










Analysis of the stomach contents of *Arapaima gigas* in the Cautário River Basin, Rondônia, Brazil

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ABSTRACT

Arapaima gigas is the largest scaled freshwater fish in the world and can grow to more than 3 meters in length and exceed 252 kg. This species naturally inhabits the flooded areas of the Amazon basin, but it has been found outside this region as a result of intentional or accidental human actions. In this study, we analyzed the items consumed by the *A. gigas* distributed in the Cautário River, a tributary of the Guaporé River Basin (Rondônia, Brazil), in which pirarucu is not native and requires actions for its eradication. Experimental fisheries were conducted from August 2023 to September 2024. A total of 128 *A. gigas* were captured (ranging from 1.16 to 2.26 m, with a mean of 1.76 ± 0.31 m, and weight of 24.70 to 111.60 kg, with a mean of 52.37 ± 22.54 kg) and had their stomachs analyzed. It was observed that 104 stomachs were full and 24 were empty. In the stomachs, five categories of food items were classified: fish and fish remains, plant material, debris, crustaceans, and worms. The items with the highest absolute frequency were plant materials (82), fish and fish remain (51), worms (32), and crustaceans (1). It was concluded that the *A. gigas* of the Cautário River Basin feed mainly on fish, but with a large participation in the diet of plant materials, which may be an indication of involuntary ingestion due to the animal's voracity or possible onset of food shortages in the study region.

Keywords: *Arapaima gigas*; Guaporé River Basin; diet; invasive fish.

Análise do conteúdo estomacal de *Arapaima gigas* na Bacia do Rio Cautário, Rondônia, Brasil

RESUMO

Arapaima gigas é o maior peixe de água doce de escamas do mundo e pode chegar a mais de 3 metros de comprimento e ultrapassar 252 kg. Essa espécie habita naturalmente as áreas inundadas da bacia amazônica, no entanto foi encontrada fora da região como resultado de ações humanas intencionais ou acidentais. Neste estudo, analisamos os itens consumidos pelo *A. gigas* distribuídos no Rio Cautário, afluente da Bacia do Rio Guaporé (Rondônia, Brasil), no qual o pirarucu não é nativo e requer ações para sua erradicação. As pescarias experimentais foram realizadas no período de agosto de 2023 a setembro de 2024. Um total de 128 *A. gigas* foram capturados (variando de 1,16 a 2,26 m, com média de $1,76 \pm 0,31$ m, e peso de 24,70 a 111,60 kg, com média de $52,37 \pm 22,54$ kg) e tiveram seus estômagos analisados. Observou-se que 104 estômagos estavam cheios e 24 vazios. Nos estômagos, cinco categorias de itens alimentares foram classificadas: peixes e restos de peixes, material vegetal, detritos, crustáceos e vermes. Os itens com maior frequência absoluta foram materiais vegetais (82), peixes e restos de peixes (51), vermes (32) e crustáceos (1). Concluiu-se que os *A. gigas* da Bacia do Rio Cautário se alimentam principalmente de peixes, mas há grande participação na dieta de materiais vegetais, o que pode ser um indício de ingestão involuntária por causa da voracidade do animal ou de possível início de escassez de alimentos na região de estudo.

Palavras-chave: *Arapaima gigas*; Bacia do Rio Guaporé; dieta; peixes invasores.

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INTRODUCTION

The species *Arapaima gigas* (Schinz, 1822), known in Brazil as the pirarucu, presents characteristics that favor its commercial attractiveness and, consequently, its intense exploitation. Its remarkable growth rate, especially in the early stages, in which it reaches around 80 cm and a weight gain of up to 10 kg in the first year of life (Castello, 2008; Coutinho et al., 2019; Val & Almeida-Val, 2012), in addition to excellent meat yield compared to other Amazonian commercial fish species, and its excellent profitability are very attractive in the Brazilian market (Cavali et al., 2023; Imbiriba et al., 1994).

The fishing of this species is facilitated by its unique respiratory habit; since *A. gigas* needs to breathe atmospheric air, it rises to the surface at intervals ranging from 15 to 20 minutes, supplying up to 90% of its oxygen demand (Salvo-Souza & Val, 1990; Silva & Duncan, 2016). This behavior makes the species vulnerable to capture, as local fishermen know their patterns of ascent and descent to the surface of the water to breathe, taking advantage of these moments to capture the pirarucu (Veríssimo, 1895).

