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ECOLOGY

Fish parasite diversity in the Amambai river, State Mato Grosso do Sul, Brazil

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ABSTRACT. This study aimed to describe the diversity of fish parasites in the Amambai River, in the Mato Grosso do Sul State, and generate information to facilitate studies of the biotic integrity of the region. During the period of September 2015 and July 2015, 48 specimens of 11 species of fish were analyzed for parasites. A total of 56.25% of the fish sampled from the Amambai River were infected with one or more metazoan species. A total of 21,514 parasite specimens belonging to 24 different species we found; they were distributed among six groups: Acanthocephala (Neoechinorhynchida), Cestoda (Proteocephalidea), Crustacea (Ergasilidae), Digenea (Cladorchiidae, *Dadaytrema*), Monogenea (Dactylogyridae) and Nematoda (Atractidae, Cucullanidae, Camallanidae, Rhabdochonidae). Of these, a monogenean, *Mymarothecium* sp. and four nematodes – *Cucullanus* sp.; *Procamallanus* (*Spirocamallanus*) *inopinatus* Travassos, Artigas, and Pereira, 1928; *Rabdochona acuminate* (Molin, 1860); and *Rondonia rondoni* Travassos, 1920 – were recorded for the first time on new hosts. This is the first work to gather information about the parasite fauna of fish from Amambai River in the Mato Grosso do Sul State, and provides records that contribute new reports of the occurrence of parasites in new locations.

Keywords: endoparasites; ectoparasites; biodiversity; parasite ecology.

Diversidade dos parasitas de peixes do rio Amambai, Estado do Mato Grosso do Sul, Brasil

RESUMO. Este estudo teve por objetivo descrever a diversidade dos parasitos de peixes do rio Amambai, Estado do Mato Grosso do Sul, a fim de gerar informações que possam servir como subsídios para estudos de integridade biótica da região. Entre o período de setembro de 2014 e julho de 2015 foram coletados e analisados 48 espécimes de peixes, pertencentes a 11 espécies. No rio Amambai, no período estudado, um total de 56,25% dos peixes estavam parasitados por pelo menos uma espécie de metazoário. Foram encontrados 21.514 espécimes de parasitos pertencentes a 24 espécies diferentes, distribuídos em seis grupos: Acanthocephala (Neoechinorhynchida), Cestoda (Proteocephalidea), Crustacea (Ergasilidae), Digenea (Cladorchiidae, *Dadaytrema*), Monogenea (Dactylogyridae) e Nematoda (Atractidae, Cucullanidae, Camallanidae, Rhabdochonidae). Destes, um Monogenea, *Mymarothecium* sp. e quatro Nematoda – *Cucullanus* sp.; *Procamallanus* (*Spirocamallanus*) *inopinatus* Travassos, Artigas, and Pereira, 1928; *Rabdochona acuminate* (Molin, 1860); e *Rondonia rondoni* Travassos, 1920 – foram registrados pela primeira vez em novos hospedeiros. Este é o primeiro trabalho que reúne informações sobre a fauna parasitária de peixes provenientes do rio Amambai, no Estado do Mato Grosso do Sul, onde os registros ampliam a lista de novos relatos de ocorrência de parasitos em novas localidades.

Palavras-chave: endoparasitos; ectoparasitos; biodiversidade; ecologia de parasitos.

Introduction

The continental waters of Brazil possess great species richness and diversity of living organisms, including parasites of natural fish populations (Agostinho, Thomaz, & Gomes, 2005). Parasites are components of most ecosystems and are involved in numerous food webs and all trophic levels. Most vertebrate species serve as a host of one or more parasite species (Lagrue, Kelly, Hicks, & Poulin, 2011). Among vertebrates, fish have the highest rates of infection by parasites because of the unique characteristics of their aquatic environment, which facilitate the spread, reproduction and life-cycle completion of each parasite group.

The parasite fauna of freshwater fish can vary in composition depending on the host species, its level in the food chain, the age, size and sex of individual fish, and other biotic and abiotic factors (Takemoto & Lizama, 2010).

Knowledge of the parasite fauna of fish can provide information on the biology of the host, the host-parasite relationship, zoonotic potential and/or importance for fish farming. Fish parasites also constitute an important instrument for biodiversity assessment (Abdallah, Azevedo, & Luque, 2004), and can help to more completely understand the biosphere (Luque & Poulin, 2007).

According to Pavanelli, Eiras, and Takemoto (2008), wild fish are parasitized by a great variety of species, however, they rarely show clinical signs of disease. This may be due to an equilibrium between their nutritional and physiological state and the environment, which may prevent the manifestation of diseases.

In this context, taxonomic studies of parasites are of great importance because one of the first steps in parasitology is to know what organisms are being dealt with. Furthermore, there remains a large number of species yet to be described (Ranzani-Paiva, Takemoto, & Lizama, 2004), since many fish are not necropsied with the intention of studying their parasites.

