

Ectoparasites and endoparasites community of *Ageneiosus ucayalensis* (Siluriformes: Auchenipteridae), catfish from Amazon River system in northern Brazil

Drielly de Oliveira Ferreira¹ · Marcos Tavares-Dias¹

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Abstract This study investigated the community of ectoparasites and endoparasites in *Ageneiosus ucayalensis* (Auchenipteridae) of a tributary from the Amazon River system, in Northern Brazil. Of 34 fish examined, 100% were parasitized by *Ichthyophthirius multifiliis*, *Cosmetocleithrum bulbocirrus*, *Demidospermus* sp., metacercariae of *Genarchella genarchella*, *Clinostomum marginatum* and *Herpetodiplostomum* sp., *Procamallanus* (*Spirocamallanus*) *belenensis*, *Cucullanus ageneiosus* and larvae of *Contracaecum* sp. digeneans *C. marginatum* and *Herpetodiplostomum* sp. were dominant parasite species, while *I. multifiliis* was the parasite with higher infection level. Such parasite species showed an aggregated dispersion, except *P. (S.) belenensis*, which showed a random dispersion. The Brillouin diversity (0.53 ± 0.29) was high, while evenness (0.28 ± 0.16) and species richness of parasites (3.7 ± 1.1) were low. The size of the hosts did not influence diversity, species richness and abundance of parasites. The ectoparasites were characterized by high prevalence and abundance, while endoparasites community presented low prevalence and abundance. The main factors responsible for structuring the parasite community in *A. ucayalensis* were mainly the behavior of this host and the availability of endoparasites infective stages in the environment. This was the first report of *I. multifiliis*, *C. bulbocirus*, *Demidospermus* sp., *Contracaecum* sp., *C. marginatum*, *Herpetodiplostomum* sp. and *G. genarchella* for *A. ucayalensis*. The presence of endohelminth larvae suggests that *A. ucayalensis* is part of the diet of other fish

at the top of the food web in the Amazonian ecosystem studied.

Keywords Amazon · Diversity · Helminths · Parasitic infection · Freshwater fish

Introduction

The Amazon River system presents a high and complex diversity of different ecosystems that comprise an almost endless number of rivers, channels, small streams, beaches, flooded forests and floodplain areas inside temporary islands (Barletta et al. 2010; Junk 2013). Such ecosystems differ in sediment types, dissolved organic matter, ion content, pH and climatic variables (rain and temperature). The fish species of the different orders (Characiformes, Siluriformes, Perciformes, Tetraodontiformes, Gymnotiformes, Osteoglossiformes, Clupeiformes, Beloniformes, etc.) are also diverse, with different trophic levels occurring in the Amazon basin. In addition, there is a high number of endemic fish species (Barletta et al. 2010; Junk 2013; Froese and Pauly 2016). The Brazilian Amazon has habitats with different anthropogenic pressure histories (deforestation, agriculture, mining, urbanization, etc.; thus, it is made up of a mosaic of habitats, favoring the existence of highly varied environments). Among these habitats is the Igarapé Fortaleza basin, in the state of Amapá, eastern Amazon region, a complex ecosystem that suffers the effects of daily tides of the Amazon River, as well as of urban eutrophication (Cunha et al. 2004; Takiyama et al. 2012). The tides influence the velocity, turbidity and conductivity of this river (Cunha et al. 2004). In addition, the waters that periodically spread out across the floodplain are rich in nutrients because of the rapid decomposition of

✉ Marcos Tavares-Dias
marcos.tavares@embrapa.br

¹ Embrapa Amapá, Rodovia Juscelino Kubitschek, Km 5, 2600, 68903-419 Macapá, AP, Brazil

grasses, animal remains and the humus layer of the forest. This hydrodynamics leads to vegetation growth (Poaceae, Cyperaceae, Fabaceae, Onagraceae, Araceae, Asteraceae, Convolvulaceae and Lentibulariaceae) and invertebrate biomass (aquatic insects, zooplanktonic crustaceans and mollusks, etc.), which are used as food by fish species (Thomaz et al. 2004; Takiyama et al. 2012). This important tributary harbors a great diversity of fish species at all life stages including *Ageneiosus ucayalensis* Castelnau 1855, a fish that is the subject of this study.

