

PARASITES IN GILLS OF *Aequidens tetramerus*, CICHLID FROM THE LOWER JARI RIVER, AN TRIBUTARY OF THE AMAZON RIVER, NORTHERN BRAZIL

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ABSTRACT

The aim of this study was to investigate for the first time the parasite fauna in the *Aequidens tetramerus* gills of the lower Jari River, State of Amapá (Brazil). In January 2017, 31 specimens of *A. tetramerus* were collected and they were necropsied using usual methodologies for parasitology of fish. A total of 1,181 parasites were collected and 100% of hosts were parasitized by *Gussevía disparoides*, *Gussevía alioides*, *Posthodiplostomum* sp., *Clinostomum* sp., *Dolops longicauda* and *Acarina* gen. sp. The dominance was of *G. disparoides* and there was aggregated dispersion of *G. disparoides*, *Posthodiplostomum* sp. and *Clinostomum* sp., while the dispersion of *G. alioides*, *D. longicauda* and *Acarina* gen. sp. was random. There were low species richness of parasites (2.45 ± 0.68), low Brillouin diversity index (0.52 ± 0.22), evenness (0.37 ± 0.15) and high Berger-Parker dominance (0.70 ± 0.15). The parasites community was characterized by low richness of species with high prevalence and low parasitic abundance. Host length had little influence on the abundance of parasites, but host behavior and availability of infective stages of parasites were the factors structuring the parasite community in *A. tetramerus*. This was the first study on parasites of *A. tetramerus* from the Jari River basin.

Key words: freshwater fish; cichlid; gills; ectoparasites.

PARASITOS DAS BRÂNQUIAS DE *Aequidens tetramerus*, CICLÍDEO DO BAIXO RIO JARI, UM TRIBUTÁRIO DO RIO AMAZONAS NO NORTE DO BRASIL

RESUMO

O objetivo deste estudo foi investigar pela primeira vez a fauna de parasitos das brânquias de *Aequidens tetramerus* do baixo Rio Jari, estado do Amapá (Brasil). Em janeiro de 2017, 31 espécimes de *A. tetramerus* foram coletados e necropsiados usando metodologias usuais para parasitologia de peixes. Um total de 1.181 parasitos foram coletados e 100% dos hospedeiros estavam parasitados por *Gussevía disparoides*, *Gussevía alioides*, *Posthodiplostomum* sp., *Clinostomum* sp., *Dolops longicauda* e *Acarina* gen. sp. A dominância foi de *G. disparoides* e houve dispersão agregada de *G. disparoides*, *Posthodiplostomum* sp. e *Clinostomum* sp., enquanto a dispersão de *G. alioides*, *D. longicauda* e *Acarina* gen. sp. foi randômica. Foi observada uma baixa riqueza de espécies de parasitos ($2,45 \pm 0,68$), baixa diversidade de Brillouin ($0,52 \pm 0,22$), baixa uniformidade ($0,37 \pm 0,15$) e elevada dominância de Berger-Parker ($0,70 \pm 0,15$). A comunidade de parasitos foi caracterizada por baixa riqueza de espécies com alta prevalência e baixa abundância parasitária. O comprimento dos hospedeiros teve pouca influência na abundância dos parasitos, mas o comportamento dos hospedeiros e a disponibilidade de formas infectantes de parasitos foram os fatores estruturando a comunidade de parasitos em *A. tetramerus*. Este foi primeiro estudo sobre parasitos de *tetramerus* da bacia do Jari River.

Palavras-chave: peixe de água doce; ciclídeo; brânquias; ectoparasitos.

INTRODUCTION

Jari River basin is located south of the State of Amapá and the extreme north of the State of Pará, with an area of approximately 57,000 m². This hydrographic basin covers the municipalities of Almeirim, in the State of Pará, Laranjal do Jari, Vitória do Jari and Mazagão, in the State of Amapá. This river rises in the Tumucumaque Mountain

on the border of Brazil with French Guiana, is approximately 780 km long and flows into the Amazon River, south of the State of Amapá. The forest along riverbanks is of tropical rainforest, and to the south of the basin there is the presence of anthropic actions, where the original vegetal cover gave place to agricultural activities and silvicultural farms (EPE, 2010; Amapá, 2012).

