

Metazoan fauna parasitizing *Peckoltia braueri* and *Pterygoplichthys pardalis* (Loricariidae) catfishes from the northeastern Brazilian Amazon

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ABSTRACT

This study investigated the metazoan parasite fauna in *Peckoltia braueri* and *Pterygoplichthys pardalis* from a tributary of the Amazon River system, in northern Brazil. In *P. braueri*, 630 parasites were collected, belonging to *Unilatus unilatus*, *Nothogyrodactylus* sp., *Genarchella genarchella*, *Proteocephalus* sp., *Gorytocephalus elongorchis*, *Dolops longicauda* and Hirudinea gen. sp., with a dominance of monogenean species. In *P. pardalis*, 120 parasites were collected, belonging to *U. unilatus*, *Acanthostomum gnerii* and *Gorytocephalus elongorchis*, and this acanthocephalan was the dominant species. In both hosts, the parasites presented clumped distribution, high prevalence, low abundance, low Shannon diversity, and low species richness, with a predominance of helminth species. In *P. braueri*, host length was positively correlated with parasite species richness and Shannon index. There was no difference in the body condition of parasitized and non-parasitized fishes in either host. This was the first report of these parasites in *P. braueri* and *P. pardalis*.

KEYWORDS: Digenea, diversity, helminths, parasites, freshwater fishes.

Fauna de metazoários parasitando *Peckoltia braueri* e *Pterygoplichthys pardalis* (Loricariidae), bagres do nordeste da Amazônia brasileira

RESUMO

Este estudo investigou a fauna de parasitos metazoários em *Peckoltia braueri* e *Pterygoplichthys pardalis* de um tributário do sistema do Rio Amazonas, no Amapá, norte do Brasil. Em *P. braueri* foram coletados 630 parasitos das espécies *Unilatus unilatus*, *Nothogyrodactylus* sp., *Genarchella genarchella*, *Proteocephalus* sp., *Gorytocephalus elongorchis*, *Dolops longicauda* e Hirudinea gen. sp., com dominância das espécies de monogenoideas. Em *P. pardalis* foram coletados 120 parasitos das espécies *U. unilatus*, *Acanthostomum gnerii* e *Gorytocephalus elongorchis*, sendo este acantocéfalo a espécie dominante. Os parasitos em ambos hospedeiros apresentaram dispersão agregada, elevada prevalência, baixa abundância, baixa diversidade de Shannon e baixa riqueza de espécies, com predominância de espécies de helmintos. Em *P. braueri*, o comprimento dos hospedeiros apresentou correlação positiva com a riqueza de espécies de parasitos e o índice de Shannon. Não houve diferença entre a condição corporal de peixes parasitados e não parasitados para os dois hospedeiros. Este foi o primeiro relato desses parasitos para *P. braueri* e *P. pardalis*.

PALAVRAS-CHAVE: Digenea, diversidade, helmintos, parasitos, peixes de água doce.

INTRODUCTION

Loricariidae are endemic fishes of rivers, lakes and streams in the Neotropical region and represent the largest family of fish, with approximately 800 known species (25% of Siluriformes diversity) (Armbruster 2011; Nelson 2016). *Peckoltia braueri* Eigenmann, 1912 (= *Hemiancistrus braueri*) and *Pterygoplichthys pardalis* Castelnau, 1855 (= *Liposarcus pardalis*), both objects of this study, are siluriforms distributed in the tropical regions of South America, mainly in the Amazon River system. *Peckoltia braueri* has a maximum length of 10.3 cm, while *P. pardalis* can reach nearly 50 cm (Froese and Pauly 2016). Both fish species have sedentary behavior and inhabit the bottom of water bodies. They are omnivores and feed mainly on particulate organic matter (detritus), as well as filaments of single-celled algae and microorganisms associated with sediment, such as protozoans, fungi and bacteria (Fisch-Muller 2003; Mazzoni *et al.* 2010 a,b; Soares *et al.* 2011; Lujan *et al.* 2015; Froese and Pauly 2016).

Peckoltia braueri and *P. pardalis* have economic importance because, in addition to feeding riverside dwellers, they are used in aquariums (Baumgartner *et al.* 2012; Porto *et al.* 2012). When in environments with low levels of dissolved oxygen, these fishes use their extremely vascularized stomach as an accessory breathing organ. Due to this physiological adaptation, the food is not retained in the stomach and goes directly to the bowel (Santos *et al.* 2006; Froese and Pauly 2016). Despite the great diversity of Loricariidae, studies on the parasites fauna associated to these fishes are limited or scarce, depending on the species.

