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Parasites in *Curimata cyprinoides* (Characiformes: Curimatidae) from eastern Amazon, Brazil

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ABSTRACT. This work is the first study on parasitic fauna of *Curimata cyprinoides* Linnaeus, 1766 (Curimatidae) and the host-parasite relationship. The total of 154,740 parasites such as *Ichthyophthirius multifiliis* (Ciliophora), trophozoite of *Spironucleus* sp. (Hexamitidae), *Urocleidoides* sp. (Monogenoidea), Digenea gen. sp. and *Polymorphus* sp. (Polymorphidae) were collected. The component community showed low diversity ($HB = 0.004 \pm 0.020$) and parasite species richness (1.4 ± 0.6). However, *I. multifiliis* and *Urocleidoides* sp. were prevalent and with higher intensity on the host population and also aggregated the distribution pattern. The occurrence of these ectoparasites in *C. cyprinoides* may be a consequence of its alimentary diet. Positive correlation between the abundance and size of *I. multifiliis* and the relative condition factor of the host were observed and discussed. These data represent increased knowledge of the biology of these parasites. Furthermore, this study expanded the geographic distribution of some parasite species for this new host from Brazil.

Keywords: ecology, gills, freshwater fish, Monogenoidea, protozoans.

Parasitos em Curimata cyprinoides (Pisces, Curimatidae) da Amazônia oriental, Brasil

RESUMO. Este trabalho providenciou o primeiro estudo sobre a fauna parasitaria de *Curimata cyprinoides* Linnaeus, 1766 (Curimatidae) e relação hospedeiro-parasito. Foram coletados 154.740 parasitos, tais como *Ichthyophthirius multifiliis* (Ciliophora); trofozoíto de *Spironucleus* sp. (Hexamitidae); *Urocleidoides* sp. (Monogenoidea); Digenea gen. sp. e *Polymorphus* sp. (Polymorphidae). A comunidade componente apresentou baixa diversidade ($HB = 0,004 \pm 0,020$) e riqueza de parasitos ($1,4 \pm 0,6$). *Ichthyophthirius multifiliis* e *Urocleidoides* sp. apresentaram a maior prevalência e intensidade na população de hospedeiro e tiveram padrão de distribuição agregado. Em *C. cyprinoides*, a ocorrência desses ectoparasitos pode ser uma consequência da ampla variedade de sua dieta alimentar. Correlação positiva da abundância de *I. multifiliis* como o tamanho e fator de condição relativo dos hospedeiros foi observada e discutida. Estes dados representam um aumento do conhecimento sobre a biologia desses parasitos. Além disso, este estudo ampliou a distribuição geográfica de algumas espécies de parasitos para este novo hospedeiro no Brasil.

Palavras-chave: ecologia, brânquias, peixe de água doce, Monogenoidea, protozoários.

Introduction

Curimatidae comprises about 130 fish species that can reach up to 30 cm in length. Benthopelagic and detritivorous fish, consume organic matter, algae, debris and microorganisms associated with the bottom of lakes and rivers. Most of species form large shoals and undertake trophic and reproductive migrations. In Amazon, Curimatidae fish species are important for commercial and riverine subsistence fishing (SANTOS et al., 2006; SOARES et al., 2011). In Brazil, the extractive fishing produced 5248.1 tons of *Curimata* spp. only in 2011 (MPA, 2013). *Curimata cyprinoides* Linnaeus, 1766 is native to South America and distributed in the Orinoco River delta and Atlantic drainage area of Guianas, lower Amazonas and Tocantins rivers (FROESE; PAULY, 2013). In the Brazilian Amazon this fish is known as "branquinha" and occur in the hydrographic basin from Igarapé Fortaleza (State of Amapá), an important tributary of Amazonas river, whose estuarine coastal sector is characterized for having extensive systemsriver floodplain, which constitute physical systems clogged river, drained by freshwater and connected to a water main course influenced by the high rainfall and tides from the Amazonas river.