Ageneiosus ucayalensis, popularly known as mandubé or ximbé, is a Siluriformes of the Auchenipteridae family and it is endemic from the Neotropical region and distributed in the Amazon, Orinoco and Paraná-Paraguay river systems (Sá-Oliveira et al. 2014; Froese and Pauly 2016). This fish occurs in the quiet zones of swamps or in the lower parts of streams and swims close to the bottom. The diet of this carnivorous fish consists mainly of microcrustaceans, fish and insects. It measures about 30 cm in maximum length, and its reproduction occurs in the Amazonian rainfall period. It has total spawning and sexual dimorphism in the period of reproduction, and its first sexual maturation occurs with about 15 cm of length (Santos et al. 2004; Sá-Oliveira et al. 2014; Froese and Pauly 2016).

Parasites play fundamental roles in natural ecosystems. They can regulate host population abundance, influence the diversity and composition of communities and stabilize food webs (Lizama et al. 2008; Rohlenová and Simková 2010; Alcântara and Tavares-Dias 2015; Oliveira et al. 2016). Sixteen species of parasites have been recorded for *Ageneiosus* species of different environments in South America; however, for *A. ucayalensis* few parasites species have been reported so far (Table 1). Most of these parasites of the *Ageneiosus* species have a life cycle that includes more than one host and infection via the trophic pathway, which increases the possibility of cumulative infections by adult and larval parasites. Therefore, little is currently available on parasitic fauna of *A. ucayalensis*, an Auchenipteridae whose main food resources suggest that this fish occupies a high position in the food web, thus increasing the probability of becoming infected by endoparasites larvae and adults. The knowledge on ecological aspects of the parasites of *A. ucayalensis* may elucidate the life cycle of species that this fish is harboring in a certain ecosystem.

In eastern Amazon (Brazil), the parasitic fauna of wild fish populations consists of diverse ectoparasites and endoparasites taxa (Bittencourt et al. 2014; Hoshino et al. 2014; Pantoja et al. 2015; Alcântara and Tavares-Dias 2015; Oliveira et al. 2016), but the structure of the parasite community of *A. ucayalensis* has not been studied yet. In addition to increasing knowledge on parasite–host–

environment relationship, such studies can indicate whether the presence or absence of some parasites in the ecosystem serves as bioindicators of environmental features (Lizama et al. 2008; Takemoto et al. 2009; Pantoja et al. 2015; Oliveira et al. 2016). Thus, this study investigated the structure of the parasite communities of *A. ucayalensis* in a tributary of the Amazon River system, in eastern Amazon, northern Brazil.

Materials and methods

Study area and fish collection

On October 2014, 34 specimens of *Ageneiosus ucayalensis* (20.6 ± 4.2 cm and 60.6 ± 42.1 g) were collected in the Igarapé Fortaleza basin, near the city of Macapá, in the state of Amapá, northern Brazil (Fig. 1), using nets (mesh 10–45 mm), for parasitological analysis. These fish were then placed in containers with ice (the euthanasia applied) and taken to the Laboratory of Aquatic Organisms Health of Embrapa Amapá, Macapá (Amapá of State, Brazil), for parasitological analysis. This study was developed in accordance with the principles adopted by the Brazilian College of Animal Experimentation (COBEA) and with authorization from the Ethics Committee in the Use of Animals of Embrapa Amapá (# 004 - CEUA/CPAFAP) and ICMBio (# 23276-1).

Parasite collection and analysis procedures

The fish were weighed (g) and their total length was measured (cm). Then, they were necropsied, and the mouth, gills, operculum and fins were examined for the presence of ectoparasites. The viscera and gastrointestinal tract were examined for the presence of endoparasites. The collection, fixation, preservation and preparation of the parasites for identification followed the recommendations of Eiras et al. (2006). To analyze the parasite infracommunities, the ecological terms used were also those recommended by Bush et al. (1997).

The following descriptors for the parasite community were calculated: species richness; Brillouin's diversity index (HB); evenness (E) in association with diversity index and Berger–Parker dominance index (d); and dominance frequency, i.e., the percentage of the infracommunities in which a given parasite species is numerically dominant (Rohde et al. 1995; Magurran 2004), using the Diversity software (Pisces Conservation Ltd., UK). The dispersion index (DI) and the discrepancy index (D) were calculated using the Quantitative Parasitology 3.0 software to detect the distribution pattern of the parasite infracommunities (Rózsa et al. 2000), for species with prevalence

Table 1 Parasites species reported for *Ageneiosus* Lacepède, 1803 from the South America

