WHO (2017); USEPA (2000).
Note: *Parameter concentrations above the quality standards of some regulations in comparison.

The microchemical composition of fish otoliths can reveal levels of environmental contamination and allow for the study of species movement. Regarding the composition of the otoliths, the concentrations of calcium, as expected, were high due to the otolith being primarily composed of calcium carbonate (MALLIK *et al*., 2023). Although there is a lack of studies on trace metal concentrations in fish, it has already been shown that Mn, Cu, and Mg affect the body development of fish (HÜSSY *et al*., 2021). The concentrations of trace metals in otoliths of *P. amazonica* are unprecedented for the Tocantins region of Maranhão, making comparisons impossible. Therefore, there is a need for further studies to compare metal concentrations at different life stages of fish.

The aluminum results are particularly noteworthy (0.823 mg/L in otoliths) (Table 5), as studies along the banks of the Tocantins River have shown elevated levels in both water (0.69 mg/L, annual average) and sediments (254.97 mg/kg, annual average) (ACIOLY *et al*., 2024; ACIOLY *et al*., 2024). Copper also showed elevated values in water (0.05 mg/L, annual average) and sediments (1.32 mg/kg, annual average). Various trace metals, including Ba, Cu, Co, Hg, Fe, Mg, Mn, Ni, Zn, and some rare earth elements, can be incorporated into the carbonate structure of otoliths, influenced by the concentration of these elements in the surrounding water. Ongoing research is essential to ensure public safety regarding contaminant levels and to assess the impact of rising urbanization. The data from these studies will support future analysis and guide conservation efforts.

Fish otolith chemistry has been used, particularly focusing on trace metal concentrations, to precisely determine fish habitats, home ranges, and migration patterns across water bodies with distinct trace metal signatures (HÜSSY *et al*., 2024; WINDOM; SAVIDGE, 2024; RANA *et al*., 2024). Otoliths consist of a complex mixture of three carbonate polymorphs—calcite, aragonite, and vaterite—with aragonite being dominant, each with its crystalline structure. Magnesium concentrations in otoliths often signal the presence of vaterite, the magnesium carbonate polymorph (GAULDIE, 1993, 1996). These crystal structures offer multiple sites, including structural defects, that can incorporate additional trace metals in response to environmental concentrations, as observed in otoliths (WINDOM; SAVIDGE, 2024; PONTUAL *et al*., 2024). Additionally, trace metals may substitute for the primary divalent cation in one of the carbonate polymorphs that make up the otolith.

Information from otoliths and other calcified structures is increasingly valuable for sustainable resource management, providing insights often unavailable from sensors or data tags (REIS-SANTOS *et al*., 2023). Contaminants like selenium have also been analyzed in otoliths across various species and ecosystems to assess levels of exposure. Rana *et al*. (2024) applied otolith microchemistry to Bangana dero, observing high bioaccumulation factors for Se in the post-monsoon period and for Hg in the pre-monsoon period. They further suggest that Bangana dero otoliths could serve as effective long-term monitoring tools for environmental assessments, aiding in historical exposure reconstructions to protect water bodies.

Studies indicate that detecting excessive or sublethal contaminant levels in biominerals is a valuable approach for identifying sources, pathways, and cumulative exposure risks, thereby supporting aquatic conservation efforts (BOULAJFENE, 2024; JOHNSON *et al*., 2020; LEONHARD; AGIADI, 2023). However, although various metal contaminants have been measured in fish otoliths from polluted areas, elements like mercury and complex compounds are rarely detected, partly due to limitations in ICP-MS techniques (REIS-SANTOS *et al*., 2023). Since otoliths can reflect both recent and historic exposure to pollutants, their analysis could offer a temporal record of environmental quality, aiding in assessing long-term pollution trends.