The wetlands from eastern Amazon are widely used for refuge and food by many fish species 2004), (GAMA; HALBOTH, including C. cyprinoides. In spite of its importance in the region under analysis, the parasite fauna and parasite ecology of this host are poorly known. On gills filaments of C. cyprinoides captured from State of Rondônia (Brazil) the ergasilid Miracetyma etimaruya was identified by Malta (1993). Domingues and Boeger (2005) described Rhinoxenus guianensis, a monogenoidean from the nasal cavities of C. cyprinoides from Iracoubo in French Guiana. Therefore, C. cyprinoides has been rarely considered in studies on parasitic ecology. For few species of Curimatidae fish studies on parasite fauna have been carried out (ABDALLAH et al., 2005; AZEVEDO et al., 2011; KOHN; FERNANDES, 1987; KOHN et al., 2011; MALTA, 1993; MOLNAR et al., 1974; MORAVEC, 1998; THATCHER, 2006). However, fish parasites can also be useful as bioindicators of habitat degradation especially when sensitive species are sampled as sentinels. These bioindicators include parasites abundance, prevalence and species diversity, variables that might increase or decrease following the longterm exposure. The present investigation was conducted to evaluate the parasite fauna in a wild

population of *C. cyprinoides* from Brazilian Amazon and the host-parasite relationship.

Material and methods

Fish and locality of collection

In September, 2011, sixty-five specimens of *Curimata cyprinoides* were collected in wetlands from Igarapé Fortaleza basin (Figure 1), a tributary from Amazonas river in the municipality of Macapá, State of Amapá, Brazil (eastern Amazon) for parasitological analysis. All fish were collected with nets of different meshes.

Collection procedures and analyses of parasites

All fish were weighed (g) and measured for total length (cm), and then necropsied for parasitological analysis. For each specimen were examined the mouth, opercula, gills and gastrointestinal tract to parasites collection (protists and metazoans). Gills were removed and analyzed with the aid of a microscope. To quantify metazoan parasites, each viscera was dissected separately and washed in running water and all the material retained on a 154 μ m mesh was examined stereomicroscopically. Parasites were fixed, preserved and stained with standard techniques (EIRAS et al., 2006).

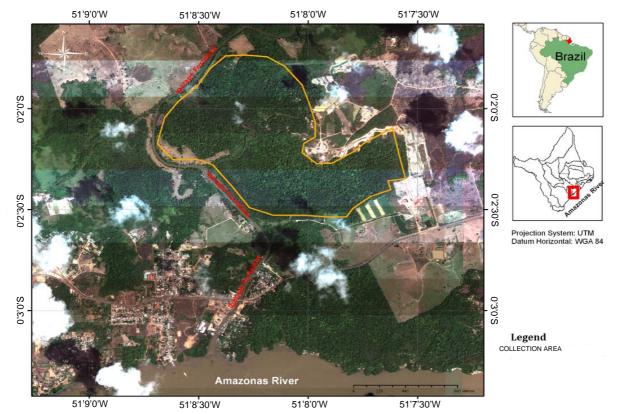


Figure 1. Collection locality of Curimata cyprinoides in wetlands from Igarapé Fortaleza basin, Amapá State (eastern Amazon), Northern Brazil.

Parasites of Curimata cyprinoides

The parasitological terminology used throughout follows that described by Bush et al. (1997). Dispersion index (ID) and discrepancy index (D) were calculated using the software Quantitative Parasitology 3.0 for detecting the distribution pattern of each parasite species in the infracommunity (ROZSA et al., 2000) with prevalent species \geq 10%. For each individual fish, the following parasite community descriptors were calculated at the infracommunity level (BUSH et al., 1997): Brillouin index (HB) and species richness, frequency of dominance (percentage of infracommunities in which a parasite species was numerically dominant) (ROHDE et al., 1995; MAGURRAN, 2004). Such diversity indexes were calculated using the software Diversity (Pisces Conservation Ltda, UK).

The data on body weight and total length were used to determine the relative condition factor (Kn) of fish hosts (LE CREN, 1951). The Pearson coefficient (r) was used to determine possible correlations of parasites intensity with the length, weight and Kn of hosts examined (ZAR, 2010).

The pH, temperature and dissolved oxygen were determined using digital devices for each purpose.