However, this ease of capture, associated with other factors, such as its high commercial value, resulted in overfishing of the species and the consequent decline of its natural populations in the Amazon basin (Allan et al., 2005). For this reason, *A. gigas* was included in the International Union for Conservation of Nature (IUCN) Red List in 1996 as an endangered species (Castello & Stewart, 2010). With the implementation of public policies and management strategies, populations have recovered, and the species was removed from this list in 2014. Despite this, *A. gigas* remains listed in the Convention on International Trade in Endangered Species of Wild Flora and Fauna (CITES), which regulates its export and import because of its commercial importance.

The farming of the species was developed to counter overfishing, which, as a consequence, later caused the introduction of *A. gigas* in several regions outside its natural area of occurrence. Its natural distribution occurs in the floodplain areas of the Araguaia-Tocantins and Solimões-Amazon River basins (Queiroz, 2000). However, in some basins of the state of Rondônia, Brazil, its invasive presence is mainly attributed to escapes from breeding grounds in the southern Peruvian Amazon (Doria et al., 2021; Miranda-Chumacero et al., 2012). Thus, one of the most emblematic situations for invasive fish species in the Amazon Basin has been consolidated, whereby, depending on the region, the species can be considered either native or invasive (Catâneo et al., 2022).

In Rondônia, *A. gigas* has spread as an invasive species in different habitats, occupying medium and large rivers and streams, configuring a conservation paradox, because while it remains threatened in its original habitat, it becomes abundant and problematic in non-native areas (Catâneo, 2019; Catâneo et al., 2022).

Among the regions in Rondônia in which *A. gigas* is considered invasive, the Guaporé River stands out for the number of records of appearances and captures. This river and its tributaries form a complex and sensitive ecosystem, which is composed of Cerrado, dense forests, floodplain areas, streams, and river islands that provide a habitat for a rich diversity of fish (Penha et al., 2017). The Cautário River, located in the northeast of the Guaporé sub-basin, is one of the most relevant tributaries, and is part of the Extractive Reserve of the Cautário River, created in 1990 to guarantee the sustainable exploitation and conservation of renewable natural resources (Brasil, 2017).

When introduced outside its natural range, *A. gigas* can negatively impact native fish communities, especially in resource-limited environments in which competitive interactions are more intense (Rejas et al., 2025; Sousa et al., 2022; Vitule et al., 2019). Its trophic position as a top predator suggests additional risks, since it regulates the stability of ecosystems through predation, with a predominantly piscivorous diet (Figueiredo, 2013). The introduction of exotic predators tends to alter prey community structures, intensify competition for resources, and unbalance populations of foraging fish (Beisner et al., 2003; Britton, 2023).

These impacts can lead to a reduction or even local extinction of native species due to habitat changes, competition, predation, genetic degradation and/or the spread of pathogens and parasites (Agostinho & Júlio Júnior, 1996; Outa et al., 2019). Such disturbances also affect trophic dynamics, promoting changes in the diet of native species and in energy flow, especially in small species (Flood et al., 2025).

In this sense, trophic plasticity is recognized as one of the main and most successful strategies of invasive freshwater fish species, allowing them to exploit different food resources and adapt quickly to new environments (Britton, 2023). This characteristic has contributed significantly to the loss of global freshwater fish biodiversity, highlighting the central role of biological invasions as one of the main factors in this decline (Dudgeon et al., 2006). In addition, insufficient control measures exacerbate the negative impacts of invasions, making it difficult for affected ecosystems to recover (Dudgeon & Strayer, 2025). As a consequence, the effects of invasions on freshwater environments become especially severe and multifaceted,

altering both the structure of communities and the functioning of ecosystems (Ricciardi & MacIsaac, 2011).

Furthermore, understanding the feeding behavior of predatory fish is fundamental to understand their somatic and reproductive growth, which are crucial aspects for trophic ecology and interspecific interactions, especially those related to predation and competition (Amundsen & Sánchez-Hernandez, 2019). In this sense, the analysis of stomach contents is an essential tool for describing eating habits and provides knowledge for fisheries management (Zacharia, 2014), offering valuable information on eating patterns and trophic relationships in the environments in which they live (Manko, 2016).

Therefore, this study evaluated the food resources consumed by populations of invasive *A. gigas* in the floodplain areas of the Cautário River Basin, to identify their food preferences in this region of Rondônia. It was intended to provide support for the

management of the eradication of the invasive species, as well as provide fisheries managers with robust information on the food habit of *A. gigas* and the current state of local native fish stocks.