CONCLUSION

This study represents an important contribution to understanding the environmental quality of the middle Tocantins River and the role of *P. amazonica* otoliths as biomarkers of trace metal pollution. The morphometric analysis confirmed the reliability and precision of otolith measurements, showing minimal differences between the left and right structures. These results align with previous studies, highlighting the robustness of otolith morphometry for environmental assessments.

The detection of elevated concentrations of aluminum, vanadium, and copper in the otoliths underscores the presence of anthropogenic contamination in the region. The findings not only establish *P. amazonica* as a potential environmental biomarker but also reveal the necessity for further studies to monitor and mitigate pollution impacts in Maranhão's aquatic ecosystems.

The results are particularly significant given the scarcity of data on trace metal bioaccumulation in the middle Tocantins River, and they provide a foundation for future research on the ecological health of this critical waterway. Ongoing studies focusing on the different life stages of *P. amazonica* and other native species are essential to develop comprehensive conservation strategies and inform sustainable management practices.

Ultimately, this study highlights the dual utility of otoliths in ecological and toxicological research, serving as precise sclerochronological biomarkers. These insights not only advance the understanding of trace metal pollution dynamics but also underscore the importance of integrating biomarker-based approaches in aquatic conservation and fisheries management.

REFERÊNCIAS

ACIOLY, T. M. D. S. *et al*. "Levels of Potentially Toxic and Essential Elements in Water and Estimation of Human Health Risks in a River Located at the Interface of Brazilian Savanna and Amazon Biomes (Tocantins River)". **Toxics**, vol. 12, n. 7, 2024.

ACIOLY, T. M. S. *et al*. "Levels of potentially toxic and essential elements in Tocantins River sediment: health risks at Brazil's Savanna-Amazon interface". **Scientific Reports**, vol. 14, n. 1, 2024.

AGIADI, K. *et al*. "The taphonomic clock in fish otoliths". **Paleobiology**, vol. 48, 2021.

ALVARIÑO, L. *et al*. "Assessment of potentially toxic metals, metalloids, and non-metals in muscle and liver tissue of Two Fish Species (*Mugil cephalus* Linnaeus, 1758 and *Odontesthes regia* (Humboldt, 1821) from the Coastal Area of Callao, Peru". **Regional Studies in Marine Science**, vol. 71, 2024.

ANDRADE, H. *et al*. "Ontogenetic movements of cod in Arctic fjords and the Barents Sea as revealed by otolith microchemistry". **Polar Biology**, vol. 43, 2020.

ANKITA, M.; KHAN, S. "Relationships between length-weight, length-length, and fish length to otolith morphometry in *Rita rita* (Hamilton, 1822)". **Zoology and Ecology**, vol. 32, n. 1, 2022.

AVIGLIANO, E. *et al*. "Otolith Sr/Ca ratio complements Sr isotopes to reveal fish migration in large basins with heterogeneous geochemical landscapes". **Environmental Biology of Fishes**, vol. 104, 2021.

AVIGLIANO, E. *et al*. "Population structure and ontogenetic habitat use of Micropogonias furnieri in the Southwestern Atlantic Ocean inferred by otolith chemistry". **Fisheries Research**, vol. 240, 2021.

AVIGLIANO, E.; VOLPEDO, A. V. “A Review of the Application of Otolith Microchemistry Toward the Study of Latin American Fishes”. **Reviews in Fisheries and Aquaculture**, vol. 24, 2016.

BHAKTA, D. *et al*. “Relationship between otolith morphometry and fish size of *Otolithoides pama* (Hamilton, 1822) from Hooghly-Matlah estuary, India”. **Indian Journal of Geo Sciences**, vol. 49, n. 10, 2020.

BHUIYA, A. *et al*. “Size frequency, length-weight relationships, condition factor and relationships between fish size and otolith dimensions in the Pama croaker *Otolithoides pama* (Hamilton, 1822) from Lower Meghna River Estuary, Bangladesh”. **Lakes amd Reservoirs: Research and Management**, vol. 27, n. 3, 2022.