Results

In the study area, the mean pH was 6.6 ± 0.2 , mean temperature of $28.1 \pm 0.3^{\circ}$ C and mean dissolved oxygen levels of 2.0 ± 0.4 mg L⁻¹. The specimens of *C. cyprinoides* measured 9.4 ± 1.7 cm and 13.1 ± 6.6 g.

In C. cyprinoides were collected 154,740 parasites from five taxa, being two Protozoa species, the Ichthyophthirius multifiliis (Ciliophora) and Spironucleus sp. (Hexamitidae), monogenoideans Urocleidoides sp. (Dactylogyridae), encysted metacercarie and (Digenea) acanthocephalans Polymorphus sp. (Polymorphidae). However, there was dominance of ectoparasites on the gills, mainly I. multifiliis (Table 1). No parasite was found in the mouth and opercula of the examined hosts.

In C. *cyprinoides*, the component community of parasites showed low mean diversity (H = 0.004 ± 0.020) and low mean species richness (1.4 ± 0.6), with predominance of individuals parasitized by 1-2 species

(Figure 2). *Urocleidoides* sp. and *I. multifiliis* showed typical aggregate distribution pattern of infection (Table 2).

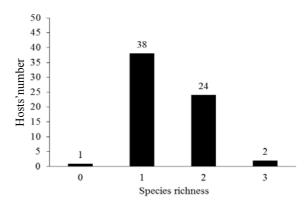


Figure 2. Parasite species richness in *Curimata cyprinoides* from Igarapé Fortaleza basin (eastern Amazon), Northern Brazil.

Table 2. Dispersion index, d statistic and discrepancy index for

 main parasite species in Curimata cyprinoides from Igarapé

 Fortaleza basin (eastern Amazon), Northern Brazil.

Parameters	I. multifiliis	Urocleidoides sp.	
Dispersion index	3.110	2.102	
d statistic	137.4	124.4	
Discrepancy index	0.324	0.826	

There was no correlation between the abundance of *Urocleioides* sp. and the total length (r = -0.0099, p = 0.9374) and body weight (r = -0.0597, p = 0.6366). In contrast, the abundance of *I. multifiliis* showed positive correlation with Kn (Figure 3) and total length and body weight (Figure 4) of hosts.

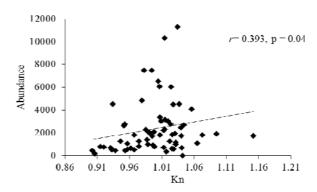


Figure 3. Correlation between relative condition factor (Kn) and the abundance of *I. multifiliis* in *C. cyprinoides* (n = 65) from Igarapé, Fortaleza basin (castern Amazon), Northern Brazil.

Table 1. Parasites in Curimata cyprinoides from Igarapé Fortaleza basin (eastern Amazon), Northern Brazil.

Parameters	I. multifiliis	Spironucleus sp.	Urocleidoides sp.	Digenea gen. sp.	Polymorphus sp
Examined fish	65	65	65	65	65
Parasitized fish	64	6	15	1	1
Prevalence (%)	98.4	9.2	23.1	1.5	1.5
Mean intensity	2416.8	1.3	3.8	2.0	1.0
Mean abundance	2379.6	0.12	0.9	0.03	0.01
Dominance (%)	99.9	0.005	0.04	0.002	0.0007
Range of intensity	174-11,285	1-3	1-20	-	-
Total number of parasites	154,672	8	57	2	1
Site of infection	Gills	Gills	Gills	Gills	Intestine

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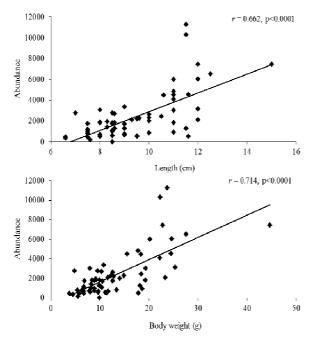


Figura 4. Correlation between abundance of *Iduthyophthirius multifiliis* and the total length and weight of *Curimata cyprinoides* (n = 65) from Igarapé Fortaleza basin (eastern Amazon), Northern Brazil.