MATERIAL AND METHODS

Study area

The collections of *A. gigas* were carried out along the main channel of the Cautário River and in its marginal lakes at the state of Rondônia (Fig. 1). This area was selected because it belongs to the Extractive Reserve of the Cautário River, which is inhabited by riverine communities that carry out in extractive activities and subsistence fishing.

Experimental fisheries

Fisheries of *A. gigas* occurred between August 2023 and September 2024 in several lakes in the Cautário River Basin. For

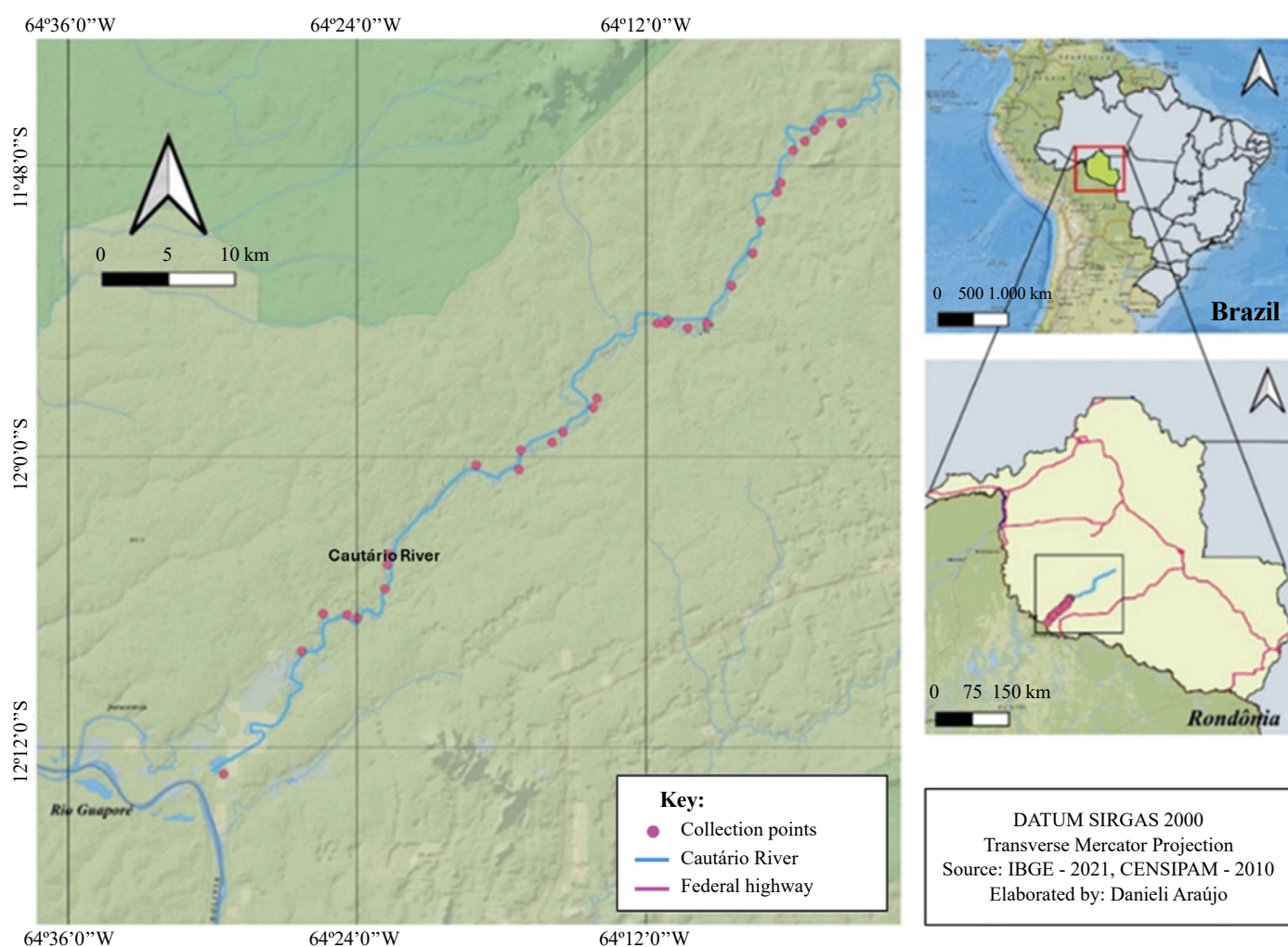


Figure 1. Study area with *Arapaima gigas* fishery sites along the Cautário River (Rondônia, Brazil).

the collections of the *A. gigas*, trawl type nets were used with meshes of 26, 28 and 30 cm, between opposite knots, 3-m high and 70-m long, which were submerged for a period of between 2 and 3 hours, then inspected. During the fisheries, hand lines with hooks attached to buoys were also used, which is traditionally known in the region as “João Bobo”.