In general, fish can host a wide range of parasite taxonomic groups, which can affect behavior, metabolism, body condition, fertility and survival of the population (Barber *et al.* 2000; Lafferty 2008; Seppänen *et al.* 2009; Lacerda *et al.* 2013). Thus, parasitic infections may be one of many factors that may lead to a decrease in the quality and quantity of the fishery. The diversity and richness of parasites may vary among host species, and also as a function of age, size, diet composition, environment quality, and seasonality (Guidelli *et al.* 2006; Alarcos and Timi 2012; Neves *et al.* 2013; Tavares-Dias *et al.* 2014). Thus, the analysis of parasite communities and diversity parameters provides relevant information about host fish populations, expanding the knowledge of parasite-host-environment interactions. *Pterygoplichthys pardalis* is known to be parasitized by monogenean, digenean and acanthocephalan species (Thatcher and Varela 1981; Luque *et al.* 2011; Porto *et al.* 2012; Mendoza-Franco *et al.* 2010), but its parasitic fauna is not well known. The parasitic fauna of *P. braueri* has not been studied. Thus, the objective of this study was to investigate the parasite fauna of these fishes in a tributary of Amazon River system, in northern Brazil.

MATERIALS AND METHODS

Study area and collection of fishes

Igarapé Fortaleza basin, located in the area of the cities of Macapá and Santana, state of Amapá (northern Brazil), is a tributary of the Amazon River (Figure 1), formed by a main channel and an extensive floodplain area. The seasonal influence of high rainfall and the daily tides of the Amazon River, due to the proximity of its confluence with the Atlantic Ocean, originate an environmental variability that provides shelter and food for a great variety of fishes. The regional vegetation consists of floodplain forests and open herbaceous areas, mainly composed of several species of macrophytes (Thomaz *et al.* 2003; Tavares-Dias *et al.* 2014).

From November 2013 to November 2014, 39 specimens of *P. braueri* and 33 specimens of *P. pardalis* were captured in Igarapé Fortaleza River (Figure 1), using gill nets of different mesh sizes (15-50 mm between knots). Captured fishes were placed in containers with water and taken to the Laboratory of Aquatic Organisms Health of Embrapa Amapá, Macapá (Amapá) for a parasitological analysis.

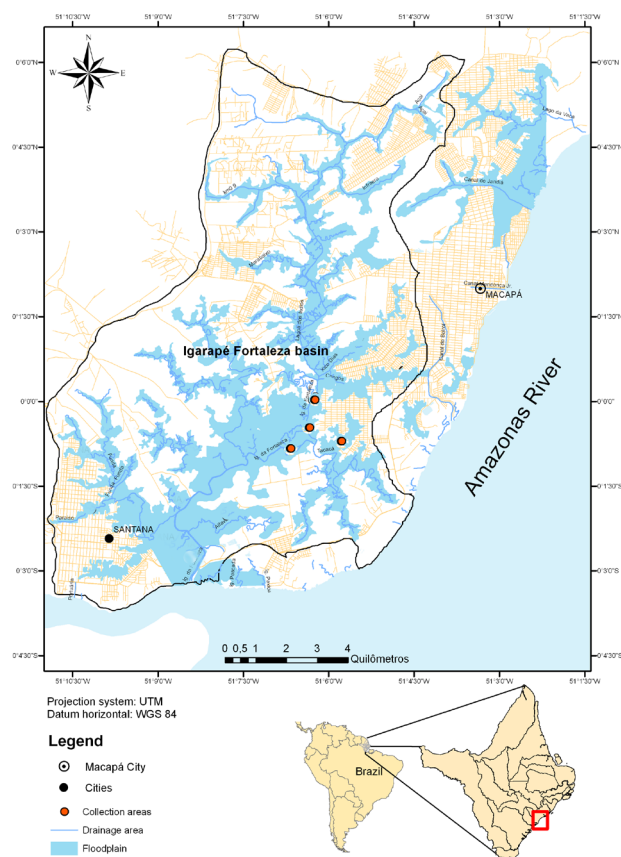


Figure 1. Collection sites of *Peckoltia braueri* and *Pterygoplichthys pardalis* in Igarapé Fortaleza basin, state of Amapá, northeastern Brazilian Amazon. This figure is in color in the electronic version.