Discussion

Curimata cyprinoides was parasitized by I. multifiliis, Urocleidoides Spironucleus sp., sp., Digenea metacercariae and Polymorphus sp., typical parasites of the tropical fish species from different environments, except Spironucleus sp. However, the component community of parasites in C. cyprinoides showed dominance of Urocleidoides sp. and I. multifiliis, with low diversity due to the number of parasite species in the site, response to the water currents, opportunity of transmission and infection, and consequently the probability of host infection. Differences in species richness and diversity of parasites may also be result of host's individual responses to parasitism and transmission rates (GUIDELLI et al., 2003; SOLER-JIMÉNEZ; FAJER-ÁVILA, 2012). Therefore, the ecological context of the population dynamics of parasites is important to understand host-parasite systems. Some environmental factors have a profound effect on several fish-parasite interactions. Overall, ectoparasites differ from endoparasites as the defense mechanisms tend to be externally reduced in fish.

Overdispersion of *I. multifiliis* and *Urocleidoides* sp. was found in *C. cyprinoides* and it can result in host mortality. Spatial heterogeneity is a feature common to many host-parasite systems, because differences in the infectivity of parasites or the host susceptibility will result in overdispersion of parasites in fish population (GUIDELLI et al., 2003;

SOLER-JIMÉNEZ; FAJER-ÁVILA, 2012). Aggregate dispersion of parasites can be attributed to different factors, such as host size, host density, and environment. The small body of *I. multifiliis* could also explain why these protozoans are most abundant than other metazoan ectoparasites, i.e. monogenoideans species. Therefore, understanding the mechanisms that generate the parasites distribution patterns of the individuals is the central tenet of ecology and naturally it has consequences for evolutionary dynamics.

Intestine is the primary site of infection, though hexamitids can invade other organs of the host. Spironucleus species causes systemic spironucleosis in wild freshwater fish (salmonids, cichlids, anguilids and cyprinids) and marine fish (salmonids and gadids) from North America, Asia and Europe (POYNTON et al., 2004; TANZOLA; VANOTTI, 2008; WILLIAMS et al., 2011). Hexamitids rarely reported in Neotropical have been fish (TANZOLA; VANOTTI, 2008), but the history of systemic spironucleosis in farmed salmonids is peculiar. Trophozoites of Spironucleus sp. were found only on gills of C. cyprinoides with low prevalence and mean intensity. This gill infection may be accidental, because no strong intestinal or systemic infections were observed. Prevalence and intensity of Spironucleus have been associated with host age. Cyst form facilitates the direct transmission of piscine Spironucleus spp. through the aquatic environment via fecal-oral route. However, the transmission via skin lesions as well as through the rectal route by both, cyst and trophozoite, have also been suggested (WILLIAMS et al., 2011). In South America, this piscine diplomonad was only described infecting the bile of Rhamdia quelen from Argentina (TANZOLA; VANOTTI, 2008). In this study, the genus Spironucleus was recorded for the first time in C. cyprinoides and the geographic distribution of this parasite was expanded to the eastern Amazon, in Brazil. Nevertheless, Spironucleus sp. infections have not been common in fish from Brazil. Despite the importance of these infections, studies on their epidemiology and ecology in wild fish populations are scarce.

Ichthyophthirius multifiliis is a widespread ciliate and well adapted to different environmental conditions since this parasite is nonspecific (TAVARES-DIAS et al., 2010). This parasite occurs mostly in environments with low oxygen levels as in this study. In addition, these protozoans are directly transmitted from fish to fish, thus the proximity among hosts might be very important for successful transmission. Hence, almost all farmed and wild freshwater fish are susceptible to these protozoan ectoparasites. In gills from *C. cyprinoides* was observed the dominance of *I. multifiliis* infection, because the abundance of this ciliate was directly correlated with the host fish growth. There was positive correlation between the *I. multifiliis* abundance and the condition factor (Kn) of the host; thus the hosts' body condition was not negatively affected. This positive correlation regarding the host growth is originated possibly from an accumulative infection, and this pattern was previously reported in others freshwater fish (OMEJI et al., 2010), once the host size is recognized as a factor influencing the parasite intensity and abundance.

