

Diversity and community ecology of metazoan parasites in *Pimelodus ornatus* (Siluriformes: Pimelodidae) from the Amazonas River in Brazil

Diversidade e ecologia da comunidade de parasitos metazoários em *Pimelodus ornatus* (Siluriformes: Pimelodidae) do Rio Amazonas no Brasil

Elvis Silva Lima^{1,2*} ; Marcos Sidney Brito Oliveira³ ; Marcos Tavares-Dias^{1,3,4} 

¹ Programa de Pós-graduação em Biodiversidade e Biotecnologia (Rede Bionorte), Universidade Federal do Amapá (UNIFAP), Macapá, AP, Brasil

² Universidade do Estado do Amapá (UEAP), Macapá, AP, Brasil

³ Programa de Pós-Graduação em Biodiversidade Tropical (PPGBio), Universidade Federal do Amapá (UNIFAP), Macapá, AP, Brasil

⁴ Embrapa Amapá, Macapá, AP, Brasil

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Abstract

The present study investigated the metazoan parasite community in *Pimelodus ornatus* from the Amazon River, in the state of Amapá (Brazil). Of 71 fish examined, 70.4% were parasitized by *Demidospermus* sp. (Monogenea), *Cucullanus pinnai*, *Procamallanus (Spirocamallanus) inopinatus* and *Contraecaecum* sp. (Nematoda) and plerocercoids from Proteocephalidae gen. sp. (Cestoda). The dominance was of nematode species such as *Procamallanus (S.) inopinatus* and *Contraecaecum* sp. The parasites showed a highly aggregated dispersion and a predominance of hosts infected by one species of parasite. The parasite community was characterized by a low Shannon diversity index, low evenness and low species richness. The richness of parasite species, Shannon's diversity index, abundance of *P. (S.) inopinatus* and Proteocephalidae gen. sp. showed a positive correlation with the length of the hosts. Therefore, the size of the hosts had an influence on the parasite community and infracommunities, as well as their intermediate position in the food web. This is the first record of *P. (S.) inopinatus* and *Contraecaecum* sp. for *P. ornatus*.

Keywords: Aggregation, ectoparasites, endoparasites, freshwater fish, infection.

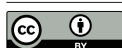
Resumo

Este estudo investigou a comunidade de parasitos metazoários em *Pimelodus ornatus* do Rio Amazonas, no estado do Amapá (Brasil). De 71 peixes examinados, 70,4% estavam parasitados. Um total de 147 parasitos foi coletado, entre *Demidospermus* sp. (Monogenea), *Cucullanus pinnai* (Nematoda), *Procamallanus (Spirocamallanus) inopinatus* e *Contraecaecum* sp. (Nematoda) e plerocercoides de Proteocephalidae gen. sp. (Cestoda). A dominância foi de nematoides como *Procamallanus (S.) inopinatus* e *Contraecaecum* sp. Os parasitos apresentaram dispersão altamente agregada e predomínio de hospedeiros infectados por uma espécie de parasito. A comunidade de parasitos foi caracterizada por baixo índice de diversidade de Shannon, baixa equitabilidade e baixa riqueza de espécies. A riqueza de espécies de parasitos, índice de diversidade de Shannon, abundância de *P. (S.) inopinatus* e Proteocephalidae gen. sp. apresentaram correlação positiva com o comprimento dos hospedeiros. Portanto, o tamanho dos hospedeiros teve influência sobre a comunidade e infracomunidades de parasitos, bem como sua posição intermediária na cadeia alimentar. Este é o primeiro registro de *P. (S.) inopinatus* e *Contraecaecum* sp. para *P. ornatus*.

Palavras-chave: Agregação, ectoparasitos, endoparasitos, peixes de água doce, infecção.

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*Corresponding author: Elvis Silva Lima. E-mail: elvisbiol10@gmail.com



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Introduction

The Amazon River is the largest drainage basin in the world and accounts for 20% of the global freshwater, 1.2×10^9 tons of sediment (Nittrouer & DeMaster, 1986; Milliman, 2001; Chong et al., 2016), 10% of the dissolved loads and 3% of the suspended loads that enter the ocean (Milliman & Syvitski, 1992; Gaillardet et al., 1997; Calvès et al., 2019). In addition, it has a high diversity of fish species.