The fisheries were authorized under environmental license No. 12/2023-Coordenação-Geral de Gestão e Monitoramento do Uso da Fauna e da Biodiversidade Aquática/Diretoria de Biodiversidade e Florestas, referring to scientific activities in the Cautário River, issued by the Instituto Brasileiro do Meio Ambiente e dos Recursos Naturais Renováveis (IBAMA), and the research project was approved by the Ethics Committee on the Use of Animals (CEUA) of the Universidade Federal de Rondônia, under No. 003-2024-A.

Analysis of stomach contents

After capture, the fish were euthanized by stunning via cerebral concussion, followed by cranial perforation, causing the disruption of the functioning of the vital organs quickly and irreversibly (Concea, 2013). Then, each specimen received a tag with individual numbering, and the weight (kg) and length (cm) were documented, followed by evisceration.

Subsequently, the stomachs of the fish were removed, labeled and fixed in 10% formalin. These were then transported to a laboratory environment in which they were washed under running water and stored in 70% alcohol. For the analysis of food items, the stomachs were prepared according to the methodology employed by Hyslop (1980).

The stomach contents were examined macroscopically (with the naked eye) and with the aid of a magnifying glass (Bioptik). Food items were classified according to their presence in the stomachs (empty or with contents), which was followed by determination of the degree of repletion, adapted from Galletti et al. (2010), in which 0 is assigned for empty stomachs, 1 for stomachs with contents occupying up to 25% of their volume, 2 for partially filled stomachs, with up to 75% of their volume occupied, and 3 for stomachs with an occupied volume of over 75%. Following this, the relative frequency and frequency of occurrence (FO%) were obtained according to the Eq. 1:

$$FO = \left(\frac{n_i}{N} \right) \times 100 \quad (1)$$

Where: n_i : the number of stomachs that contain the food item; N : the total number of stomachs analyzed (Hyslop, 1980).

Statistical analysis

The biometric measurements of the *A. gigas* specimens and the quantitative data of the food items were organized and

submitted to descriptive analysis (frequencies, mean, and standard deviation), in order to describe the main patterns and trends of the variables studied (Babbie, 2010). In addition, the identified food items were organized in tables for simple counts (absolute frequency—AF) and the comparison of this count with the total (relative frequency—RF). Additionally, the linear function $f(x) = ax + b$, in which a is the angular coefficient and b is the linear coefficient (point where the line crosses the y axis), was used to verify the relationship between the independent variable ($x = SL$, standard length) and the dependent variable ($y = TL$ total length). All the analyses were performed using R software, version 4.5.1 (R Core Team, 2025), adopting a significance level of $\alpha = 0.05$.

RESULTS

Stomachs from 128 *A. gigas* individuals were analyzed. The fish had total length (TL) ranging from 1.16 to 2.26 m (mean \pm standard deviation: 1.76 ± 0.31 m) and weight from 24.70 to 111.60 kg (52.37 ± 22.54 kg). The fish presented a robust linear function that describes the TL (dependent variable) according to the standard length (SL) (independent variable) equal to $TL = 0.9182 \cdot SL - 0.0425$ with $R^2 = 0.9775$, and a potential function to describe the increment in weight according to the TL equal to $W = 9.5715 \cdot TL - 2.9918$ with $R^2 = 0.9686$.

Among the total fish analyzed, 104 (81.2%) had contents in the stomach (some type of food ingested), with degree of repletion ranging from 1 to 3 (with mode 1 and mean of 1.42 ± 0.88) and 24 empty (0). In the stomachs with contents, 178 food items were identified, which were classified into five categories: fish and fish remains, plant material, debris, crustaceans, and worms (Table 1). The items with the highest frequency of occurrence were plant materials, and the item with the lowest value was crustaceans.

Table 1. Frequency of occurrence of food items found in the stomachs of 128 samples of *Arapaima gigas* captured in the Cautário River basin.

