First study on parasites of *Hemibrycon surinamensis* (Characidae), a host from the eastern Amazon region

Primeiro estudo sobre parasitos de *Hemibrycon surinamensis* (Characidae), um hospedeiro na Amazônia oriental

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Abstract

This study was the first investigation of communities and infracommunities of parasites of *Hemibrycon surinamensis*. All the fish collected in a tributary of the Amazon river were parasitized by one or more parasite species. The Brillouin diversity index (*HB*) was 0.46 ± 0.28 and the mean species richness was 3.5 ± 1.2 parasites per host. A total of 14,734 parasites were collected, including *Ichthyophthirius multifiliis* and *Piscinoodinium pillulare* (Protozoa); *Jainus hexops* and *Tereancistrum* sp. (Monogenoidea); *Ergasilus turucuyus* and *Argulus* sp. (Crustacea); metacercariae of Derogenidae gen. sp.; metacercariae and adults of *Genarchella genarchella* (Digenea); and *Cucullanus* larvae and *Contracaecum* larvae (Nematoda). The dominant parasite was *I. multifiliis*, followed by *P. pillulare*. The parasites showed aggregated dispersion, except for *E. turucuyus*, which had random dispersion. The condition factor (Kn) indicated that the parasitism levels had not affected host body condition. The high levels of infection observed were due to host behavior, and this was discussed. This was the first report of *I. multifiliis*, *P. pillulare*, *Argulus* sp., *E. turucuyus*, *G. genarchella*, *J. hexops* and *Tereancistrum* sp. in *H. surinamensis*, and it expanded the occurrence of *E. turucuyus* and *G. genarchella* to the eastern Amazon region.

Keywords: Brazil, diversity, ectoparasites, freshwater fish, infracommunity.

Resumo

Este estudo é a primeira investigação sobre as comunidades e infracomunidades de parasitos de *Hemibrycon surinamensis*. Todos os peixes coletados no tributário do Rio Amazonas estavam parasitados por uma ou mais espécies de parasitos. O índice de diversidade de Brillouin (*HB*) foi 0,46 ± 0,28 e a riqueza de espécies foi 3,5 ± 1,2 parasitos por hospedeiro. Foram coletados 14.734 parasitos, entre *Ichthyophthirius multifiliis, Piscinoodinium pillulare* (Protozoa), *Jainus hexops* e *Tereancistrum* sp. (Monogenoidea), *Ergasilus turucuyus, Argulus* sp. (Crustacea), metacercárias de Derogenidae gen. sp., metacercárias e adultos de *Genarchella genarchella* (Trematoda), larvas de *Cucullanus* e larvas de *Contracaecum* (Nematoda). A dominância foi de *I. multifiliis*, seguida por *P. pillulare*. Os parasitos mostraram dispersão agregada, exceto *E. turucuyus*, que teve dispersão aleatória. O fator de condição (Kn) indicou que os níveis de parasitismo não afetaram as condições corporais dos hospedeiros. Foram discutidos os elevados níveis de infecção os quais estão associados ao comportamento do hospedeiro. Este é o primeiro relato de *I. multifilis, P. pillulare, Argulus* sp., *E. turucuyus, G. genarchella, J. hexops* e *Tereancistrum* sp. para *H. surinamensis*, expandindo a ocorrência de *E. turucuyus* e *G. genarchella* para a Amazônia oriental.

Palavras-chave: Brasil, diversidade, ectoparasitos, peixe de água doce, infracomunidade.

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Introduction

The Neotropical freshwater fish fauna has the highest diversity and species richness in the world (AZEVEDO, 2010). The order Characiformes is considered to be one of the most representative in freshwater environments of Brazil, and the family Characidae is the largest within this order of Neotropical fish. The family Characidae includes numerous species of small, medium and large size (REIS et al., 2003). Among the small-sized fish is the genus *Hemibrycon*, which has 32 species distributed in Panama, the Caribbean, Venezuela, Trinidad and Tobago, Brazil, Bolivia, Ecuador and Peru (BERTACO; MALABARBA, 2010; FROESE; PAULY, 2013).