The diversity of fish in the Amazon River is of economic importance for many riverine populations that live of fishing and use different species of fish for their subsistence (Salo et al., 2013). This diversity is due to the particular characteristics of this large river and its tributaries, which are rich ecosystems with complex trophic chains and with vegetation cover that vary according to the flood regime and regional seasonality (Costa Sousa et al., 2017; Arantes et al., 2019). However, many species of Amazonian fish and their biology are little known (Tavares-Dias & Oliveira, 2017; Negreiros et al., 2018), such as the *Pimelodus ornatus* (Kner, 1857).

Pimelodus ornatus is a Pimelodidae, popularly known as mandi-guaru, silver mandi, mandi-pinini or painted mandi. This Siluriformes can be found in the Amazon, Madeira, Parnaíba, Negro, Alto Paraná, Orinoco, which are large rivers in the Guianas, Paraguay, Bolivia, Peru and Venezuela (Nomura, 1984; Torrente et al., 2013; Froese & Pauly, 2021). Omnivorous fish, active at night, can occur in the main river channels, in rocky bottoms with dead tree trunks, upstream and downstream from rapids and backwaters (Froese & Pauly, 2021). Their first sexual maturation occurs from 15.4 cm in length (Vazzoler, 1996) and females can preserve sperm with their secretions by inserting them in the epithelium of their genital tract, being indicative of internal fertilization (Vazzoler, 1996; Boujard, 1997; Le Bail et al., 2000). This host fish has been parasitized by species of Monogenea, Nematoda, Trematoda, Cestoda and Pentastomida (Table 1). However, little is known about the ecological interactions of *P. ornatus* with its parasite community.

Adequate knowledge of parasite biodiversity is crucial for environmental management and conservation initiatives (Poulin, 2004; Negreiros et al., 2019). It is known that among the processes responsible for the spatial distribution of the parasites, they can be related to the constant scenarios of changes in the environment and

Table 1. List of metazoan parasites species reported for *Pimelodus ornatus* from South. America.

Taxon/parasite species	Locality	References
Monogenea		
<i>Demidospermus peruvianus</i>	Peru	Mendoza-Palmero et al. (2019)
<i>Demidospermus curvovaginatus</i>	Peru	Mendoza-Palmero et al. (2019)
Nematoda		
<i>Cucullanus pinnai</i>	Brazil	Thatcher (2006), Kohn et al. (2011)
<i>Pseudocladorchis cylindricus</i>	Brazil	Travassos et al. (1928)
Trematoda		
<i>Dadaytrema oxycephala</i>	Brazil	Travassos et al. (1928)
<i>Genarchella parva</i>	Argentina	Kohn et al. (2007)
<i>Genarchella genarchella</i>	Brazil	Fernandes & Kohn (2001), Kohn et al. (2011)
Cestoda		
<i>Mariauxiella pimelodi</i>	Brazil	De Chambrier & Rego (1995)
<i>Spasskyellina mandi</i>	Brazil	Pavanelli & Takemoto (1996)
<i>Nomimoscolex</i> sp.	Peru	De Chambrier et al. (2015)
Pentastomida		
<i>Porocephalus gracilis</i>	Brazil	Travassos et al. (1928)
<i>Leiperia gracile</i>	Brazil	Luque et al. (2013)

biological invasions, which can lead to diseases (McLeod & Wing, 2008; Harvell et al., 2009; Altizer et al., 2013). Therefore, biotic and abiotic factors are commonly responsible for the diversity, richness and infection rates by parasites in wild fish (Tavares-Dias et al., 2014; Blasco-Costa et al., 2015; Oliveira et al., 2017; Tavares-Dias & Oliveira, 2017; Negreiros et al., 2018), affecting the parasite-host relationship. Thus, the objective of the present study was to investigate the diversity and community ecology of metazoan parasites in *P. ornatus* in the Amazon River region of the state of Amapá, northern Brazil.