BOSTANCI, D. *et al*. “Otolith morphometry and scanning electron microscopy analysis of three fish species from the Black Sea”. **Acta Biologica Turcica**, vol. 37, n. 1, 2024.

BOULAJFENE, W. “Toxicity Testing Using Marine Mollusks' Larvae”. *In*: LARVAE, M. **Developments applications**. London: CRC Press. 2024.

BOYLE, R. “Otolith adaptive responses to altered gravity”. **Neuroscience and Biobehavioral Reviews**, vol. 122, 2021.

BUTLER, E. C. V. *et al*. “Influence of life history variation and habitat on mercury bioaccumulation in a high-order predatory fish in tropical Australia”. **Environmental Research**, vol. 212, n. 113152, 2022.

CAMPANA, S. E. *et al*. “Otolith Microstructure Examination and Analysis”. **Canadian Special Publication of Fisheries and Aquatic Sciences**, vol. 117, 1992.

CAVALCANTE, A. M. S. *et al*. “Determinação de metais pesados (Fe, Mn, Cd, Cu, Co, Cr, Ni, Zn e Pb), matéria orgânica e carbono orgânico no sedimento de áreas de manguezal NO Município de Vigia De Nazaré, NE–PA”. **Holos Environment**, vol. 21, n. 1, 2021.

CHEN, S.; DING, Y. “Tackling heavy metal pollution: evaluating governance models and frameworks”. **Sustainability**, vol. 15, n. 22, 2023.

CONAMA - Conselho Nacional do Meio Ambiente. **Dispõe sobre a Classificação dos Corpos de Água e Diretrizes Ambientais para o seu enquadramento, bem como estabelece as condições e padrões de lançamento de efluentes, e dá outras providências**. Brasília: CONMA, 2005. Available in: <www.mma.gov.br>. Access in: 22/09/2024.

CONAMA. Conselho Nacional do Meio Ambiente. **Resolução n. 357, de 17 de março de 2005**. Brasília: CONMA, 2005. Available in: <www.mma.gov.br>. Access in: 22/09/2024.

CONAMA. Conselho Nacional do Meio Ambiente. **Resolução n. 454, de 01 de novembro de 2012**. Brasília: CONMA, 2012. Available in: <www.mma.gov.br>. Access in: 22/09/2024.

CURTHOYS, I. S. “The Anatomical and Physiological Basis of Clinical Tests of Otolith Function. A Tribute to Yoshio Uchino”. **Frontiers in Neurology**, vol. 11, 2020.

D’IGLIO, C. *et al*. “Intra- and interspecific variability among congeneric Pagellus otoliths”. **Scientific Reports**, vol. 11, 2021.

D'IGLIO, C. *et al*. "Otolith Analyses Highlight Morpho-Functional Differences of Three Species of Mullet (Mugilidae) from Transitional Water". **Sustainability**, vol. 14, 2021.

FEY, D. P. *et al*. "Effects of temperature on somatic growth, otolith growth, and uncoupling in the otolith to fish size relationship of larval northern pike, Esox lucius L". **Fisheries Research**, vol. 236, 2021.

GAGLIANO, M. *et al*. "Feeding history influences otolith shape in tropical fish". **Marine Ecology Progress Series**, vol. 278, 2004.

GAULDIE, R. W. "Effects of temperature and vaterite replacement on the chemistry of metal ions in the otoliths of *Oncorhynchus tshawytscha*". **Canadian Journal of Fisheries and Aquatic Sciences**, vol. 53, n. 9, 1996.

GAULDIE, R.W. "Polymorphic crystalline structure of fish otoliths". **Journal of Morphology**, vol. 218, 1993.

GHANI, J. *et al*. "Multivariate statistical methods and GIS-based evaluation of potable water in urban children's parks due to potentially toxic elements contamination: a children's health risk assessment study in a developing country". **Sustainability**, vol. 15, 2023.