Parasite analysis

After measuring body weight (g) and total length, each fish was subject to necropsy for parasitological analysis. From each fish, mouth, opercula and gills were examined for the collection of ectoparasites and the gastrointestinal tract for the collection of endoparasites. Collection, fixation, preparation, counting, and staining of parasites for identification were according to Eiras *et al.* (2006). The ecological terms used in this study were according to Bush *et al.* (1997).

Shannon index (*H*), evenness (*E*), parasite species richness (Magurran, 2004) and dominance frequency (Rohde *et al.* 1995) were calculated to assess the component community of parasites using the Diversity software (Pisces Conservation Ltda, UK). Dispersion index (DI) and discrepancy index (D) were calculated using the Quantitative Parasitology 3.0 software to detect the distribution pattern of parasite infra-communities (Rózsa *et al.* 2000) for species with a prevalence higher than 10%. For each infra-community, the DI significance was tested using the *d* statistical test (Ludwig and Reynolds 1988).

Weight (g) and total length (cm) data of each host species were used to calculate the relative condition factor (Kn) (Le-Cren 1951) of parasitized and non-parasitized fishes, which was compared using Mann-Whitney test (*U*). The Spearman's correlation coefficient (*r_s*) was used to determine possible correlations of host weight and length with parasite abundance, Shannon index, species richness and *H* (Zar 2010).

RESULTS

From the 39 *P. braueri* examined, 48.8% were parasitized and 630 metazoans, i.e. monogeneans, acanthocephalan, digenean, cestode, argulid and leech species were collected, with a dominance of *Unilatus unilatus* Mizelle & Kritsky, 1967 and *Nothogyrodactylus* Kritsky & Boeger, 1991 (Monogenoidea), followed by *Genarchella genarchella*

Travassos, Artigas & Pereira, 1928, while *Dolops longicauda* Heller, 1857 (Crustacea) and leeches were the less prevalent parasites. Larvae of *Proteocephalus* Weinland, 1958 (Cestoda) were also found (Table 1).

From the 33 *P. pardalis* examined, 60.6% were parasitized by monogeneans, acanthocephalans and digeneans, and 120 of these metazoan parasites were collected (Table 2). There was a dominance of *Gorytocephalus elongorchis* Thatcher, 1979 (Acanthocephala), followed by *Acanthostomum gnerii* Szidat, 1954 (Digenea). *Peckoltia braueri* and *P. pardalis* parasites presented clumped dispersion (Table 3). In both species, non-parasitized hosts were predominant (Figure 2).

For *P. braueri*, the Shannon index was 0.27 ± 0.45 , evenness was 0.15 ± 0.25 , and parasite species richness was 1.23 ± 1.62 parasites per host. Host length was positively and significantly correlated with parasite species richness ($r_s = 0.662$, $p = 0.0001$) and the Shannon index ($r_s = 0.578$, $p = 0.0001$). For *P. pardalis*, the Shannon index was 0.24 ± 0.38 , equitability was 0.22 ± 0.38 , and parasite species richness was 0.91 ± 1.15 parasite per host. The hosts length was not significantly correlated with parasite species richness ($r_s = -0.212$, $p = 0.237$) nor the Shannon index ($r_s = -0.328$, $p = 0.063$). In *P. braueri*, the abundance of *G. genarchella* and *G. elongorchis* was positively and significantly correlated with the length and weight of hosts. In *P. pardalis*, no significant correlation of length and weight of hosts with parasite abundance was observed (Table 4).

The equation that describes the weight-length relation showed a negative allometric growth for both *P. braueri* ($y = 0.1702x^{2.082}$, $r^2 = 0.890$) and *P. pardalis* ($y = 4.0497x^{1.281}$, $r^2 = 0.317$), which indicates a higher increase in body mass than in length in these hosts. The Kn in non-parasitized (1.000 ± 0.004) and parasitized (1.000 ± 0.005) *P. braueri* was similar ($U = 143.0$, $p = 0.949$), as it was in non-parasitized (1.000 ± 0.003) and parasitized (1.000 ± 0.001) *P. pardalis* ($U = 79.0$, $p = 0.060$).

Table 1. Metazoan parasites in *Peckoltia braueri* from Igarapé Fortaleza basin, northeastern Brazilian Amazon. P: Prevalence, MI: Mean intensity, MA: Mean abundance, FD: Frequency of dominance, TNP: Total number of parasites, SI: Site of infection.