Hosts	Parasite species	Locality	References
Cestoda			
<i>Ageneiosus inermis</i>	<i>Luciaella ivanovae</i> Gil-Pertierra (2009)	Colastiné River, Argentina	Gil-Pertierra (2009)
	<i>Gibsoniela mandube</i> Woodland (1935)	Amazon River (AM), Brazil	Rego (1992)
	<i>Gibsoniela mandube</i> Woodland (1935)	Paraná River (PR), Brazil	Takemoto et al. (2009)
	<i>Ageneiella brevifilis</i> Chambrier and Vaucher (1999)	Colastiné River, Argentina	Gil-Pertierra (2009)
Nematoda			
<i>Ageneiosus ucayalensis</i>	<i>Cucullanus ageneiosus</i> Giese et al. (2010)	Guajará Bay (PA), Brazil	Giese et al. (2010)
	<i>Pseudoproleptus</i> sp.	Guamá River (PA), Brazil	Melo et al. (2011)
	<i>Procamallanus (Spirocamallanus) rarus</i> Travassos, Artigas and Pereira (1928)	–	Moravec (1998)
	<i>Procamallanus (Spirocamallanus) belenensis</i> Giese, Santos and Lanfredi (2009)	Guamá River (PA), Brazil	Giese et al. (2009)
	<i>Acanthocephala</i> gen. sp.	Paraná River (PR), Brazil	Takemoto et al. (2009)
<i>Ageneiosus militaris</i>	<i>Cucullanus pinnai pinnai</i> Travassos, Artigas and Pereira (1928)	Paraná River (PR), Brazil	Kohn et al. (2011)
	<i>Goezia</i> sp.	Paraná River (PR), Brazil	Moravec (1998); Kohn et al. (2011)
Digenea			
<i>Ageneiosus militaris</i>	<i>Clinostomidae</i> gen. sp.	Paraná River (PR), Brazil	Kohn et al. (2011)
<i>Ageneiosus inermis</i>	<i>Creptotrema lamothei</i> Curran (2008)	Paraguay River, Paraguay	Curran (2008)
	<i>Crepidostomum macrorchis</i> Szidat (1954)	Parana River, Argentina	Hamann (1988)
	<i>Cladocaeum tomasscholzi</i> Oréllis-Ribeiro et al. (2016)	Nanay River, Peru	(Oréllis-Ribeiro et al. 2016)
Crustacea			
<i>Ageneiosus inermis</i>	<i>Gamispinus diabolicus</i> Thatcher and Boeger (1984)	Amazon River (AM), Brazil	Thatcher and Boeger (1984)
	<i>Excorallana berbicensis</i> Boone (1918)	Amazon River (PA), Brazil	Thatcher (1995)
<i>Ageneiosus ucayalensis</i>	<i>Excorallana berbicensis</i> Boone (1918)	Araguari River (AP), Brazil	Vasconcelos and Tavares-Dias (2015)

>10%. The significance of the dispersion index for each infracommunity was tested using the *d* statistics (Ludwig and Reynolds 1988).

Fish weight (g) and total length (cm) data were used to calculate the relative condition factor (Kn) of the hosts, which was compared with the standard value (Kn = 1.00) using the *t* test. Body weight (g) and total length (cm) were also used to calculate the length–weight relationship ($W = aL^b$) after logarithmic transformation of length (L) and weight (W). Subsequently, two straight lines were fitted to the data, thus obtaining $\ln y = \ln A + B \ln x$ (Le-Cren 1951). The Spearman correlation coefficient (*rs*) was

used to determine possible correlations of parasite abundance with length, weight, parasite species richness and Brillouin's diversity of the hosts (Zar 2010).

Results

Of 34 *A. ucayalensis* examined, 100% were infected and 30,897 parasites were collected, being 1 species of protozoan, 2 monogeneans, 3 nematodes and 3 digeneans. There was a dominance of *Clinostomum marginatum* Rudolphi, 1819 and *Herpetodiplostomum* Dubois, 1936 (Digenea),