GUT, C. *et al*. "Identification of past and present gobies: distinguishing Gobius and Pomatoschistus (Teleostei: Gobioidei) species using characters of otoliths, meristics and body morphometry". **Contributions to Zoology**, vol. 89, 2020.

HERMANN, T. W. *et al*. "Harnessing the potential for otolith microchemistry to foster the conservation of Amazonian fishes". **Aquatic Conservation: Marine and Freshwater Ecosystems**, vol. 31, 2021.

HOFF, N. T. *et al*. "Past and contemporaneous otolith fingerprints reveal potential anthropogenic interferences and allows refinement of the population structure of *Isopisthus parvipinnis* in the South Brazil Bight". **Biology**, vol. 11, n. 7, 2022.

HUANG, Y. F. *et al*. "Ontogenetic development, allometric growth patterns, and daily increment validation of larvae and juvenile Culter alburnus". **Environmental Biology of Fishes**, vol. 104, 2021.

HÜSSY, K. *et al*. "Into the wild: coupling otolith and archival tag records to test assumptions underpinning otolith chemistry applications in wild fish". **Frontiers in marine science**, vol. 11, 2024.

HÜSSY, K. *et al*. "Trace element patterns in otoliths: the role of biomineralization". **Reviews in Fisheries Science and Aquaculture**, vol. 29, n. 4, 2021.

IBGE – Instituto Brasileiro de Geografia e Estatística. **Cidades e estados**. Rio de Janeiro: IBGE, 2022. Available online: <www.ibge.govol.br>. Access in: 23/09/2024.

JAWAD, L. *et al*. "Fluctuating Asymmetry in Asteriscii Otoliths of Common Carp (*Cyprinus carpio*) Collected from Three Localities in Iraqi Rivers Linked to Environmental Factors". **Fishes**, vol. 7, n. 2, 2022.

JOHNSON, R. C. *et al*. "Lifetime chronicles of selenium exposure linked to deformities in an imperiled migratory fish". **Environmental Science and Technology**, vol. 54, n. 5, 2020.

KALISH, J. M. “Otolith microchemistry: validation of the effects of physiology, age and environment on otolith composition”. **Journal of Experimental Marine Biology and Ecology**, vol. 132, 1989.

KHAWAR, M. *et al*. “Trace metals and nutrient analysis of marine fish species from the Gwadar coast”. **Scientific Reports**, vol. 14, n. 1, 2024.

KIKUCHI, E. *et al*. “Using growth rates and otolith shape to identify the population structure of *Umbrina canosai* (Sciaenidae) from the Southwestern Atlantic”. **Marine Biology Research**, vol. 17, 2021.

KOEBERLE, A. L. *et al*. “Fluctuating asymmetry of adult Chinook Salmon (*Oncorhynchus tshawytscha*) otoliths from wild and hatchery origins”. **Aquatic Ecology**, vol. 54, 2019.

KOROSTELEV, N. B. *et al*. “Micro- and Ultramicroelemental Content in Otoliths of Blue Antimora Antimora rostrata and Pacific Flatnose *A. microlepis* (Moridae, Teleostei)”. **Oceanology**, vol. 60, 2021.

KUMKAR, P. *et al*. “Contaminants and their ecological risk assessment in beach sediments and water along the Maharashtra coast of India: A comprehensive approach using microplastics, heavy metal (loid) s, pharmaceuticals, personal care products and plasticisers”. **Science of the Total Environment**, vol. 892, 2023.

LEONHARD, I.; AGIADI, K. “Addressing challenges in marine conservation with fish otoliths and their death assemblages”. *In*: NAWROT, R. *et al*. **Conservation Palaeobiology of Marine Ecosystems**. London: Geological Society, 2023.

LOEPPKY, A. R. *et al*. “Influence of ontogenetic development, temperature, and pCO2 on otolith calcium carbonate polymorph composition in sturgeons”. **Scientific Reports**, vol. 11, 2021.