Aequidens tetramerus Heckel, 1840 is a Cichliformes of the Family Cichlidae with benthopelagic behavior, and endemic to South America, with distribution in the Amazon River basin system (Peru, Colombia, Ecuador, Brazil and Bolivia). It is also present in the Tocantins and Parnaíba rivers, Guyana, French Guiana, Suriname and Orinoco river basins, Venezuela and Colombia (Ferreira et al., 1998; Kullander, 2003; Froese and Pauly, 2018). It is one of the most colorful species of the genus *Aequidens*, mainly in the breeding period (Froese and Pauly, 2018). They can be caught in streams and floodplain areas with lentic, clear and shallow waters. It has omnivorous habit, feeding mainly on insects and secondarily on fish, crustaceans, arthropods and aquatic plants (Ferreira et al., 1998; Pinheiro et al., 2016; Cardoso and Couceiro, 2017; Froese and Pauly, 2018). Despite the high fish richness of the Jari River basin, few studies have been carried out on fish parasites (Oliveira et al., 2015; Oliveira et al., 2017a, 2017b, 2017c; Gonçalves et al., 2018; Santos et al., 2018). Parasites play a key role in natural ecosystems, mainly considering that their life cycles involve different hosts, either vertebrates or invertebrates. Hence, they play an important role in the ecological balance of the aquatic ecosystems, sometimes acting as a control mechanism for the size of wild fish populations (Oliveira et al., 2017b). In addition, parasites can cause grave pathologies for host fishes, with prejudicial to fishery and aquaculture.

In *A. tetramerus* from the Igarapé Fortaleza basin were reported protozoans *Ichthyophthirius multifiliis* Fouquet, 1876; *Tripartiella tetramerii* Martins, Marchiori, Bittencourt & Tavares-Dias, 2016 and *Trichodina nobilis* Chen, 1963; crustaceans *Dolops longicauda* Heller, 1857; monogeneans *Gussevia alioides* Kritsky, Thatcher & Boeger, 1986 and *Gussevia disparoides* Kritsky, Thatcher & Boeger, 1986; metacercariae of Digenea, larvae of nematodes Anisakidae gen. sp. and *Pseudoproleptus* sp.; larvae of cestodes Proteocephalidea and acanthocephalans *Gorytocephalus spectabilis* Machado, 1959 (Tavares-Dias et al., 2014; Martins et al., 2016). However, there are no other studies on parasites of *A. tetramerus*. Thus, the aim of this work was to study the parasites in gills of *A. tetramerus* from the lower Jari River, State of Amapá (Brazil).

MATERIAL AND METHODS

Fish and collection site

In January 2017 (rainy season), 31 specimens of *A. tetramerus* (12.8 ± 2.0 cm and 121.1 ± 22.2 g) were collected in the Bacabal Stream, in the lower Jari River, near Jarilândia riverine community in the municipality of Vitória do Jari (Figure 1), State of Amapá (Brazil), and subjected to parasitological analysis. Fish were collected with gill nets with different mesh sizes (ICMBio: 23276-1). This study was developed according to the principles recommended by the Brazilian College of Animal Experimentation (COBEA)

and with the authorization of the Embrapa Amapá Animal Ethics Committee (Protocol N° 014 - CEUA/CPAFAP).

Collection procedures and analysis of parasites

All fish were weighed (g) and measured for total length (cm), and then necropsied for parasitological analysis. The gills were removed and fixed in 5% formalin, and analyzed with the aid of microscope and stereomicroscope. Previously described techniques were used to collect, count, fix, preserve, and stain the parasites for identification (Eiras et al., 2006; Boeger and Viana, 2006).