Materials and Methods

Sampling area and fish collection

Seventy-one specimens of *P. ornatus* (15.8 ± 2.4 cm and 18.9 ± 8.1 g) were collected every two months, from January to September 2020, on the Amazon River near the municipality of Santana, Amapá state (Figure 1). The fish were collected with nets of different sizes and meshes (15, 20, 25, 30 and 35), throw nets (20 mm mesh) and hand lines. The collected specimens were conserved in 10% formaldehyde and transported to the Embrapa Aquaculture and Fisheries Laboratory, Macapá, Amapá state, Brazil.

During the fish collection, the water quality parameters of electrical conductivity, pH and total dissolved solids were measured using a multiparameter (COMBO5-02-1016), and the dissolved oxygen and temperature were measured using an oximeter (MO-900). The average water temperature was 29.4 ± 1.2 °C, dissolved oxygen 5.9 ± 0.1 mg/L, pH 7.1 ± 0.2 , total dissolved solids 52.2 ± 2.0 mg/L and electrical conductivity 85.8 ± 4.9 μ S/cm.

The present study was carried out according to the recommendations and guidelines of the Brazilian College of Animal Experimentation (COBEA) and with authorization from the Ethics Committee on the Use of Animals of Embrapa Amapá (Protocol N° 014 - CEUA/CPAFAP).