LOMARTIRE, S. *et al*. “Biomarkers based tools to assess environmental and chemical stressors in aquatic systems”. **Ecological Indicators**, vol. 122, 2021.

LORENC, W. *et al*. “LC/ICP-MS and complementary techniques in bespoke and nontargeted speciation analysis of elements in food samples”. **Mass Spectrometry Reviews**, vol. 41, 2020.

MACDONALD, J. I. *et al*. “Isolating the influence of ontogeny helps predict island-wide variability in fish otolith chemistry”. **Reviews in Fish Biology and Fisheries**, vol. 30, 2019.

MACIEL, T. R. *et al*. “Integrated use of otolith shape and microchemistry to assess Genidens barbus fish stock structure”. **Estuarine, Coastal and Shelf Science**, vol. 261, 2021.

MADADI, M. *et al*. “Using Amaranthus green proteins as universal biosurfactant and biosorbent for effective enzymatic degradation of diverse lignocellulose residues and efficient multiple trace metals remediation of farming lands”. **Journal of Hazardous Materials**, vol. 406, 2021.

MAHÉ, K. *et al*. “Directional bilateral asymmetry in fish otolith: A potential tool to evaluate stock Boundaries?”. **Symmetry**, vol. 13, 2021.

MALLIK, A. *et al*. “Fish Otolith Microchemistry as a Biomarker of Metal Pollution in the Estuarine Ecosystem”. **Science of The Total Environment**, vol. 807, n. 3, 2023.

MARTINO, J. C. *et al*. “Corrigendum to: Identifying physiological and environmental influences on otolith chemistry in a coastal fishery species”. **Marine and Freshwater Research**, vol. 72, 2021.

MEJRI, M.; T. *et al*. “Fluctuating asymmetry in the otolith shape, length, width and area of Pagellus erythrinus collected from the Gulf of Tunis”. **Cahiers de Biologie Marine**, vol. 61, 2020.

MESA, M. L. *et al*. “Comparative analysis of otolith morphology in icefishes (Channichthyidae) applying different statistical classification methods”. **Fisheries Research**, vol. 230, 2020.

MOORE, B. R. *et al*. “Otolith shape as a tool for species identification of the grenadiers *Macrourus caml* and *M. whitsoni*”. **Fisheries Research**, vol. 253, 2022.

MORAES, J. N. **Caracterização cromossômica de seis espécies de peixes da família Curimatidae (Characiformes) da Amazônia** (Dissertação de Mestrado em Genética, Conservação e Biologia Evolutiva) Manaus: INPA, 2021.

MORALES, C. J. *et al*. “Otolith Morphometric and Shape Distinction of Three Redfin Species under the *Genus Decapterus* (Teleostei: Carangidae) from Sulu Sea, Philippines”. **Fishes**, vol. 8, 2023.

MORISSETTE, O. *et al*. “Quantifying migratory capacity and dispersal of the invasive tench (*Tinca tinca*) in the St. Lawrence River using otolith chemistry”. **Canadian Science Publishing**, vol. 78, 2021.

NEVES, J. *et al*. “Comparing otolith shape descriptors for population structure inferences in a small pelagic fish, the European sardine *Sardina pilchardus* (Walbaum, 1792)”. **Journal of Fish Biology**, vol. 102, 2023.

NGUYEN, T.; DINH, Q. “Otolith dimensions and their relationship with the size of *Glossogobius sparsipapillus* fish along the coastline of Mekong Delta, Vietnam”. **Egyptian Journal of Aquatic Biology and Fisheries**, vol. 24, n. 2, 2020.

NYGAARD, T. F. *et al*. “Environmental Adaptation of Robot Morphology and Control Through Real-World Evolution”. **Evolutionary Computation**, vol. 29, 2021.

ODUM, H. T. “The Stability of the World Stronium Cycle”. **Science**, vol. 114, 1951.