To analyze the parasites, the ecological terms used were those recommended by Rohde et al. (1995) and Bush et al. (1997). The following parasite community descriptors were calculated at the infracommunity level: species richness, Brillouin diversity index (*HB*), Evenness (*E*), dominance index of Berger-Parker (*d*) and dominance frequency (percentage of infracommunities whose species was numerically dominant) were determined (Rohde et al., 1995; Magurran, 2004) using the software Diversity (Pisces Conservation Ltda, UK). The index of dispersion (ID), and the Poulain discrepancy index (D) were calculated using the Quantitative Parasitology 3.0 software to detect the distribution pattern of parasite infracommunity (Rózsa et al., 2000) for species with prevalence > 10%. The ID significance for each infracommunity was tested using the *d*-statistics (Ludwig and Reynolds, 1988). Spearman correlation coefficient (*rs*) was applied to determine correlations of parasite abundance with length, and weight of hosts and with the parasite diversity index (*HB*) and species richness (Zar, 2010).

RESULTS

In the gills of *A. tetramerus*, were collected a total of 1,181 parasites, being them: *Gussevia disparoides*, *Gussevia alioides*, *Posthodiplostomum* sp., *Clinostomum* sp., *Dolops longicauda* and undetermined mites. *Gussevia disparoides* and digenean species were the parasites prevalent and abundant. However, the dominance of *G. disparoides*, followed by digeneans *Posthodiplostomum* sp. and *Clinostomum* sp. (Table 1). These parasites presented aggregated or random dispersion with a high discrepancy (Table 2).

There was a predominance of hosts parasitized by 2 to 3 species (Figure 2).

Brillouin diversity index ranged from 0 to 1.02, evenness from 0 to 0.67, species richness of parasites varied from 1 to 4 and Berger-Parker dominance index, from 0.41 to 1.00 (Table 3). There was no correlation of host weight with the Brillouin diversity ($rs = 0.11$, $p = 0.54$) and parasite species richness ($rs = -0.06$, $p = 0.74$). In addition, there was no correlation between host length and the Brillouin diversity ($rs = 0.22$, $p = 0.22$) parasite species richness ($rs = 0.13$, $p = 0.48$).

A weak positive correlation of the abundance of *G. disparoides* and *G. alioides*, *Posthodiplostomum* sp. and *Clinostomum* sp. with host length was found. However, no correlation of parasite abundance with host weight was found (Table 4).