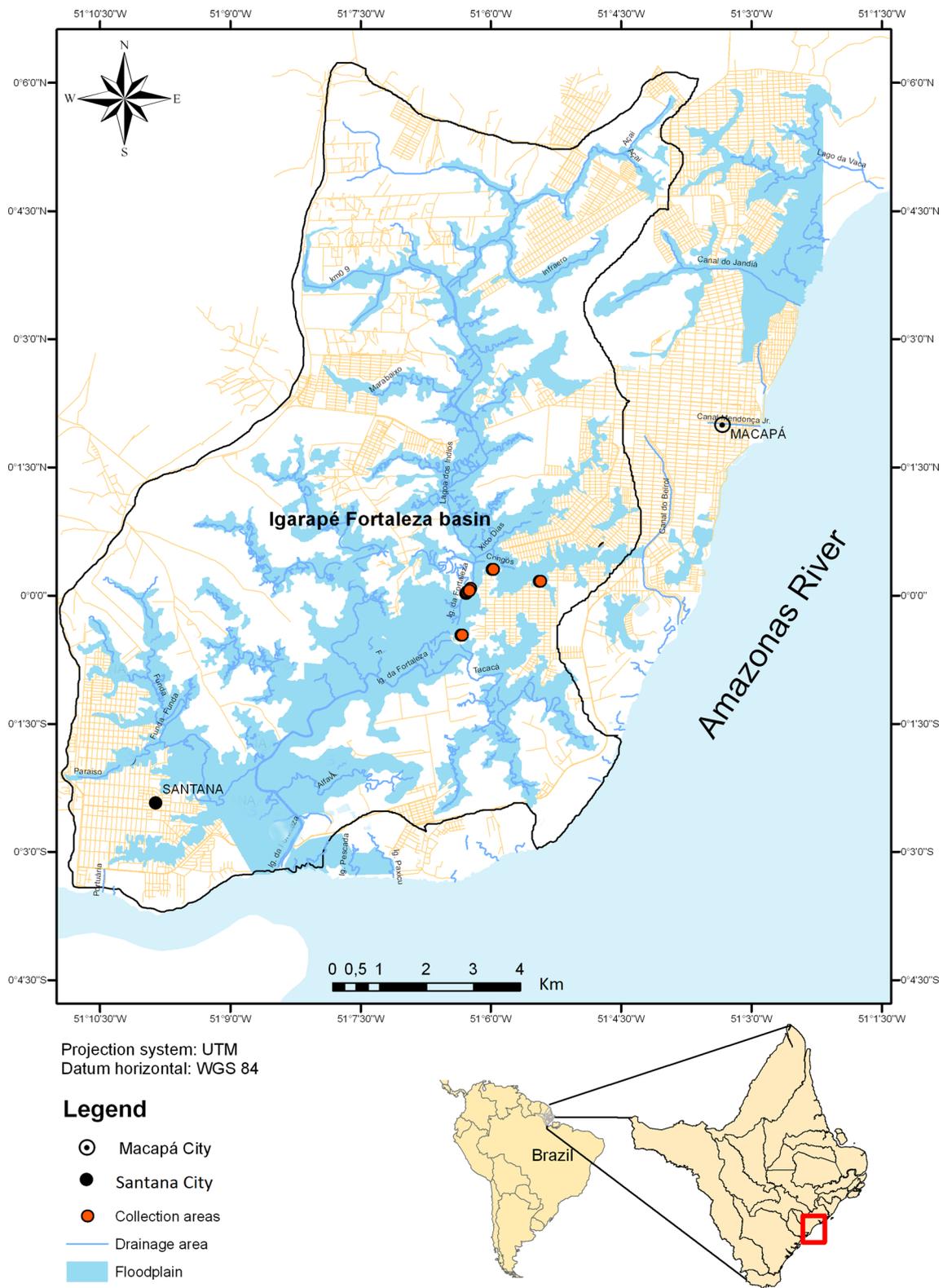


Fig. 1 Species richness of parasites in *Ageneiosus ucayalensis* from Amazon River system, in Brazil

but the highest infection levels were of the ectoparasites *Ichthyophthirius multifiliis* Fouquet, 1876 (Protozoa),

Cosmetocleithrum bulbocirrus Kritsky, Thatcher and Boeger, 1986, *Demidospermus* Suriano, 1983 (Monogenea)

Table 2 Parasites of *Ageneiosus ucayalensis* from Amazon River system, in Brazil