ONYENA, A. P. *et al*. “Engaging one health in heavy metal pollution in some selected Nigerian Niger delta cities. A Systematic review of pervasiveness, bioaccumulation and subduing environmental health challenges”. **Biological Trace Element Research**, vol. 202, n. 4, 2024.

ORDOÑEZ, A. *et al*. “Automatic Fish Age Determination across Different Otolith Image Labs Using Domain Adaptation”. **Fishes**, vol. 7, 2022.

OSMAN, Y. A. A. *et al*. “Relationship between body and otolith morphological characteristics of sabre squirrelfish (*Sargocentron spiniferum*) from the southern Red Sea: Difference between right and left otoliths”. **Oceans**, vol. 2, n. 3, 2021.

PEREIRA, D. P. **Avaliação de impactos ambientais no Lago Açu (Área de proteção ambiental da Baixada Maranhense e Sítio Ramsar com a aplicação de biomarcadores em espécies nativas** (Dissertação de Mestrado em Recursos Aquáticos e Pesca) São Luís: UEMA, 2019.

PEREIRA, D. V. *et al*. “Vulnerability to overfishing of fish stocks in the Amazon Basin”. **Fisheries Research**, vol. 265, 2023.

PHILLIPS, A. A. *et al*. “Sulfur isotope analysis of cysteine and methionine via preparatory liquid chromatography and elemental analyzer isotope ratio mass spectrometry”. **Rapid Communications in Mass Spectrometry**, vol. 35, 2020.

POLITIKOS, D. V. *et al*. “Automating fish age estimation combining otolith images and deep learning: The role of multitask learning”. **Fisheries Research**, vol. 242, 2021.

PONTUAL, H. *et al*. “Heterogeneity of otolith chemical composition from two-dimensional mapping: Relationship with biomineralization mechanisms and implications for microchemistry analyses”. **Journal of Fish Biology**, vol. 104, n. 1, 2024.

RAHNMAMA, B. *et al*. “The use of different otolith-shape analyses for stock discrimination of Yellowtail Snapper Ocyurus chrysurus, (Bloch, 1791) in the coastal waters of northeastern Brazil”. **International Journal of Aquatic Biology**, vol. 11, 2023.

RANA, A. *et al*. “Otolith microchemistry of freshwater indigenous minor carp (Bangana dero) as a biomonitoring tool to trace heavy metals in River Sutlej, Ropar Wetland (Ramsar site), Punjab, India”. **Environmental Monitoring and Assessment**, vol. 195, n. 11, 2023.

REIS-JÚNIOR, J. *et al*. **Otólitos**: As pedras preciosas dos peixes. Aracajú: Criação Editora, 2023.

REIS-SANTOS, P. *et al*. “Reading the biomineralized book of life: expanding otolith biogeochemical research and applications for fisheries and ecosystem-based management”. **Reviews in Fish Biology and Fisheries**, vol. 33, n. 2, 2023.

SANTOS, J. A. *et al*. “Otolith shape analysis supports three cryptic species in the *Stellifer punctatissimus* complex (Acanthuriformes: Sciaenidae)”. **Neotropical Ichthyology**, vol. 20, 2022.

SANTOS, K. F. S. *et al*. Morfometria e morfologia de otólitos de peixes no litoral do estado do Maranhão, Brasil. **Brazilian Journal of Biology**, vol. 84, 2024.

SAYGIN, S. *et al*. “Otolith Shape Analysis and the Relationships between Otolith Dimensions–Total Length of European Bitterling, Rhodeus amarus (Cyprinidae) Sampled from Samsun Province, Turkey”. **Journal of Icthyology,** vol. 60, 2020.

SILVA, A. R. “Análise da paisagem em um afluente do rio Tocantins no município de Imperatriz, Maranhão”. **Revista Contexto Geográfico**, vol. 7, n. 15, 2022.

SMOLINSKI, S. *et al*. “Assessing the performance of statistical classifiers to discriminate fish stocks using Fourier analysis of otolith shape”. **Canadian Science Publishing**, vol. 77, 2019.