Parasite species	P (%)	MI	MA	FD (%)	TNP	SI
<i>Unilatus unilatus</i> and <i>Nothogyrodactylus</i> sp.	46.5	9.8	4.6 ± 9.8	0.31	196	Gills
<i>Genarchella genarchella</i>	9.3	6.25	0.6 ± 0.2	0.04	25	Gills
<i>Genarchella genarchella</i>	23.3	24.0	5.6 ± 0.2	0.38	240	Intestine
<i>Genarchella genarchella</i>	6.9	6.0	0.4 ± 2.7	0.03	18	Stomach
<i>Genarchella genarchella</i>	6.9	8.0	0.6 ± 17.2	0.04	24	Abdominal cavity
<i>Proteocephalus</i> sp. (larvae)	2.3	2.0	0.05 ± 1.8	-	2	Stomach
<i>Proteocephalus</i> sp. (larvae)	13.9	14.7	2.0 ± 2.6	0.14	88	Intestine
<i>Gorytocephalus elongorchis</i>	18.60	3.9	0.7 ± 0.3	0.05	31	Intestine
<i>Gorytocephalus elongorchis</i>	6.9	1.3	0.09 ± 7.3	0.01	4	Abdominal cavity
<i>Dolops longicauda</i>	2.3	1.0	0.02 ± 1.9	-	1	Gills
Hirudinea gen. sp.	2.3	1.0	0.02 ± 0.4	-	1	Gills

Table 2. Metazoan parasites in *Pterygoplichthys pardalis* from Igarapé Fortaleza basin, northeastern Brazilian Amazon. P: Prevalence, MI: Mean intensity, MA: Mean abundance, FD: Frequency of dominance, TNP: Total number of parasites, SI: Site of infection.

Parasite species	P (%)	MI	MA	FD (%)	TNP	SI
<i>Unilatus unilatus</i>	15.2	3.4	0.52 ± 1.7	0.142	17	Gills
<i>Acanthostomum gnerii</i>	30.3	4.8	1.45 ± 3.9	0.400	48	Intestine
<i>Acanthostomum gnerii</i>	6.1	1.0	0.06 ± 0.2	0.017	2	Abdominal cavity
<i>Acanthostomum gnerii</i>	3.0	4.0	0.12 ± 0.7	0.033	4	Stomach
<i>Gorytocephalus elongorchis</i>	45.5	3.3	1.48 ± 2.2	0.408	49	Cavidade abdominal

Table 3. Dispersion Index (DI), Statistic-*d* and discrepancy index (D) for parasite infracommunities of two Loricariidae species from Igarapé Fortaleza basin, northeastern Brazilian Amazon.

Parasite species	DI	<i>d</i>	D
<i>Peckoltia braueri</i>			
<i>Unilatus unilatus</i> and <i>Nothogyrodactylus</i> sp.	3.600	7.88	0.664
<i>Genarchella genarchella</i> (intestine)	4.394	9.61	0.793
<i>Proteocephalus</i> sp. (intestine)	3.171	6.86	0.872
<i>Gorytocephalus elongorchis</i> (intestine)	2.491	5.10	0.833
<i>Pterygoplichthys pardalis</i>			
<i>Unilatus unilatus</i>	2.547	4.83	0.858
<i>Acanthostomum gnerii</i> (intestine)	2.644	5.08	0.777
<i>Gorytocephalus elongorchis</i>	1.777	2.73	0.666

Table 4. Spearman correlation coefficient (*rs*) and the associated probability (*p*) of the abundance of parasite species with the total length and body weight for Loricariidae species from the Igarapé Fortaleza basin, northeastern Brazilian Amazon.

Parasite species	Length		Weight	
	<i>rs</i>	<i>p</i>	<i>rs</i>	<i>p</i>
<i>Peckoltia braueri</i>				
<i>Unilatus unilatus</i> and <i>Nothogyrodactylus</i> sp.	0.178	0.279	0.226	0.166
<i>Genarchella genarchella</i>	0.635	0.0001	0.643	0.0001
<i>Proteocephalus</i> sp.	0.299	0.064	0.218	0.182
<i>Gorytocephalus elongorchis</i>	0.573	0.0001	0.476	0.002
<i>Pterygoplichthys pardalis</i>				
<i>Unilatus unilatus</i>	0.178	0.320	0.089	0.621
<i>Acanthostomum gnerii</i>	0.185	0.303	-0.060	0.742
<i>Gorytocephalus elongorchis</i>	0.047	0.793	-0.281	0.113