Parasites species	P (%)	MI	MA	FD (%)	Range	TNP	SI
<i>Ichthyophthirius multifiliis</i>	88.2	93.9	82.9 ± 65.5	0.091	18–243	2818	Gills
<i>Cosmetocleithrum bulbocirrus</i> and <i>Demidospermus</i> sp.	100	40.8	40.8 ± 49.5	0.045	2–250	1386	Gills
<i>Genarchella genarchella</i> (metacercariae)	64.7	23.3	15.1 ± 20.7	0.017	2–69	515	Gills
<i>Clinostomum marginatum</i> and <i>Herpetodiplostomum</i> sp. (metacercariae)	47.1	1634.1	769 ± 1058	0.846	2–2565	26,146	Intestine
<i>Contracaecum</i> sp. (larvae)	8.8	1.3	0.1 ± 0.4	–	1–2	4	Intestine
<i>Contracaecum</i> sp. (larvae)	5.9	1.0	0.1 ± 0.2	–	–	2	Abdominal cavity
<i>Procamallanus (Spirocamallanus) belenensis</i> (larvae and adults)	32.4	1.4	0.4 ± 0.7	–	1–2	15	Intestine
<i>Procamallanus (Spirocamallanus) belenensis</i> (larvae and adults)	8.8	1.4	0.1 ± 0.5	–	1–2	5	Abdominal cavity
<i>Cucullanus ageneiosus</i> (larvae)	8.8	3.0	0.2 ± 0.6	–	1–3	6	Intestine
<i>Cucullanus ageneiosus</i> (larvae)	5.9	1.0	0.1 ± 0.2	–	–	2	Abdominal cavity

P prevalence, MI mean intensity, MA mean abundance, FD frequency of dominance, TNP total number of parasites, SI site of infection

Table 3 Dispersion index (DI), *d*-statistic and discrepancy index (D) for the parasites infracommunities in *Ageneiosus ucayalensis* from Amazon River system, in Brazil

Parasites	DI	<i>d</i>	D
<i>Ichthyophthirius multifiliis</i>	2.161	3.88	0.373
<i>Cosmetocleithrum bulbocirrus</i> and <i>Demidospermus</i> sp.	1.979	3.37	0.299
<i>Genarchella genarchella</i>	2.869	5.70	0.517
<i>Clinostomum marginatum/Herpetodiplostomum</i> sp.	2.498	4.78	0.611
<i>Procamallanus (Spirocamallanus) belenensis</i>	1.207	0.86	0.652

Table 4 Diversity descriptors for communities of parasites in *Ageneiosus ucayalensis* from Amazon River system, in Brazil

Diversity indices	Mean ± SD (Range)
Species richness	3.7 ± 1.1 (2–6)
Brillouin’s index (<i>HB</i>)	0.53 ± 0.29 (0.03–1.11)
Evenness (<i>E</i>)	0.28 ± 0.16 (0.02–0.60)
Berger–Parker index (<i>d</i>) dominance	0.78 ± 0.17 (0.44–0.99)

and *Genarchella genarchella* Travassos, Artigas and Pereira, 1928 (Digenea). Among the endoparasites, the predominance was of nematodes such as *Contracaecum* Railliet and Henry, 1912, *Procamallanus (Spirocamallanus) belenensis* Giese et al. 2009 and *Cucullanus ageneiosus* Giese et al. 2010 (Table 2). The main parasite species of *A. ucayalensis* presented an aggregated dispersion pattern, except for *P. (S.) belenensis*, which showed a random dispersion (Table 3).

The mean Brillouin diversity index was high, while the mean evenness and mean parasite species richness of hosts were low (Table 4). The hosts were predominantly infected by 3 parasite species (Fig. 2). The lengths of the hosts were

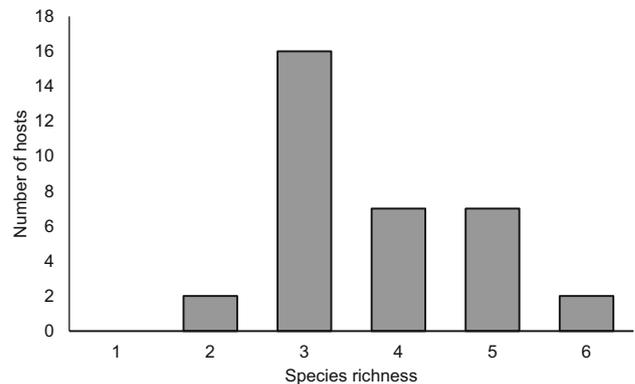


Fig. 2 Collection locality of *Ageneiosus ucayalensis* in a tributary from Amazon River system, in Brazil

not correlated with the Brillouin diversity ($r_s = 0.195$; $p = 0.270$) or with the richness of parasite species ($r_s = -0.185$; $p = 0.370$). However, only the abundance of *G. genarchella* presented a weak negative correlation with host length and weight of the hosts (Table 5).