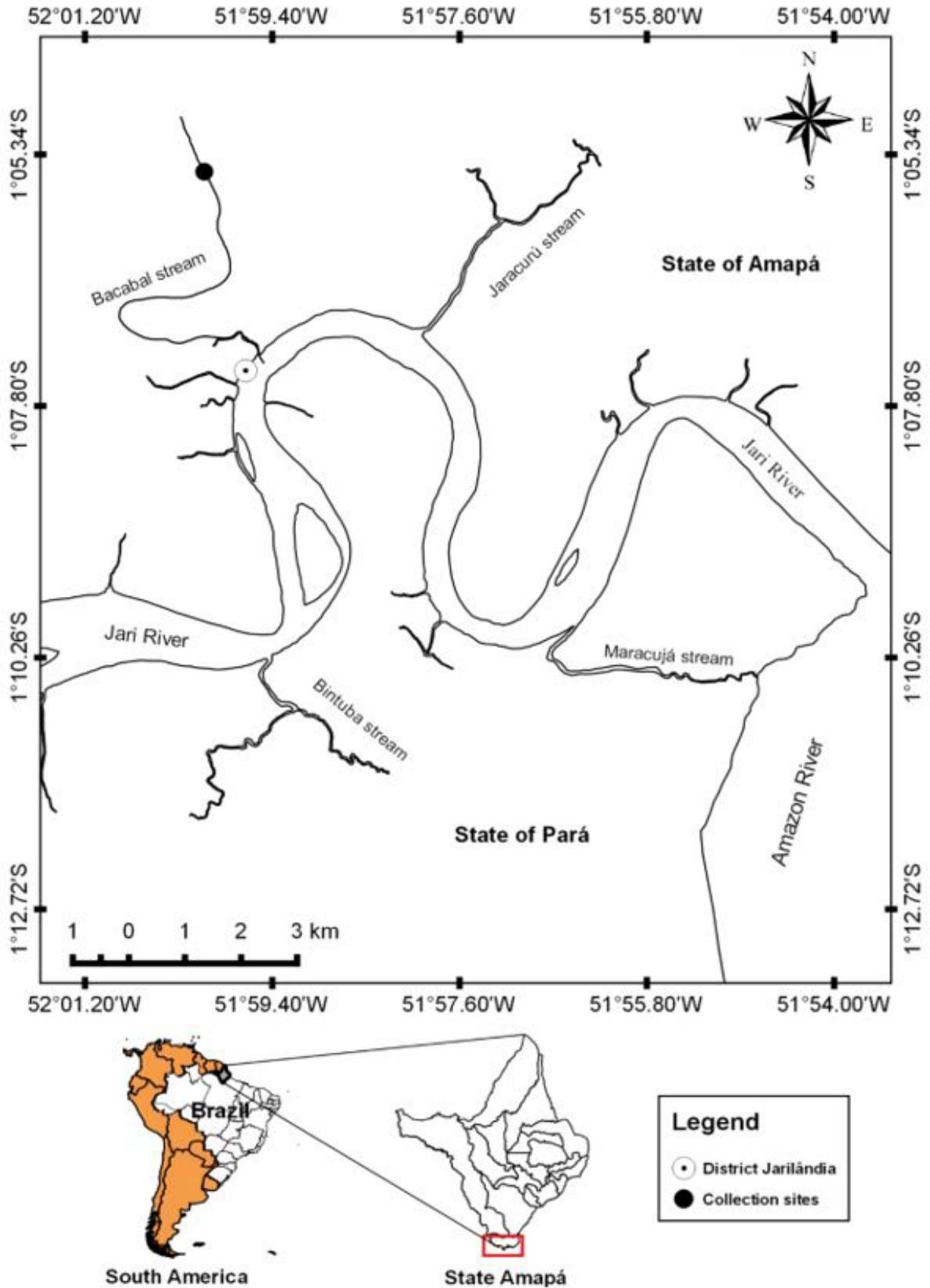


Figure 1. Collection site of *Aequidens tetramerus* in the lower Jari River, State of Amapá (Brazil).

Table 1. Parasites of *Aequidens tetramerus* gills from the lower Jari River, State of Amapá (Brazil).

Species of parasites	P (%)	MI	MA ± SD	Ranges	TNP	FD (%)
<i>Gussevia disparoides</i>	100	21.3 ± 10.9	21.3 ± 10.9	3-46	660	0.559
<i>Gussevia alioides</i>	12.9	1.2 ± 0.5	0.2 ± 0.4	1-2	5	0.004
<i>Posthodiplostomum</i> sp. and <i>Clinostomum</i> sp. (metacercariae)	87.1	17.7 ± 16.8	15.4 ± 16.8	1-60	479	0.406
Acarina gen. sp.	32.3	3.3 ± 6.6	1.1 ± 3.9	1-22	33	0.028
<i>Dolops longicauda</i>	12.9	1.0 ± 0	0.1 ± 0.3	1	4	0.003

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

Table 2. Dispersion index (ID), statistic-*d* and discrepancy index (D) for the parasites of *Aequidens tetramerus* gills from the lower Jari River, State of Amapá (Brazil).

Species of parasites	ID	<i>d</i>	D	Type of dispersion
<i>Gussevia disparoides</i>	1.855	2.87	0.295	Aggregated
<i>Gussevia alioides</i>	1.280	1.08	0.863	Random
<i>Posthodiplostomum</i> sp. and <i>Clinostomum</i> sp.	2.870	5.44	0.383	Aggregated
Acarina gen. sp.	1.305	1.17	0.723	Random
<i>Dolops longicauda</i>	0.900	-0.33	0.844	Random

Table 3. Descriptors of diversity for parasites communities of parasites in *Aequidens tetramerus* gills from the lower Jari River, State of Amapá (Brazil).