Hemibrycon surinamensis Géry, 1962, the species that forms the focus of the present study, is only distributed in South America: in coastal basins of French Guiana and Suriname, and in the basins of the Tapajós, Tocantins and Xingu rivers in Brazil (REIS et al., 2003; BERTACO; MALABARBA, 2010; FROESE; PAULY, 2013). These fish have benthopelagic behavior; the males reach a maximum length of 8.0 cm and the females, 9.1 cm (FROESE; PAULY, 2013). In the eastern Amazon region, H. surinamensis is known as "matupiri" and is present in the basin of the Igarapé Fortaleza. This basin is located in the estuarine coastal sector, which is characterized by having extensive river-floodplain systems. These are physical river systems that fill up with accumulations of detritus, sedimentary material and organic matter, and are drained by freshwater and connected to a main water course. These systems are influenced by rainfall in the Amazon region and also by the daily tides of the Amazon River (TAKIYAMA et al., 2004). This unique ecosystem is rich in aquatic vegetation, especially macrophytes (THOMAZ et al., 2004), and therefore its lowland areas are widely used for shelter and food by many fish, including H. surinamensis.

Studies on the biology of H. surinamensis are still scarce, especially regarding its parasites and feeding habits. However, studies on parasites should be directed not only towards fish of economic importance, but also towards others, given the biological importance of fish in food chains (EIRAS et al., 2011). Several factors influence the diversity and structure of parasite infracommunities, such as the habitat, age, size and life history of the host fish, particularly regarding their trophic category and migratory capacity (GUIDELLI et al., 2003; TAVARES-DIAS et al., 2010; AZEVEDO et al., 2011; BELLAY et al., 2012). Knowledge about parasite infracommunities and their relationships with host fish is of great importance, since these parasites also play a key role in ecosystems by regulating the abundance or density of natural fish populations, thus stabilizing food chains and host community structures (ZRNČIĆ et al., 2009; TAKEMOTO et al, 2009; AZEVEDO et al., 2011). In this way, the present study was the first investigation on several aspects of parasites of H. surinamensis from a tributary of the Amazon river, state of Amapá, Brazil.

Materials and Methods

Fish and sampling site

From October to November 2011, 93 specimens of *Hemibrycon* surinamensis (11.1 \pm 0.7 cm and 24.5 \pm 3.7 g) were collected in the basin of the Igarapé Fortaleza (0°02'31.4" S, 051°05'52.18" W), Macapá (state of Amapá, Brazil), for parasitological analysis. The fish were collected using gill nets of different mesh sizes (ICMBio License: 23276-1) and were immediately transferred on ice to the Aquatic Organism Health Laboratory of Embrapa Amapá (in Macapá), for parasitological analysis.

Procedures for parasite collection and analysis

The fish collected were weighed (g) and measured for total length (cm). Each individual was macroscopically evaluated regarding body surface, mouth, eyes, opercula and gills. The gills were removed to collect ectoparasites. The gastrointestinal tract was removed and examined in order to collect endoparasites. All the parasites were collected, fixed, stained for identification (EIRAS et al., 2006) and quantified (TAVARES-DIAS et al., 2001). The parasitological terms adopted were those recommended by ROHDE et al. (1995) and BUSH et al. (1997). Voucher specimens were deposited at the Scientific and Technological Research Institute of the State of Amapá (Instituto de Pesquisas Científicas e Tecnológicas do Estado do Amapá, IEPA), in the Scientific Collection Curation Office for the Fauna of Amapá (Curadoria das Coleções Científicas, Fauna do Amapá, CCFA), under accession number IEPA 012-018-P.

The Brillouin index (*HB*), evenness (*E*), Berger-Parker dominance index (*d*) and species richness (MAGURRAN, 2004) were calculated for the parasite component community, by using the Diversity software (Pisces Conservation Ltd., UK). The dispersion index (ID) and the discrepancy index (D) were calculated using the Quantitative Parasitology 3.0 software, in order to detect the distribution pattern of each parasite infracommunity (RÓZSA et al., 2000) in species with prevalence \geq 10%. The significance of ID for each parasite species was tested using *d*-statistics (LUDWIG; REYNOLDS, 1988).