Currently, 18 species of Urocleidoides Mizelle and Price, 1964 are known infecting the Neotropical freshwater fish such as Curimatidae, Characidae, Ctenoluciidae, Erythrinidae, Hypopomidae, Lebiasinidae, Poeciliidae, Anostomidae and Parodontidae. Thus, diversification of Urocleidoides spp. in the tropics is highly due to the opportunity to colonize and speciate on members from a wide number of piscine families (ROSIM et al., 2011) from South and Central America. On the other hand, only Urocleidoides curimatae (Dactylogyridae) is known infecting Curimatidae species, the wild Curimata argentea gills from Trinidad (MOLNAR et al., 1974). On gills of C. cyprinoides from Igarapé Fortaleza basin the low prevalence and mean intensity of Urocleidoides sp. was similar to those reported for Hyphessobrycon copelandi from middle Negro River (TAVARES-DIAS et al., 2010). Monogenoideans on fish in natural environment often occur in lower infection level, and site specificity can facilitate mating in these low-density populations for these parasites (SOLER-JIMÉNEZ; FAJER-ÁVILA, 2012).

In this study, only one digenean metacercarie was found on the gills of C. cyprinoides. In contrast, Tavares-Dias et al. (2011) observed high parasitism Posthodiplostomum Herpetodiplostomum by and metacercariae on gills of Astronotus ocellatus from a lake in eastern Amazon. High metacercariae infections were also reported on stomach, intestine, fins, gills and skin of Cichlasoma urophthalmus (JIMÉNEZ-GARCÍA; VIDAL-MARTÍNEZ, 2005), as well as on gills, muscles, intestine and fins from cyprinids fish (SAENPHET et al., 2008). Therefore, digeneans are usually parasites of intestine, but have also adopted new sites of infection in hosts such as fish gills. However, infection dynamics by digeneans can vary according to the environment and host (JIMÉNEZ-GARCÍA; behavior VIDAL-MARTÍNEZ, 2005; TAVARES-DIAS et al., 2011), because it can be related to water temperature, geographical latitude, longevity and particularly, the hosts' feeding habit.

Most endohelminthes trophically transmitted, such as acanthocephalans, are associated with a particular niche and host diet (TAKEMOTO et al., 2009; THATCHER, 2006). Thus, factors controlling host specificity of these fish parasites such as hosts habit and habitat are primarily ecological. Crustaceans represent possible intermediate hosts of Polymorphus species that may infect fish paratenic hosts in their complex life cycle (AMIN et al., 2010). In Brazilian freshwater fish (i.e. ocellatus, Geophagus brasiliensis Astronotus and Oligosarcus hepsetus), species of Polymorphus have not been described (SANTOS et al., 2008), because the adult forms are found in birds. For C. cyprinoides from Igarapé, Fortaleza State basin, only one Polymorphus sp. was found and this infection was accidental. Oligosarcus hepsetus, a paratenic host of the same acanthocephan genus, was reported with higher prevalence (10.0%) and intensity (1-10 parasites per host) (ABDALLAH et al., 2004). This study extends the occurrence of the genus Polymorphus to a new freshwater host, C. cyprinoides, and enlarged the geographic distribution of this parasite for Brazilian Amazon.

Conclusion

This study showed a quantitative analysis of parasitism in C. cyprinoides, which had low species richness. As the fish size can be a factor determining the parasite species richness, larger host species may harbor more parasites; thus this low species richness in C. cyprinoides could be due to the hosts small size, besides its inferior level in the food chain, and another factors. Hosts with higher parasitic intensity levels can be more easily prey to fish-eating fish; therefore, rarely these fish can be collected of a wild population. The parasites community of C. cyprinoides was dominated by I. multifiliis and Urocleidoides sp., both typical parasites of poor-condition environments as the Igarapé Fortaleza basin, which are suffering from a strong eutrophication impact of human actions. Thus, further studies might concentrate in this impact that may be affecting the parasites diversity and infection levels in this fish.

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