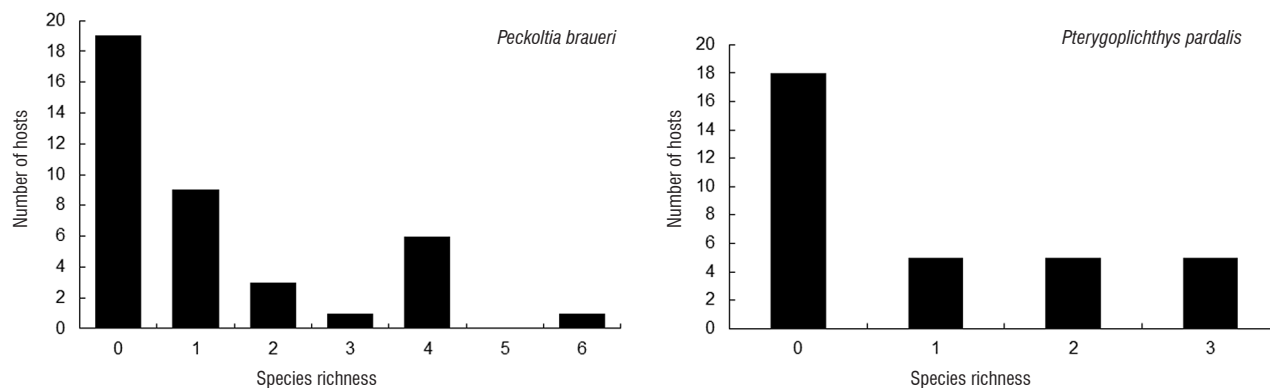


Figure 2. Species richness of parasites in two Loricariidae species from the Igarapé Fortaleza basin, northeastern Brazilian Amazon.

DISCUSSION

In wild fish populations, the impact of parasitic infections can indicate a potential decrease in fishery production for consumption due to losses in quality and quantity, reduction in fish weight and body injuries that may lead to discarding the venture. The parasitic fauna of *P. braueri* and *P. pardalis* was composed of monogenean, acanthocephalan, digenean, cestode, crustacean and hirudinean species. However, *Proteocephalus* sp., *D. longicauda*, *G. genarchella* and hirudineans only occurred in *P. braueri*, while *A. gnerii* parasitized only *P. pardalis*. *Pterygoplichthys pardalis* from different locations have been parasitized by *Unilatus* spp., *Heteropriapulus* spp., *Megacoelium spinispecum* Thatcher & Varella, 1981, *Austrodiplostomum compactum* (Lutz, 1928) Dubois, 1970 and *Gorytocephalus* sp. (Thatcher and Varella 1981; Mendoza-Franco *et al.* 2010; Luque *et al.* 2011; Porto *et al.* 2012). When the same endoparasite is found in more than one host species in the same environment, it indicates the occurrence of new intermediate hosts in the habitat, in addition to the fact that different fish species share one or more food items (Marcogliese 2002).

In nature, the patterns among parasitic communities of host populations can be detected through quantitative and qualitative descriptors (Magurran 2004; Luque *et al.* 2013). In *P. braueri* and *P. pardalis*, Shannon diversity, equitability and species richness of parasites were low. In host fish populations, parasite diversity and richness depend directly on the population dynamics of birth, death, migration, age and diet of the host, presence of infective forms, environment quality, seasonality and geographical factors, among other biotic and abiotic factors (Luque *et al.* 2013; Hoshino and Tavares-Dias 2014; Tavares-Dias *et al.* 2014).

Monogeneoidea is the most diverse taxa in fish from South America, with 835 species reported from hosts of different countries, most of them from freshwater habitats (Luque *et al.* 2016). Thirteen genera of monogeneans can be found parasitizing several species of Loricariidae, but *Unilatus* Mizelle & Kritsky, 1967 and *Trinigyryus* Hanek, Molnar & Fernando, 1974 (Dactylogyridae) are the most frequently encountered in these hosts (Cohen *et al.* 2013). In this study, *P. braueri* was parasitized by *U. unilatus* and *Nothogyrodactylus* sp., while *P. pardalis* was parasitized only by *U. unilatus*, but the level of infection was higher in *P. braueri* gills. These differences in parasitism levels are likely species-specific. This is the first record of *U. unilatus* in *P. braueri* and *P. pardalis*.