For *A. ucayalensis*, the equation that described the length–weight relationship was $y = 0.0267x^{2.5176}$ ($r^2 = 0.852$), thus showing that growth was allometrically

Table 5 Spearman correlation coefficient (*rs*) for parasite abundance in relation to total length and body weight of *Ageneiosus ucayalensis* from Amazon River system, in Brazil

Parasites	Length		Weight	
	<i>rs</i>	<i>p</i>	<i>rs</i>	<i>p</i>
<i>Ichthyophthirius multifiliis</i>	0.299	0.086	0.222	0.207
<i>Cosmetocleithrum bulbocirrus</i> and <i>Demidospermus</i> sp.	0.251	0.152	0.268	0.125
<i>Genarchella genarchella</i>	−0.439	0.009	−0.358	0.037
<i>Clinostomum marginatum</i> and <i>Herpetodiplostomum</i> sp.	−0.083	0.639	−0.004	0.982
<i>Procamallanus (Spirocamallanus) belenensis</i>	−0.232	0.204	−0.288	0.099

negative, i.e., that there were greater increases in body mass than in size. The Kn (1.00 ± 0.05) of the hosts did not differ ($t = 0.0075$; $p = 0.994$) from the standard (Kn = 1.00) and did not show any correlation ($p = 0.05$) with the abundance of parasite species.

Discussion

For *A. ucayalensis*, only 5 nematodes and 1 crustacean species have been recorded, and such studies have been performed in hosts from eastern Amazon (Table 1). Therefore, this was the first report of *I. multifiliis*, *C. bulbocirrus*, *Demidospermus* sp., *Contraecaecum* sp., *C. marginatum*, *Herpetodiplostomum* sp. and *G. genarchella* for *A. ucayalensis*—parasites found in different organs of the hosts and in varied levels of infection. Moreover, this was the second record of *C. ageneiosus* and *P. (S.) belenensis* for *A. ucayalensis*. In fish, the structure and composition of the parasite communities is made up of a selection of the parasite species available in the environment (Choudhury and Dick 2000; Alcântara and Tavares-Dias 2015).

In wild fish populations, abundance, structure of the communities and infracommunities of parasites may vary according to various biotic and abiotic factors, such as sex, age, behavioral patterns, feeding diet of fish, availability of infective stages of parasites, presence of hosts adequate for colonization by parasites, migration pattern and quality of water. (Rohde et al. 1995; Choudhury and Dick 2000; Lizama et al. 2008; Takemoto et al. 2009; Rohlenová et al. 2010; Hoshino et al. 2014; Bittencourt et al. 2014; Pantoja et al. 2015; Alcântara and Tavares-Dias 2015; Oliveira et al. 2016). Thus, in *A. ucayalensis*, the communities of parasites were constituted by ectoparasites and endoparasites, but with a predominance of endoparasites larvae and adults, helminths with heteroxenic life cycle, as the diet of this carnivorous fish consists mainly of microcrustaceans, fish and aquatic insects (Santos et al. 2004; Sá-Oliveira et al. 2014; Froese and Pauly 2016). The diet based on infected mollusks caused infections by *C. marginatum* and

Herpetodiplostomum sp. in *A. ucayalensis*, and the diet based on infected crustaceans caused infections by *Contraecaecum* sp., *C. ageneiosus* and *P. (S.) belenensis*. Therefore, *A. ucayalensis* is a secondary intermediate host for these endoparasite species, because mollusk species are the primary intermediate host for *C. marginatum* and *Herpetodiplostomum* spp. and fish-eating bird are definitive hosts (Klaas 1963; Tavares-Dias et al. 2011), while crustaceans such as copepods are primary intermediate hosts for species of *Contraecaecum*, *Cucullanus* and *Procamallanus* (Moravec 1998; Anderson 2000).