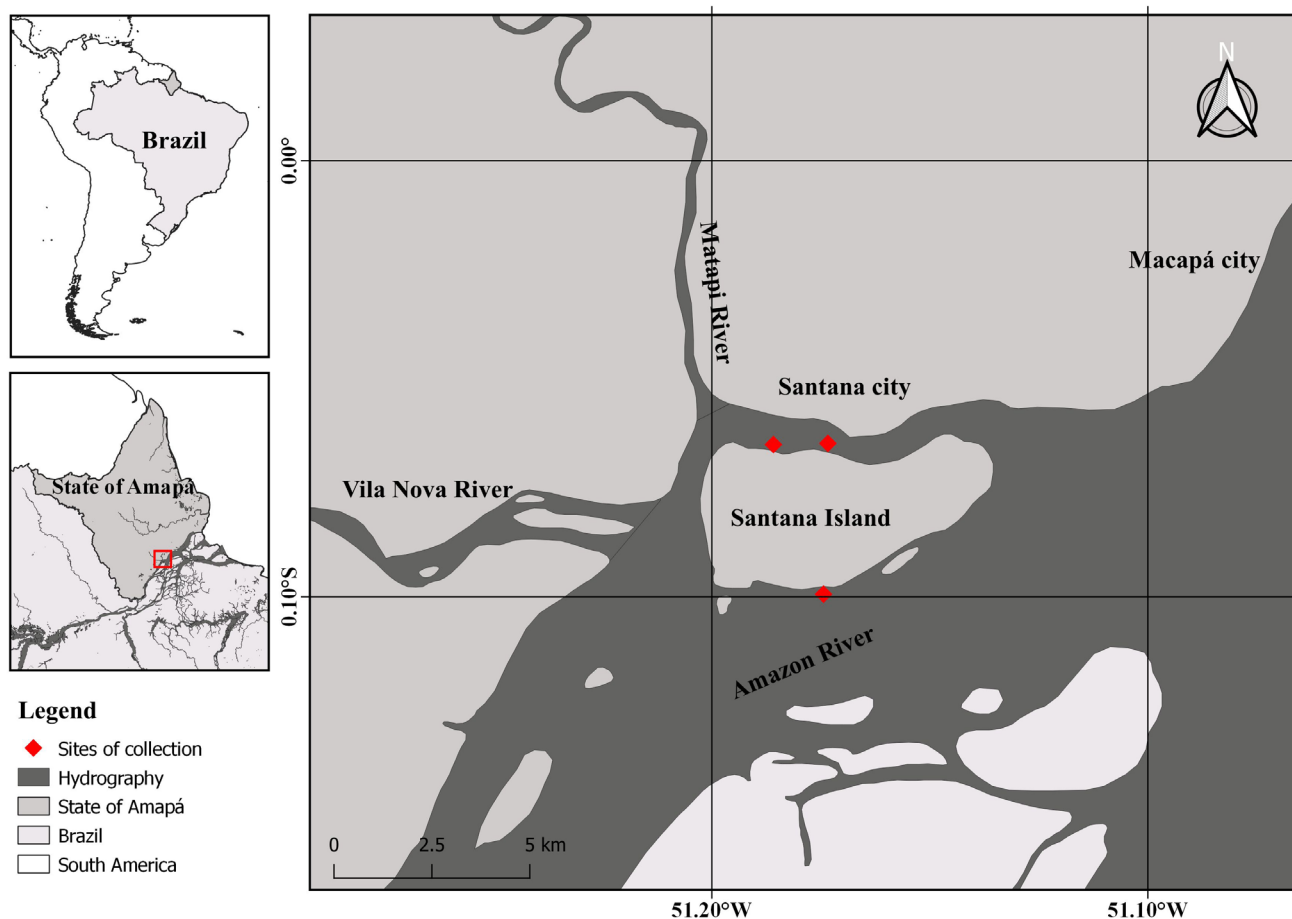


Figure 1. Collection site of *Pimelodus ornatus* in Amazonas River, State of Amapá, northern Brazil.

Collection and analytical procedures of the parasites

All fish were weighed (g) and the total length (cm) was measured, and the gills, gastrointestinal tract and viscera were examined for the presence of metazoan parasites. The gills, gastrointestinal tract and viscera were examined with a stereomicroscope and the observed parasites were fixed with 70% ethyl alcohol. The parasites were prepared for identification using the methodology described in Eiras et al. (2006). Parasites were identified according to Moravec (1998) and Thatcher (2006), and specialized papers. Voucher specimens were deposited at Instituto de Pesquisas Científicas e Tecnológicas do Estado do Amapá (IEPA), Macapá, AP, Brazil, in the Scientific Collection Curation Office for the Fauna of Amapá, under accession number IEPA:160-164P.

Data analysis

Prevalence, mean abundance, and mean intensity of parasite infracommunities (Bush et al., 1997) and frequency of dominance (Rohde et al., 1995) were calculated. The dispersion index (ID) and the significance of the ID were calculated using the *d*-statistic with the software Quantitative Parasitology 3.0, as well as the Poulin discrepancy index (D) (Ludwig et al., 1988). These parameters were calculated for species with a prevalence > 10%.

All data were previously evaluated on the assumptions of normality and homoscedasticity using the Shapiro-Wilk and Bartlett tests, respectively. The Shannon index (*H*), evenness (*E*) and species richness of parasites (Rohde et al., 1995, Magurran, 2004) were used to estimate parasite diversity. Spearman's correlation coefficient (*rs*) was used to investigate possible correlations between host body length and weight with the richness of parasite species, Shannon index and parasite abundance.

Results

Pimelodus ornatus specimens were parasitized by *Demidospermus* Suriano, 1983, *Cucullanus pinnai* (Travassos, Artigas & Pereira, 1928); *Procamallanus (Spirocamallanus) inopinatus* (Travassos, Artigas & Pereira, 1928), larvae of *Contraecum* Railliet & Henry, 1912 and plerocercoids of Proteocephalidae La Rue, 1911 (Table 2). However, the dominance was of nematode species and no parasites were found in the mouth and operculum of the examined hosts. The parasites showed aggregate dispersion (Table 3).

Table 2. Parasitic helminths in *Pimelodus ornatus* from Amazonas River, State of Amapá, in Brazil