Food items	Absolute frequency	Relative frequency (%)	Frequency of occurrence (%)
Plant material	82	46.07	64.06
Fish and fish remains	51	28.65	39.84
Worms	32	17.98	25.00
Debris	12	6.74	9.38
Crustaceans	1	0.56	0.78

In the analysis of the stomach contents of the analyzed fish, it was possible to identify six groups of fish species (two or more species of the same genus or of different genera), as well as remains of spines and scales of unidentified fish (Fig. 2). Also, during the analysis of stomach contents, a case of cannibalism for pirarucu was recorded (Fig. 2a).

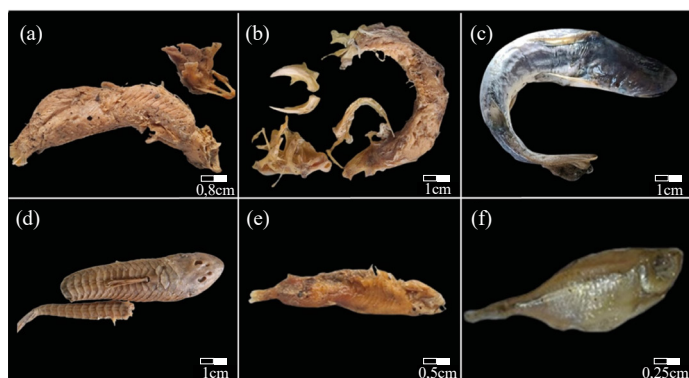


Figure 2. Group of fish species found in the stomachs of *Arapaima gigas* in the Cautário River Basin. (a) *A. gigas*, (b) *Hoplias malabaricus*, (c) *Ageneiosus inermis*, (d) *Pterygoplichthys*, (e) Characiformes (Characidae), and (f) *Astyanax* gr. *bimaculatus*. The bars in the figures represent the scale in cm.

In the stomachs analyzed, the presence of worms (unidentified), plant material (roots, stems, and leaves) and fragments of crustaceans, such as shrimp remains (*Macrobrachium amazonicum*), was observed (Fig. 3).



Figure 3. Worms and plant remains found in the stomachs of *Arapaima gigas* in the Cautário River Basin. The arrow highlights the presence of (a) worms and (b) vegetable remains — roots, stems and leaves. The bars in the figures represent the scale in cm.

DISCUSSION

The present study investigated the food found in the stomachs of *A. gigas* captured in the Cautário River Basin, and a high frequency of plant material was observed in the stomachs of

these animals. This factor may be an indication that the species is in a situation of food shortage, involuntary ingestion of plant material during the capture of its prey, or that there has been a decrease in native fish stocks.

However, the presence of plant material found in the stomach contents of pirarucus, whether absorbed voluntarily or not, contributes to the formation of their body biomass derived from allochthonous resources, a factor that corroborates with other studies confirming this pattern for fish in tropical stream systems (Chipps & Garvey, 2007; Reis et al., 2020).

During the research, it was observed that native fish, in the period of low water (dry season), tend to group in pools, deeper areas in the river (Costa et al., 2013). This behavior of adaptation and survival, given the retraction of the main channel of the river, makes the alternative refuge areas for native fish populations scarce, so when *A. gigas* individuals enter these places in search of food, they encounter their prey (native fish), which are vulnerable, or take refuge in ecotone areas (areas with vegetation on the riverbank).

Seasonal variations in flooded areas have a significant influence on the behavior and feeding habits of neotropical fish species, constituting the main factor controlling the trophic structures of these environments (Mérona & Vigouroux, 2009). In this context, adaptations observed in foraging fish species are quickly noticed by predators, such as *A. gigas*, resulting in changes in the trophic chain of the new colonized aquatic ecosystems (Lowe-McConnell, 1999).

However, in our study, we found that the fish species found in the stomachs of the analyzed individuals (N = 128; TL varying between 116 to 226 cm) were similar to those recorded in the populations of *A. gigas* captured along the middle Juruá River (N = 113; TL varying between 60 to 245 cm) in Amazonas, Brazil, in which fish of the orders Characiformes and Siluriformes were identified (Jacobi et al., 2020), followed by the items plant material and remains of fish bones.