Descriptors	Mean ± SD	Ranges
Species richness of parasites	2.45 ± 0.68	1-4
Brillouin diversity index (<i>HB</i>)	0.52 ± 0.22	0-1.02
Evenness (<i>E</i>)	0.37 ± 0.15	0-0.67
Berger-Parker dominance index	0.70 ± 0.15	0.41-1

Table 4. Spearman correlation coefficient (*rs*) of the parasites abundance with length and body weight of *Aequidens tetramerus* from the lower Jari River, State of Amapá (Brazil).

Species of parasites	Peso (g)		Comprimento (cm)	
	<i>rs</i>	p	<i>rs</i>	p
<i>Gussevia disparoides</i> and <i>Gussevia alioides</i>	0.15	0.42	0.39	0.03
<i>Posthodiplostomum</i> sp. and <i>Clinostomum</i> sp.	0.28	0.12	0.45	0.01
Acarina gen. sp.	0.05	0.79	0.17	0.36
<i>Dolops longicauda</i>	-0.08	0.64	-0.06	0.75

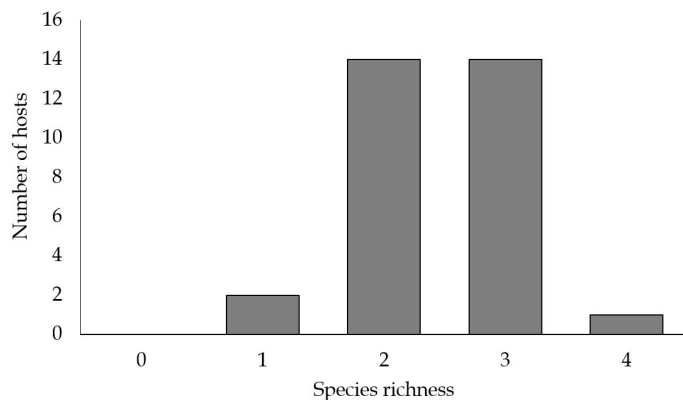


Figure 2. Species richness of parasites in *Aequidens tetramerus* gills from the lower Jari River, State of Amapá (Brazil).

DISCUSSION

In the *A. tetramerus* gills from the lower Jari River, the parasite community was composed of 2 species of Monogenea, 2 Digenea, 1 Crustacea and 1 Acarina gen. sp., with dominance of monogeneans *G. disparoides*. In contrast, the parasite community in the *A. tetramerus* gills from the Fortaleza Igarapé basin (AP) was composed of 2 species of Monogenea, 1 Digenea, 1 Crustacea and 3 Protozoa (Tavares-Dias et al., 2014). In gills of *A. tetramerus* of this study, there was no presence of protozoans *I. multifiliis* and trichodinid species as occurred in this host from the Fortaleza Igarapé basin (Tavares-Dias et al., 2014). The community structure and infracommunities of ectoparasites in wild host populations vary depending on geographical distance and habitat due to differences in abiotic factors, such as environmental characteristics (Dogiel, 1961; Oliveira et al., 2017b).

Gussevius disparoides, *Posthodiplostomum* sp. and *Clinostomum* sp. had an aggregated dispersion in *A. tetramerus*, while *G. alioides*, *D. longicauda* and mites had a random dispersion, which is typical of pathogenic species with little opportunity to colonize hosts (Guidelli et al., 2003). However, for *A. tetramerus* from the Fortaleza Igarapé basin, the parasite dispersion pattern was only aggregated (Tavares-Dias et al., 2014), a common dispersion pattern for freshwater fish (Guidelli et al., 2003; Neves et al., 2013; Tavares-Dias et al., 2014; Oliveira et al., 2017b; Gonçalves et al., 2018; Santos et al., 2018). In general, aggregated dispersion is mainly related to environmental factors, host behavior and immunological susceptibility to infection, as well as genetic factors, which control the size of parasite populations in hosts (Guidelli et al., 2003; Neves et al., 2013; Tavares-Dias et al., 2014; Gonçalves et al., 2018).