Parasite species	P (%)	MA ± SD	MI ± SD	TNP	FD (%)	SI
Monogenea						
<i>Demidospermus</i> sp.	2.8	0.04 ± 0.3	1.5 ± 1.0	3	2.1	Gills
Nematoda						
<i>Contraecum</i> sp. (larvae)	22.5	0.4 ± 1.0	1.9 ± 1.4	31	21.2	Intestine
<i>Contraecum</i> sp. (larvae)	2.8	0.03 ± 0.2	1.0 ± 0	3	1.4	Stomach
<i>Procamallanus (S.) inopinatus</i>	52.1	1.2 ± 1.5	2.4 ± 1.3	88	60.3	Intestine
<i>Procamallanus (S.) inopinatus</i>	1.4	0.01 ± 0.1	1.0 ± 0	1	0.7	Stomach
<i>Procamallanus (S.) inopinatus</i>	1.4	0.01 ± 0.1	1.0 ± 0	1	0.7	Abdominal cavity
<i>Cucullanus pinnai</i>	5.6	0.1 ± 0.2	1.0 ± 0	4	2.7	Intestine
Cestoda						
Proteocephalidae gen. sp. (plerocercoids)	11.3	0.2 ± 0.6	1.6 ± 1.1	13	8.9	Intestine
Proteocephalidae gen. sp. (plerocercoids)	4.2	0.04 ± 0.2	1.0 ± 0	3	2.0	Stomach

P: Prevalence, MA: Mean abundance, MI: Mean intensity, TNP: Total number of parasites, FD: Frequency of dominance, SI: Site of infection, SD: Standard deviation.

The parasite component community showed a low Shannon diversity index, low evenness and low species richness, and predominance of endoparasite species (Table 4). There was a predominance of hosts infected by one species of parasite (Figure 2).

Table 3. Index of dispersion (ID), *d*-statistical (*d*) and discrepancy index (D) of parasite infracommunities in *Pimelodus ornatus* from the Amazonas River, Amapá state, in Brazil.

Parasite species	ID	<i>d</i>	D	Dispersion type
<i>Contracaecum</i> sp.	2.74	5.5	0.83	Aggregated
<i>Procamallanus</i> (S.) <i>inopinatus</i>	1.77	4.0	0.60	Aggregated
Proteocephalidae gen. sp.	1.85	4.3	0.83	Aggregated

Table 4. Component community of metazoan parasites in *Pimelodus ornatus* from the Amazon River, state of Amapá, in Brazil.

Parameters	Values
All species of parasites	
Number of hosts examined	71
Total prevalence (%) of parasites	70.4
Total number of parasites	147
Number species of parasites	5
Diversity of Shannon	0.2 ± 0.3
Evenness	0.1 ± 0.2
Species richness of parasites	1.0 ± 0.8
Species of endoparasites	
Number species of endoparasites	4
Percentage of endoparasites (%)	97.9
Species of endoparasites (larvae)	2
Species of endoparasites (adults)	2
Species of ectoparasites	
Number species of ectoparasites	1
Percentage of ectoparasites (%)	2.1
Species of ectoparasites (larvae)	0

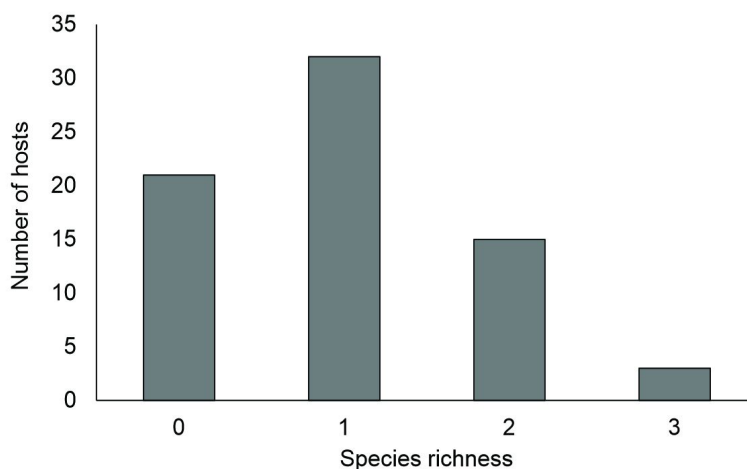


Figure 2. Species richness of metazoan parasites in *Pimelodus ornatus* from the Amazon River, Amapá of State, in Brazil.