SOUZA, A. T. *et al*. “Ontogenetic and interpopulation differences in otolith shape of the European perch (Perca fluviatilis)”. **Fisheries Research**, vol. 230, 2020.

SPICH, K.; FEY, D. P. “Using otolith microstructure analysis in studies on the ecology of the early life stages of cod, *Gadus morhua* L.: A review”. **Fisheries Research**, vol. 250, 2022.

SPILSBURY, F. *et al*. “Multivariate analysis of otolith microchemistry can discriminate the source of oil contamination in exposed fish”. **Comparative Biochemistry and Physiology Parte C: Toxicology and Pharmacology**, vol. 254, 2022.

TASLIMA, K. *et al*. "Impacts of heavy metals on early development, growth and reproduction of fish– A review". **Toxicology reports**, vol. 9, 2022.

USEPA - US Environmental Protection Agency. **National Primary Drinking Water Regulations**. Washington: Environmental Protection Agency, 2000.

VASCONCELOS, J. *et al*. "Choosing wavelet methods for otolith contour studies". **Reviews in Fish Biology and Fisheries**, vol. 24, n. 1, 2024.

VIGNON, M.; MORAT, F. "Environmental and genetic determinant of otolith shape revealed by a non-indigenous tropical fish". **Marine Ecology Progress Series**, vol. 411, 2010.

VRDOLJAK, D. *et al*. "Otolith fingerprints reveals potential pollution exposure of newly settled juvenile Sparus aurata". **Marine Pollution Bulletin**, vol. 160, 2020.

VU, A. V. *et al*. "Life history strategies of Mekong pangasiid catfishes revealed by otolith microchemistry". **Fisheries Research**, vol. 249, 2022.

WAGNER, A. R. *et al*. "Impact of Canal-Otolith Integration on Postural Control". **Frontiers in Integrative Neuroscience**, vol. 15, 2021.

WANG, R. *et al*. "Design and implementation of a jellyfish otolith-inspired MEMS vector hydrophone for low-frequency detection". **Microsystems and Nanoengineering**, vol. 7, 2021.

WHO - World Health Organization. "Guidelines for Drinking-Water Quality". Geneva: WHO, 2017. Available in: <www.who.int>. Access in: 23/09/2024.

WHO - World Health Organization. **Guidelines for drinking-water quality**: recommendations. Geneva: WHO, 2004. Available in: <www.who.int>. Access in: 23/09/2024.

WIĘCASZEK, B. *et al*. "Fish size effect on sagittal otolith outer shape variability in round goby *Neogobius melanostomus* (Pallas 1814)". **Journal of Fish Biology**, vol. 97, n. 5, 2020.

WINDOM, H. L.; SAVIDGE, W. B. "Sources and transport pathways of trace metals to the outer continental shelf off South Carolina and Georgia, USA revealed from the otoliths of moray eels". **Continental Shelf Research**, vol. 282, 2024.

WOOD, R. S. *et al*. "Quantifying fish otolith mineralogy for trace-element chemistry studies". **Scientific Reports**, vol. 12, 2022.

YEDIER, S. "Otolith shape analysis and relationships between total length and otolith dimensions of European barracuda, *Sphyraena sphyraena* in the Mediterranean Sea". **Iranian Journal of Fisheries Sciences**, vol. 20, n. 4, 2021.

YEDIER, S. *et al*. "Morphological and morphometric features of the abnormal and normal saccular otoliths in flatfishes". **The Anatomical Record**, vol. 306, 2022.

ZISCHKE M. T. *et al*. "Otolith morphology of four mackerel species (*Scomberomorus spp*.) in Australia: Species differentiation and prediction for fisheries monitoring and assessment". **Fisheries Research**, vol. 176, 2016.

BOLETIM DE CONJUNTURA (BOCA)

Ano VI | Volume 19 | Nº 57 | Boa Vista |2024

http://www.ioles.com.br/boca