Data on body weight (g) and total length (cm) were used to calculate the relative condition factor (Kn) of the fish (LE CREN, 1951), which was compared with the standard value (Kn = 1.0) by means of the t test. The Pearson correlation coefficient (r) was used to check for correlations between host length and the Brillouin index (*HB*) and parasite abundance (ZAR, 2010), using the BioEstat 5.0 software.

Results

All the fish examined were parasitized by one or more parasite species. A total of 14,734 parasites were collected and these included *Ichthyophthirius multifiliis* Fouquet, 1876 (Ciliophora); *Piscinoodinium pillulare* Schäperclaus, 1954, Lom 1981 (Dinoflagellida); *Jainus hexops* Kritsky & Leiby, 1972; *Tereancistrum* Kritsky, Thatcher & Kayton, 1980 (Dactylogyridae); *Ergasilus turucuyus* Malta & Varella, 1996 (Ergasilidae); *Argulus* Muller, 1785 (Argulidae); metacercariae of Derogenidae gen. sp.; metacercariae and adults of *Genarchella genarchella* Kohn & Fernandes (1988) (Derogenidae); larvae of *Cucullanus* Muller, 1777 (Cucullanidae) and larvae of *Contracaecum* Railliet & Henry, 1912 (Anisakidae). Ectoparasite species predominated and *I. multifiliis* was the dominant parasite species, but endoparasite species were also found (Table 1). The parasites presented aggregated dispersion, except for *E. turucuyus*, which had random dispersion (Table 2).

The Brillouin diversity index (*HB*) was 0.46 ± 0.28 , the evenness (*E*) was 0.24 ± 0.15 , the dominance (*d*) was 0.78 ± 0.19 and the mean species richness was low (3.5 ± 1.2 parasites per host). The *HB* did not show any significant correlation (r = 0.121, p = 0.247) with total host length. Hosts parasitized by three to four parasite species predominated (Figure 1).

The condition factor (Kn = 1.000 ± 0.092 , t = 0.023, p = 0.982) of the parasitized fish did not differ from the standard value (Kn = 1.0), thus indicating that the parasitism had not impaired host body condition. The total host length only showed a positive correlation with the abundance of *Tereancistrum* sp. in the gills of *H. surinamensis* (Figure 2).

Discussion

The parasite community of *H. surinamensis* showed high diversity, consisting of two Protista, two Monogenoidea, two Crustacea, two Digenea and two Nematoda. The highest richness of ectoparasites (seven species) reflects environmental conditions that were favorable to their transmission, because they did not

need intermediate hosts. The low richness of endoparasites may be related to the living habits of this host, which occupies the second trophic level in the food chain. Infection by *I. multifiliis, P. pillulare, J. hexops, Tereancistrum* sp., metacercariae and adults of *G. genarchella* and *Contracaecum* sp. larvae presented aggregated dispersion, which is a common pattern among freshwater fish (GUIDELLI et al., 2003). Nevertheless, *E. turucuyus* showed random dispersion in the gills of *H. surinamensis*, which is typical of parasites with moderate or high pathogenicity, since these parasites regulate the population density of hosts, while overdispersion of parasites tends to stabilize the host-parasite relationship (MOLLER, 2006). Meanwhile, besides presenting high pathogenicity (MALTA; VARELLA, 1996), *E. turucuyus* appears to have presented reduced ability to colonize *H. surinamensis* due to competition with other species of parasites that were overdispersed in the gills.

Only one specimen of *Argulus* sp. was collected from the gills of *H. surinamensis* in the basin of the Igarapé Fortaleza, eastern Amazon region. *Argulus pestifer*, *Argulus multicolor*, *Argulus juparanaensis* and *Argulus amazonicus* have frequently been observed in different fish species in the Brazilian Amazon region (MALTA, 1998). *Ergasilus turucuyus*, another crustacean parasitizing the gills of *H. surinamensis*, presented low infection levels (prevalence = 10.8% and mean intensity = 1.1), in comparison with *Acestrorhynchus falcatus* and *Acestrorhynchus falcatus* in the Pacaás Novos river in the western Amazon region (MALTA; VARELLA, 1996). Because only these Characiformes hosts were infected by *E. turucuyus*, it seems that these copepods have higher specificity than other species of crustaceans.