The species richness of parasites ($rs = 0.24$; $p = 0.04$) and Shannon's diversity index ($rs = 0.29$; $p = 0.01$) showed a weak positive correlation with the length of the hosts. The richness of parasite species ($rs = 0.18$, $p = 0.12$) had no correlation with the weight of the hosts, but the Shannon's diversity index ($rs = 0.27$; $p = 0.02$) showed weak positive correlation with the weight of the hosts. The abundance of *P. (S) inopinatus* ($rs = 0.21$; $p = 0.01$) and Proteocephalidae gen. sp. ($rs = 0.22$; $p = 0.05$) showed a weak positive correlation with the length of the hosts. There was no correlation between the weight of the hosts and the abundance of *P. (S) inopinatus* ($rs = 0.08$; $p = 0.50$), Proteocephalidae gen. sp. ($rs = 0.22$; $p = 0.06$) and *Contracaecum* sp. ($rs = -0.05$; $p = 0.66$). The abundance *Contracaecum* sp. ($rs = -0.01$, $p = 0.91$) there was no correlation between host length.

Discussion

The component community of metazoan parasites in *P. ornatus* from the Amazon River consisted of one species of Monogenea, three species of Nematoda and one species of Cestoda. Few parasite species known for *P. ornatus* were found here (Table 1). For *P. ornatus* sampled from the reservoir of the Hydroelectric Power Station of Itaipu, State of Paraná (Brazil), Kohn et al. (2011) reported only one Nematoda species and one Digenea species. In contrast, the metazoan community in *Pimelodus blochii* (Valenciennes 1840) from the Acre and Iaco rivers, was made up of five species of Monogenea, 10 Nematoda, three Digenea, one Cestoda and three Crustacea (Negreiros et al., 2018); in *Pimelodus pohli* Ribeiro & Lucena, 2006 of the São Francisco river, for two species of Monogenea, three Nematoda, one Digenea and one Acanthocephala (Sabas & Brasil-Sato, 2014) and in *Pimelodus maculatus* Lacepède, 1803 of the Guandu River for one species of Nematoda and one species of Cestoda (Albuquerque et al., 2008). These results are probably related to omnivorous feeding habit of these host fish species (Froese & Pauly, 2021), considering that acquiring of nematodes and other endoparasites are linked to the diet (Sabas & Brasil-Sato, 2014; Blasco-Costa et al., 2015; Oliveira et al., 2017; Tavares-Dias & Oliveira, 2017; Negreiros et al., 2018). However, such differences in the component community and parasite richness may be attributed to the difference in host species, diet and different environments.

In *P. ornatus*, the parasites showed an aggregate dispersion pattern, which is common in several species of freshwater fish from various natural environments (Sabas & Brasil-Sato, 2014; Tavares-Dias & Oliveira, 2017; Oliveira et al., 2019; Neves et al., 2020a). This aggregation may be associated with the genetic variability of the host population, decreased interspecific competition between parasites, decreased damage to the host and other environmental factors (Poulin, 2013; Tavares-Dias & Oliveira, 2017; Salgado-Maldonado et al., 2019).

Monogeneans are parasites that may serve as indicators of environmental quality, since they are generally present in greater abundance in aquatic environments with low conditions in water (Dogiel, 1961; Oliveira et al., 2017, 2019; Negreiros et al., 2018). In addition, most monogenean parasites are species-specific, parasitizing a host or phylogenetically related hosts. The depth of the water body and flow velocity limit the exploitation of monogenean species in the host fish (Negreiros et al., 2018). In *P. ornatus* of the Amazon River, an environment with a high average water flow ($1.0 \times 10^5 \text{m}^3/\text{s}$), which prevents the accumulation and permanence of pollutants and eutrophication (Abreu et al., 2020), and with a high level of oxygen, there was a low level of infection by monogeneans *Demidospermus* sp. Negreiros et al. (2018) reported that anthropogenic development of the Rio Acre influenced the levels of infection by *Demidospermus peruvianus* Mendoza-Palermo & Scholz, 2011; *Demidospermus striatus* Mendoza-Palermo & Scholz, 2011; *Demidospermus* sp. and *Ameloblastella* Kritsky, Mendoza-Franco & Schoz, 2000 in the gills of *P. blochii*.