Genarchella genarchella is a widely distributed digenean in Characiformes and Siluriformes species from Brazil, Argentina and Uruguay (Scholz *et al.* 1995; Kohn *et al.* 2003; Franceschini *et al.* 2013). As this digenean species has mollusks and Cypriniformes species as intermediate hosts, and Characiformes and Siluriformes species are definitive hosts (Martorelli 1989;

Lefebvre and Poulin 2005). *Peckoltia braueri* in our study were infected by the ingestion of mollusks containing infective forms of this digenean. *Acanthostomum gnerii*, a digenean commonly found in the digestory tract of freshwater Siluriformes, has mollusk species as primary intermediate hosts, fishes of several species as secondary intermediate hosts and siluriform species as definitive hosts (Ostrowski-Nunez and Gil-Pertierra 1991; Gil-Pertierra and Ostrowski-Nunez 1995). In *P. braueri*, the levels of infection by *G. genarchella* were similar to those of *A. gnerii* in *P. pardalis*. However, the levels of infection by *A. gnerii* were higher than the ones reported for *Rhamdia quelen* Quoy and Gaimard, 1824 (Gil-Pertierra and Ostrowski-Nunez 1995). The abundance of *G. genarchella* increased with the size of *P. braueri*, indicating that larger individuals and, thus, older ones, host more parasites. Similar results were reported for *Astronotus ocellatus* Spix & Agassiz, 1831 parasitized by *Posthodiplostomum* sp. (Neves *et al.* 2013). This is the first report of *G. genarchella* in *P. braueri* and of *A. gnerii* in *P. pardalis*.

Amazonian acanthocephalans are a group of parasitic worms that have attracted relatively little attention in comparison to other endohelminths. However, they are found in all classes of fish besides aquatic birds and mammals. *Gorytocephalus elongorchis*, described for the first time parasitizing *Hipostomus carinatus* Steindachner, 1881 (= *Plecostomus carinatus*) from Janauacá Lake, western Amazon region (Thatcher 1979), was found in *P. pardalis* and *P. braueri*, which are new hosts for this acanthocephalan. These results also expand the geographical distribution of *G. elongorchis* to the eastern Amazon region. The levels of infection by *G. elongorchis* in *P. pardalis* were high, while they were moderate in *P. braueri*. Furthermore, the abundance of *G. elongorchis* increased with the size of *P. braueri*, indicating that older individuals are more parasitized. Likewise, for other fish populations, the abundance or intensity of endohelminths increases with the age or size of the hosts (Rohde *et al.* 1995; Lacerda *et al.* 2013). This is the first report of *G. elongorchis* in *P. pardalis* and *P. braueri*.

Cestoda represents the third species-rich group of helminths in South America, with 460 species found in different countries (Luque *et al.* 2016). *Proteocephalus* species are the most frequent cestodes in Siluriformes from South America (Rego *et al.* 1999; Chambrier *et al.* 2006). Diaptomidae crustaceans and cyclopoid copepods are intermediate hosts for *Proteocephalus* species. Metacercariae or procercooids of this cestode develop in the body cavity of fishes, definitive or paratenic hosts, which are directly infected after consuming such infected crustaceans (Scholz 1999; Rego *et al.* 1999). Plerocercariae of *Proteocephalus* sp. were only found in *P. braueri*, and with low levels of infection, indicating that this fish may be a secondary intermediate host for this cestode species.

This first report of *D. longicauda* in *P. braueri* showed infestation in only one host, similar to the findings in *Metynniss lippincottianus* Cope, 1870 (Hoshino and Tavares-Dias 2014), also from the same habitat of this study. This argulid presents a wide distribution in the Amazon River system (Tavares-Dias et al. 2015). Other ectoparasites with low levels of infestation in the gills were hirudinean species, which were also found in *P. braueri* gills. Therefore, the infections caused by *D. longicauda* and hirudineans in fish seem to be accidental.

CONCLUSIONS

This first study on *P. braueri* parasites shows that the parasitic fauna of *P. braueri* and *P. pardalis* is composed by a low diversity of metazoan species, predominantly by helminth species with high prevalence, low abundance and clumped distribution. This low abundance of parasites did not influence the body conditions of these two hosts. Finally, for *P. braueri*, host length explains 60% of the species richness and diversity of parasites. However, in *P. pardalis*, other factors that were not analyzed herein are responsible for parasite richness, low diversity and abundance.

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