In the stomachs of the *A. gigas* (total weight varying between 24.70 to 111.60 kg), we identified the species *Hoplias malabaricus*, *Pterygoplichthys*, and Characiformes (Characidae), which were also reported in the study by Queiroz (2000) with *A. gigas* samples (total weight varying between 7.38 to 12.58 kg) carried out in Amazonian floodplains, as well as the species tamoatá (*Megalechis thoracata*), pictus catfish (*Pimelodus* spp.), giant raphael catfish (*Megalodoras uranoscopus*) and knifefish (*Sternopygidae*, *Gymnotidae*, *Rhamphichthyidae*).

The species *Ageneiosus inermis*, known as manduba, *Pterygoplichthys*, and *Loricariichthys* cf. *acutus* (Loricariidae; Siluriformes), which are endemic to the Amazon Basin, have

also been reported as prey of *A. gigas*. As for the fish of the genus *Astyanax* (Characiformes-Characidae), which are popularly known as tetra, these are considered the main sources of food for piscivorous fish (Dzindzik, 2021).

However, the presence of pieces of wood found in the stomachs of the target species of the study may have occurred because these fish capture their food through suction (Embrapa, 2015; Fontenele, 1948; Luz, 2016; Pinese, 1996); thus, involuntarily, they also swallow a lot of plant matter that is close to their prey (Watson et al., 2013). Therefore, this process of prey capture, coupled with the period in which the individuals of the target species were captured (low-water period), can justify the high frequency of plant remains (roots, stems and leaves), which were also observed in the study by Oliveira et al. (2005), who identified organic debris, inorganic particles with portions of fine sand and plant remains, demonstrating the ability of *A. gigas* to catch food near the riverbed.

However, it was observed that the items found in the stomachs of individuals of the target species confirm that this species is a piscivorous fish, but that in situations of food scarcity, it can take advantage of other items that it swallows involuntarily, when it sucks up its prey.

According to the research conducted by Villafán et al. (2020), medium and small fish, especially those belonging to the Characidae family (order Characiformes), make up a significant part of the diet of *A. gigas*. The authors indicate that, in the Madre de Dios River Basin (Bolivia), the diet of *A. gigas* is composed of a high frequency of fish and plants, with frequency of occurrence values of 84.3 and 89.2%, respectively. These data indicate that plants may not be being consumed on an occasional basis and suggest the omnivorous nature of the species.

Studies report that throughout its ontogenic development, *A. gigas* has a diverse food habit and feeds on items of plant and animal origin, such as fruits, sprouts, seeds, flowers, aquatic macrophytes, microcrustaceans, worms, insects, mollusks, amphibians, and reptiles. However, its food base consists of fish and shrimp (Lima & Batista, 2012; Queiroz, 2000).

Therefore, the high frequency of plant parts found in the stomachs of *A. gigas* in the present study contrasts with other studies that describe this fish as a predominantly carnivorous species (Oliveira et al., 2005). This shows that *A. gigas*, even though it is a voracious predator, can also take advantage of other resources available in the environment, which is typical behavior of the ichthyic communities of the floodplain areas that vary their diet according to the dynamics and seasonal availability of food in each environment (Corrêa & Smith, 2019).

The presence of *A. gigas* in the Guaporé River Basin, in which it is invasive, has proven to be a legitimate and very serious threat to native fish species, since local residents cite a decrease in fish stocks, including the disappearance of many species that were once abundant (Catâneo et al., 2022).

Given this, through other studies, the establishment of *A. gigas* as an invasive species exerts strong pressure on native species, because of their characteristics that allow them to colonize new environments successfully, especially in places where it has no natural predator (Araújo-Lima & Goulding, 1997; Castello & Stewart, 2010; Dário & Carvalho, 2020; Freitas & Lima, 2008).

However, in addition to the threat to native fish populations through predation, *A. gigas* can also make these fish populations vulnerable to parasites, microorganisms and diseases that are brought into the environments by the invasive fish, which can lead to increased mortality and decreased genetic variability of these natural populations (Doria et al., 2020; Laikre et al., 2010; Villanúa et al., 2008; Waples & Drake, 2004).

An important topic that stands out in this study was the presence of an *A. gigas* specimen found in the stomach of one of the specimens studied (Fig. 2a), characterizing cannibalism, which may be indicative of the reduction in the diversity and abundance of native fish. This cannibalism behavior is ecologically significant and consistent with reports from other Amazonian populations (Jacobi et al., 2020). Cannibalism can emerge under high conspecific density and/or prey scarcity and often signals strong intraspecific competition, which is a typical pattern of broader invasion impacts in freshwater ecosystems (Ricciardi & MacIsaac, 2011).