In wild fish populations, patterns among parasite communities can be detected using quantitative and qualitative descriptors (Magurran, 2004; Tavares-Dias et al., 2014; Oliveira et al., 2017b; Gonçalves et al., 2018; Santos et al., 2018). We observe that *A. tetramerus* collected in the rainy season showed a similar Brillouin diversity index and lower species richness of parasites than the described for this same host from the Fortaleza Igarapé basin, collected in the dry season. However, the Brillouin diversity index and species richness of parasite was greater than that reported for *A. tetramerus* in the rainy season (Tavares-Dias et al., 2014). Nevertheless, in addition to the seasonal period difference, the present study included only the parasite community of *A. tetramerus* gills, while Tavares-Dias et al. (2014) studied the ecto- and endoparasite communities of this same host. Therefore, such differences may also be related to environmental factors and the presence of viable intermediate hosts in the environment for those parasites with a heteroxenic life cycle (Takemoto et al., 2009; Tavares-Dias et al., 2017; Oliveira et al., 2017b).

Monogeneans, ectoparasites with direct and short life cycle, are present in fish from lentic and lotic environments, but they have a preference for lentic environments (Boeger and Viana, 2006; Tavares-Dias et al., 2017; Oliveira et al., 2017b; Gonçalves et al., 2018). High abundance of monogeneans in the fish gills can cause hypoxia due to injuries (Boeger and Viana, 2006). In gills of *A. tetramerus* from the lower Jari River, infestation of *G. disparoides* and *G. alioides* occurred, but *G. disparoides* presented the highest levels of parasitism, probably due to interspecific competition for microhabitat on the gills of hosts (Oliveira and Tavares-Dias, 2016). However, the prevalence was highest than that described by Tavares-Dias et al. (2014), and mean abundance was lower, while the mean intensity was similar. Therefore, such differences in infestation levels are related to environmental differences.

Metacercariae of *Clinostomum* spp. are digeneans belonging to the Clinostomidae with a complex life cycle and that have zoonotic potential for human (Bullard and Overstreet, 2008). *Posthodiplostomum* and *Clinostomum* species have mollusks as primary intermediate hosts and fish as secondary intermediate hosts, and fish eating-birds are definitive hosts (Bullard and Overstreet, 2008; Ritossa et al., 2013). In the *A. tetramerus* gills from the lower Jari River, there was infestation of metacercariae of *Posthodiplostomum* sp. and *Clinostomum* sp., while for this same host from the Fortaleza Igarapé basin was reported the

presence of encysted metacercariae (Tavares-Dias et al., 2014). The prevalence of *Posthodiplostomum* sp. and *Clinostomum* sp. in *A. tetramerus* was higher than described by Tavares-Dias et al. (2014), but mean intensity was lower and mean abundance was similar. Therefore, these differences were caused by the availability of infectious stages in different environments.

Usually, parasitic crustaceans presents a low abundance of infestation in wild fish populations (Oliveira and Tavares-Dias, 2016). In the *A. tetramerus* gills of the lower Jari River, there was low levels of infestation by *D. longicauda*, similar to what was reported by Tavares-Dias et al. (2014), for this same host. Generally, mites are found externally and internally in a variety of vertebrates, including fish and piscivorous birds (Gonçalves et al., 2018). Low levels of mite infestation occurred in *A. tetramerus* gills, similar to that reported by Gonçalves et al. (2018), for *Colossoma macropomum*. This is the first report of mites for *A. tetramerus*.

The body size of the hosts may or may not influence the community and infracommunities of parasites in host fish populations (Oliveira et al., 2017b), when reflects or not variations in the susceptibility to the parasites. For *A. tetramerus*, there was no correlation of host size and the Brillouin and diversity of species richness of parasites, but rather a weak positive correlation of length with abundance of monogeneans *G. disparoides* and *G. alioides*, as well as with *Posthodiplostomum* sp. and *Clinostomum* sp., similar to that reported for this same host by Tavares-Dias et al. (2014).

CONCLUSIONS

The ectoparasites community in *A. tetramerus* was characterized by a low species richness, low diversity index and low evenness, with moderate prevalence and low abundance and dominance of *G. disparoides* and metacercariae of digeneans. Host population length had little influence on parasites abundance, but no effect on parasites richness and diversity. The behavior of the hosts and the availability of infective stages of parasites in the environment were factors structuring the community of ectoparasites in *A. tetramerus*.

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