In *P. ornatus*, larval stage of *Contracaecum* sp. and Proteocephalidae gen. sp. were present, indicating that this fish is an important intermediate host in the transmission of these endoparasites perhaps due to its omnivorous feeding habit (Sánchez-Botero & Araújo-Lima, 2001; Froese & Pauly, 2021). We observed that *P. ornatus* was feeding on small crustaceans such as crabs and shrimp. In addition, *P. ornatus* also feeds on small fish (Sá-Oliveira et al., 2014). These results indicate that *P. ornatus* occupies a lower position in the food chain, thus facilitating these infections by such endoparasites with different life cycles. However, this was the first record of *Contracaecum* sp. for *P. ornatus*.

Procamallanus (S.) inopinatus, a nematode with wide geographic distribution and is found in different species of fish in Brazil (Neves et al. 2020b), was recorded here for the first time in *P. ornatus*. This endoparasite was the dominant species in *P. ornatus* and had a higher level of infection when compared to *Contracaecum* sp. and *C. pinnai*, and occurred in the intestine, stomach and abdominal cavity. However, the levels of *C. pinnai* infection in *P. ornatus* were low when compared to *P. blochii* from the Iaco and Acre rivers (Negreiros et al., 2018; Negreiros et al., 2019).

and *P. maculatus* from the Rio Guandu (Albuquerque et al., 2008). *Cucullanus pinnai* has also been reported to parasitize *Pimelodus albicans* (Valenciennes, 1840) (Chemes & Takemoto, 2011) and *Pimelodus clarias* (Linnaeus, 1758) (Kohn & Fernandes, 1987), demonstrating that this is a common nematode infecting species of *Pimelodus*.

No crustacean parasite species was found on the gills of *P. ornatus* from the Amazonas River. Similar findings were reported by Neves & Tavares-Dias (2019) for *Ageneiosus ucayalensis* (Castelnaud, 1855), *Pimelodella eigenmanni* (Boulenger, 1891), *Colomesus asellus* (Muller & Troschel, 1849), *Pimelodus blochii* (Valenciennes, 1840), *P. ornatus*, *Platynemateichthys notatus* (Jardine, 1841) and *Peckoltia lineola* (Armbruster, 2008) from Matapi River, a tributary of the Amazonas River, in Amapá State (Brazil). This absence of parasitic crustacean species may be due to the water velocity and daily tides from the Amazonas River (Abreu et al., 2020), given that these ectoparasites need to swim to find adequate hosts, while others depend on the flow of water and swimming speed (Neves & Tavares-Dias, 2019).

In *P. ornatus*, regarding the host-parasite relationship, there was a positive correlation of the species richness of parasites and Shannon diversity index with the total length of the host, demonstrating that the size of the fish influenced these parameters. Similar findings have been reported for *Chaetobranchopsis orbicularis* (Steindachner, 1975) (Tavares-Dias & Oliveira, 2017), *Colossoma macropomum* Cuvier, 1816 (Gonçalves et al., 2018) and *Acestorhynchus falcirostris* (Cuvier, 1819) (Hoshino et al., 2016). Moreover, the body length of *P. ornatus* also influenced in the abundance of *P. (S) inopinatus* and Proteocephalidae gen. sp., in which larger fish tended showed more parasites when compared to smaller ones, although these correlations explicit only 22% of occurrence of this parasite. Determining the factors that affect the parasites present in wild fish populations is important for parasite ecology studies. However, in fish populations, the influence of the body size on parasite load may vary, and the causes of variations are little understood.

Conclusions

The parasite community of *P. ornatus* in the Amazon River was composed of helminth species with low prevalence, low abundance, low diversity and low species richness, predominance of ectoparasites and with aggregate dispersion. The size of the host fish influenced the diversity of parasites, explaining less than 25% of the occurrence of parasites. In addition, new reports of parasites have been recorded for *P. ornatus*. The data obtained here emphasize the importance of the Amazon River as a source of biodiversity. It was possible to contribute to an increase in the knowledge of the freshwater biodiversity from the Amazon, expanding the ecological interactions of parasites and biological information on this Amazonian siluriform with gaps in the literature. This information may be used for future comparisons in studies on the impacts of anthropogenic actions on the parasitic diversity of *P. ornatus* in the Amazon River.

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