Endohelminths are parasites known to modulate the population dynamics of invertebrate hosts and indirectly determine the diversity and structure of fish communities. Choudhury and Dick (2000) hypothesized that the endohelminth component communities in many tropical freshwater fish consist of lower species richness and have lower infection rates than fish from certain areas of temperate regions. Metacercariae of *C. marginatum* and *Herpetodiplostomum* sp. were the dominant parasites in *A. ucayalensis* and were the endoparasites with higher infection level. In contrast, low infection level by *Clinostomum* sp. and *Herpetodiplostomum* sp. was also reported for the forage fish, *Astyanax altiparanae* (Lizama et al. 2008). Therefore, the infections by metacercariae of these digeneans in *A. ucayalensis* suggest that the presence of intermediate and definitive hosts from the Igarapé Fortaleza (eastern Amazon, Brazil) were the causes of the infections. However, metacercariae of *G. genarchella* were found only in gills of *A. ucayalensis*, indicating direct contact with larval forms (cercariae) of this ectoparasite species in the environment (Morley 2012). Although the pathogeny of these digeneans for *A. ucayalensis* is not known, some metacercariae species cause a high pathogenicity to hosts (Dezfuli et al. 2016).

Cucullanus ageneiosus was found in low prevalence and abundance in *A. ucayalensis*. Giese et al. (2010) stated that *C. ageneiosus* is a nematode of brackish water fish. Larvae of *Contraecaecum* sp. are the most common anisakid of the Brazilian freshwater fish due to low parasitic specificity (Takemoto et al. 2009; Luque et al. 2011) and had a low

infection level in *A. ucayalensis* of this study. However, carnivorous fish, such as *A. ucayalensis*, have higher risk of infection by larvae of *Contracaecum* in comparison to omnivorous fish that are at the lower position in the food web (Hoshino et al. 2014). In contrast, *P. (S.) belenensis* is a nematode specific to *A. ucayalensis* and had moderate prevalence and low abundance and intensity. Nevertheless, Giese et al. (2009) reported a low prevalence and intensity of *P. (S.) belenensis* for this same host from the Guajar Bay (Par state, Brazil). Local factors may be the key determinants of abundant parasites in a locality. Moreover, different parasite species may have different intrinsic properties that determine their abundance in hosts.

In *A. ucayalensis*, from the Igarap Fortaleza basin, *I. multifiliis* and monogeneans *C. bulbocirrus* and *Demidospermus* occurred in the gills and were the parasites with the higher infection levels. This high parasitism level of monogeneans and ciliates may be due to the direct cycle of these ectoparasites, which is facilitated by a eutrophized environment suffering a high influence of urbanization (Hoshino et al. 2014; Pantoja et al. 2015; Oliveira et al. 2016). Monogeneans *C. bulbocirrus* and *Demidospermus* spp. are parasites mostly of siluriforms fish from South America. Brazil holds the highest number of these monogenean species in South America, and 24 species of *Demidospermus* are known mainly in fish in Brazil, Peru and Argentina (Cohen et al. 2013).

The body size of *A. ucayalensis* showed no correlation of the abundance with the ectoparasites and endoparasites species, unlike what was reported for *Lepomis macrochirus* (Neff and Cargnelli 2004) and *A. altiparanae* (Lizama et al. 2008). The condition factor has been recommended as an indirect indicator of body condition in fish populations (Le-Cren 1951; Neff and Cargnelli 2004; Rohlenov et al. 2010; Guidelli et al. 2011; Alcntara and Tavares-Dias 2015; Oliveira et al. 2016). However, the interactions between the parasite abundance and the condition factor may show distinct results. Negative or positive correlations have been reported between host body conditions and parasite abundance, suggesting the existence of a trade-off, with the decreasing or increasing host body conditions in response to abundance and pathogenicity of the parasites (Neff and Cargnelli 2004; Rohlenov and Simkov 2010; Guidelli et al. 2011), probably due to an energy investment in response to parasitism. As the potential impact of the parasites is obviously dependent on the infection levels, it achieves in a given host population, the body conditions of *A. ucayalensis* parasitized by ectoparasites and endoparasites were not affected by moderate infection levels.

In conclusion, the parasites community of *A. ucayalensis* was characterized by an overdispersion, low species richness, low uniformity and high diversity and was dominated by endoparasites species with low prevalence and

abundance. Furthermore, the host body size is not a determinant factor in the variation of parasite species richness and diversity among the *A. ucayalensis* population. Therefore, the main factors responsible for structuring the parasite community in *A. ucayalensis* were mainly the behavior of this host and the availability of endoparasites infective forms in the environment. The presence of endohelminth larvae suggests that this host is part of the diet of other fish at the top of the food web in the ecosystem studied. This was the first study on the structure of the parasite community in *A. ucayalensis*.

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