The high presence of *A. gigas* in the Cautário River may have occurred due to several factors: the construction of hydroelectric projects in the Madeira River (Agostinho et al., 2009; Lima, 2017; Melo et al., 2019), which favored the significant presence of this animal in the rivers of Rondônia; the accidental (escapes from fish farms) or intentional (release into rivers) introduction of the species, which culminated in sign of a reduction of the ichthyic community (Catâneo, 2019; Catâneo et al., 2022; Rejas et al., 2025); and mainly because they do not have a natural predator that is capable of reducing their populations (Lima & Batista, 2012; Migdalski, 1957; Vitule et al., 2012).

Additionally, we found that the specimens of the analyzed species presented a robust linear function in the ratio of the increase in weight as a function of length. Another important characteristic observed is that the specimens presented a high degree of repletion, which indicates that these fish may be limited in obtaining food or have an advanced digestive process,

which reinforces the possibility of scarcity of the main source of food, native fish, which has also been impacted by the dams in the rivers of the region (Agostinho et al., 2009; Dzindzik, 2021; Lima, 2017; Melo et al., 2019).

Therefore, it is essential to determine the origin, the invasion pathways and vectors of *A. gigas* populations in newly occurring areas, in order to guide the formulation of appropriate management strategies for native and invasive populations (Catâneo et al., 2022). Thus, mitigating measures for the eradication of this species when established as invasive in Amazon must be implemented to the detriment of the protection of the native fish species that make up the genetic database of the ichthyofauna of this region and that, in addition to the ecological value, also serve as a food base for many riverine families which depend on fish for their subsistence (Hrbek et al., 2007).

Moreover, future studies can be carried out using stable isotope analyses to complement the data on the stomach contents of the invasive *A. gigas*, integrating the assimilated diet in longer time scales, offering a more enlightening view of the entire community and its trophic structure (Reis et al., 2020; Sturbois et al., 2022).

CONCLUSION

Analysis of the diet of *A. gigas* in the Cautário River Basin revealed that, although the species predominantly consumes fish, a significant proportion of its diet is composed of plant materials. This suggests that the population of this species in the study region has a mixed diet, even though it is classified as carnivorous. On the other hand, the quantity of plant items found in the stomachs of the specimens may signal an adaptation in the dietary patterns of these animals in the face of a possible shortage of native fish or other food resources.

These results highlight the need to continuously monitor changes in food availability and feeding behavior of *A. gigas* to better understand ecological dynamics and ensure adequate conservation of local fish stocks. Thus, the information regarding changes in the feeding habit of this species, together with the capture data of native and invasive species in the region, represent relevant information for decision-making by managers, in fisheries management, aiding the elaboration of standards for the capture of *A. gigas* for population control as an invasive pirarucu species and standards for its marketing chain.

It is essential to implement, in future stages, strategies aimed at monitoring spatial heterogeneity, followed by participatory management. These actions should be based on successful projects developed in Amazonian systems, as exemplified by the

experience of the Mamirauá region (state of Amazonas). In this context, it is essential to consider hydrological change as a cross-cutting factor, given its direct interaction with the dynamics of species invasion or ecological processes.

Given the relevance of *A. gigas* at the top of the trophic chain and its commercial importance, regulations for the capture of this invasive fish can influence the entire dynamics of fisheries, as well as economic agents involved in the productive chain of fisheries in the region. Knowledge of the feeding behavior of this species can contribute to the sustainable use of biodiversity in aquatic environments in the Amazon basin.

CONFLICT OF INTEREST

Nothing to declare.

DATA AVAILABILITY STATEMENT

All data relevant to the study are included in the article and as supporting information.

AUTHORS' CONTRIBUTIONS

Conceptualization: Pereira, S.B.B., Sousa, R.G.C.; **Methodology:** Pereira, S.B.B., Oliveira, N.S.; **Writing – revision and editing:** Fernandes, F.H., Chagas, T.Q., Meante, R.E.X., Beltrão, H.; **Validation:** Lima, S.A.O., Sousa, R.G.C.; **Final approval:** Sousa, R.G.C.

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DECLARATION OF USE OF ARTIFICIAL INTELLIGENCE TOOLS

The authors declare that no artificial intelligence tools were used in the preparation, writing, data analysis, or review of this manuscript.

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