Table 1. Parasites of *Hemibrycon surinamensis* in the Igarapé Fortaleza basin, eastern Amazon region, Brazil. FE: number of fish examined;

 FP: number of fish parasitized; P: prevalence; MI: mean intensity; MA: mean abundance; TNP: total number of parasites; IS: infection site.

FP: number of fish parasitized; P: preva	lence; MI: mean	intensity; MA	: mean abundance;	I INP: total numbe	r of parasites;	15: miection site.
Parasites	FE/FP	P (%)	MI (Range)	MA ± SD	TNP	IS
Ichthyophthirius multifiliis	93/61	65.6	197.1 (15-1890)	129.3 ± 246.4	12,025	Gills
Piscinoodinium pillulare	93/16	17.2	104.5 (15-450)	18.0 ± 61.9	1672	Gills
<i>Tereancistrum</i> sp.	93/87	93.5	6.4 (1-25)	6.0 ± 5.3	556	Gills
Jainus hexops	93/66	71.0	3.9 (1-19)	2.7 ± 3.4	255	Gills
Ergasilus turucuyus	93/10	10.8	1.1 (1-2)	0.1 ± 0.4	11	Gills
Argulus sp.	93/1	1.1	1.0	0.01	1	Gills
Derogenidae gen. sp. (metacercariae)	93/62	66.7	2.8 (1-24)	1.9 ± 3.0	176	Gills
Genarchella genarchella adults	93/5	5.4	1.2 (1-2)	0.3 ± 0.3	6	Intestine
Cucullanus sp. (larvae)	93/6	6.5	1.2 (1-2)	0.08 ± 0.3	7	Intestine
Contracaecum sp. (larvae)	93/14	15.1	1.8 (1-7)	0.06 ± 1	25	Intestine

Table 2. Dispersion index (DI), *d*-statistic, discrepancy index (D) and relative dominance (RD) for main parasites of *Hemibrycon surinamensis* in the Igarapé Fortaleza basin, eastern Amazon region, Brazil.

Parasites	DI	d	D	RD
Ichthyophthirius multifiliis	8.182	25.3	0.560	0.816
Piscinoodinium pillulare	6.333	20.6	0.347	0.113
Tereancistrum sp.	1.801	4.67	0.347	0.038
Jainus hexops	2.558	8.17	0.540	0.017
Ergasilus turucuyus	1.075	0.53	0.892	0.0007
Derogenidae gen sp.	1.938	5.35	0.560	0.012
Contracaecum sp.	1.443	2.77	0.870	0.002

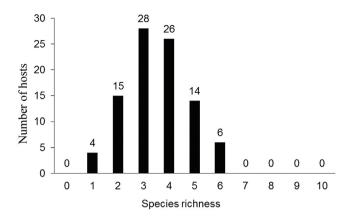


Figure 1. Species richness of parasites in *Hemibrycon surinamensis* in the Igarapé Fortaleza basin, eastern Amazon region, Brazil.

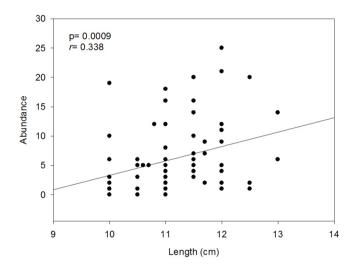


Figure 2. Correlation between the abundance of *Tereancistrum* sp. and total length of *Hemibrycon surinamensis* in the Igarapé Fortaleza basin, eastern Amazon region, Brazil.

Ichthyophthirius multifiliis was the dominant parasite in *H. surinamensis*, followed by *P. pillulare*, but infections by these protozoa occurred more frequently than in *Carnigiella martae* (TAVARES-DIAS et al., 2010), *Cobitis elongatoides* and *C. elongatus* in natural environments (ZRNČIĆ et al., 2009). However, neither of these parasites presents specificity, and high levels of parasitism are more frequent in lentic environments, such as rearing tanks (ZRNČIĆ et al., 2009; TAVARES-DIAS et al., 2010). Therefore, these high infection levels in *H. surinamensis* were influenced by the aggregating behavior of those hosts, which in general live under the macrophytes, where they spend most of their time on feeding.

In the gills of *H. surinamensis*, the level of infection by *Tereancistrum* sp. was relatively higher than by *J. hexops*. However, the prevalence of these monogenoidean species was higher than that of *Tereancistrum arcuatus* and *Jainus iocensins* in *Salminus brasiliensis* in the Paraná river, Brazil (COHEN et al., 2012). In addition, the abundance of *Tereancistrum* sp. increased with the length of *H. surinamensis*, thus indicating that accumulation occurs over the period of host growth. Similarly, in *Geophagus brasiliensis*, the abundance of *S. frequens* was also positively correlated with

its length (BELLAY et al., 2012). Six species of *Tereancistrum* are known and they parasitize fish of different host families in Brazil. There were also two species Bryconidae, one of Anostomidae and three of Prochilodontidae (COHEN et al., 2013). *Jainus hexops* was originally described in the gills of *Astyanax fasciatus* (Characidae) in Costa Rica (KRITSKY and LEIBY, 1972), and it has also been reported parasitizing *Moenkhausia sanctaefilomenae* (Characidae) in the Paraná river (TAKEMOTO et al., 2009) in Brazil. Therefore, the present report provided the third record of *J. hexops*, which also occurred in other Characidae species.

Metacercariae of Derogenidae, probably species of the genus *Genarchella*, were found at high levels of infection in the gills of *H. surinamensis*, in comparison with parasitism by metacercariae and adults of *G. genarchella* in the intestine of this same host. Kohn et al. (2011) also reported infection by *G. genarchella* in two specimens of *Pimelodus ornatus* (Siluriformes) in the Itaipu reservoir, state of Paraná. This digenean species has mollusks and Cypriniformes as intermediate hosts, and Characiformes and Siluriformes as definitive hosts (MARTORELLI, 1989; LEFEBVRE; POULIN, 2005). Thus, in the Igarapé Fortaleza basin, the Characiforme *H. surinamensis* may still be infected by this digenean through ingestion of mollusks (LEFEBVRE; POULIN, 2005) and/or direct contact with larval forms (cercariae) in the aquatic environment (MORLEY, 2012).

Larvae of Contracaecum sp. and Cucullanus sp. were detected at low infection levels in the intestine of *H. surinamensis*, as expected, since omnivorous fish have low risk of infection in comparison with carnivorous fish, which are at the top of the food chain. Low parasitism by Contracaecum sp. larvae has also been reported in relation to Metynnis lippincottianus, an omnivorous fish in the Paraná river basin (MOREIRA et al., 2009), as well as by Cucullanus zungaro in Hemisorubim platyrhynchos in the Baía river (GUIDELLI et al., 2003). The intermediate hosts of these nematodes are microcrustaceans (MORAVEC, 1998; MOREIRA et al., 2009), and H. surinamensis is the second intermediate or paratenic host for Cucullanus sp. In Brazil, 24 species of the genus Cucullanus mainly parasitizes Siluriforme species of freshwater and marine ecosystems, but a few species are known to have Characiformes hosts, including C. brevispiculus, C. mogi and C. pinnai pinnai (LUQUE et al., 2011).

In the Igarapé Fortaleza basin, *H. surinamensis* feeds on mollusks and microcrustaceans. Thus, it is an omnivorous fish with an intermediate position in the food chain of the fish community, possibly serving as a forage species for predatory fish that are the intermediate hosts of *Contracaecum* sp., an anisakid for which the definitive hosts are fish-eating birds and fish-eating aquatic mammals. This was the first report of these parasite species in *H. surinamensis*, and it expanded the occurrence of *E. turucuyus* and *G. genarchella* to the eastern Amazon region.

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