Knowledge regarding the parasite fauna of Brazilian fish, and especially in the Mato Grosso do Sul State, is poor, especially considering their great diversity. Therefore, the objective of the present study was to identify and list the species of fish parasites in the Amambai River, in order to generate information to facilitate studies of the biotic integrity of the region.

Material and methods

The parasite species list presented herein was based on samples collected at three sites on the Amambai River (Site 1: 23° 31' 192" S 53° 83' 460" W; Site 2: 23° 13' 803" S 54° 20' 133" W; Site 3: 22° 92' 257" S 54° 62' 900" W), in the Mato Grosso do Sul State, Brazil. Fish were captured during December 2014 and July 2015 by local fishermen using gill nets and cast nets of various mesh sizes.

Gills were removed from collected individuals for analysis of ectoparasites and the visceral cavity opened and the intestines exposed for analysis of endoparasites (according to Eiras, Takemoto, & Pavanelli, 2006). Parasites were then collected with the aid of a stereomicroscope.

The parasite species list provided here follows the classification and systematic arrangements of the following studies: Thatcher (2006) for acanthocephalans and cestodes; Boxshall and Halsey (2004) for crustaceans; Kohn, Fernandes, and Cohen (2007) for digenetic trematodes; Cohen and Kohn (2008) and Thatcher (2006) for monogenetic trematodes and Moravec (1998) for nematodes. In addition, checklists of the parasites of Brazilian fish, and works specific for each group of parasites, where used when necessary. This work was performed at the *Núcleo de Pesquisas em Limnologia, Ictiologia e Aquicultura* (Nupélia) of the Universidade Estadual de Maringá – Maringá (Paraná State).

Results and discussion

Forty-eight fish of 11 species were collected from the Amambai River for analysis: *Salminus brasiliensis* Cuvier, 1816 (n = 5), *Salminus hilarii* Valenciennes, 1850 (n = 1), *Serrasalmus marginatus* Valenciennes, 1837 (n = 3), *Raphiodon vulpinus* Spix and Agassiz, 1829 (n = 2), *Schizodon borelli* (Boulenger, 1900) (n = 3), *Prochilodus lineatus* Valenciennes, 1837 (n = 16), *Sorubim lima* (Bloch & Schneider, 1801) (n = 1), *Pseudoplatystoma corruscans* (Spix & Agassiz, 1829) (n = 3), *Auchenipterus osteomystax* (Ribeiro, 1918) (n = 1), *Pterodoras granulosus* Valenciennes, 1840 (n = 4) and *Pterygoplichthys ambroseti* (Holmberg, 1893) (n = 9). Of these, a total of 27 (56.25%) were parasitized by at least one metazoan species. Of the parasitized fish, 70.3% were parasitized by ectoparasites, 51.8% by endoparasites and 29.6% by both.

A total of 21,514 specimens of parasites of 24 different taxa were found (Figure 1, Table 1) (Acanthocephala – Neoechinorhynchida; Cestoda – Proteocephalidea; Crustacea – Ergasilidae; Digenea – Cladorchiidae, *Dadaytrema*; Monogenea – Dactylogyridae; and Nematoda – Atractidae, Cucullanidae, Camallanidae, Rhabdochonidae), including five new host records (NHR): a monogenean, *Mymarothecium* sp. Kritsky, Boeger, and Jégu, 1996; and four nematodes: *Cucullanus* sp.; *Procamallanus* (*Spirocamallanus*) *inopinatus* Travassos, Artigas, and Pereira, 1928; *Rabdochona acuminate* (Molin, 1860); and *Rondonia rondoni* Travassos, 1920.

The main sites of infection were the gills, where 12 different parasite species were collected including monogeneans and crustaceans. The intestine had the second highest number of species (five), including digeneans, cestodes, nematodes and acanthocephalans.

Salminus brasiliensis was the most parasitized fish species, with seven species of metazoan parasites. Of all the ectoparasites found, crustaceans exhibited the lowest parasitic specificity since the copepod *Ergasilus* sp. was found parasitizing three different species of fish (*P. lineatus*, *S. brasiliensis* and *S. marginatus*). The endoparasite with the lowest parasitic specificity was the nematode *Procamallanus*

(*Spirocamallanus*) *inopinatus* Travassos, Artigas, and Pereira, 1928, which was parasitizing two different host species (*R. vulpinus* and *S. marginatus*). The species *Rhabdochona acuminata* was the only component of the parasitic fauna of *Auchenipterus osteomistax*. *Rondonia rondoni* had the highest abundance of *Pterodoras granulosus* and *Pterigoplichthys ambrosetti*, with the latter being a new species for that host.

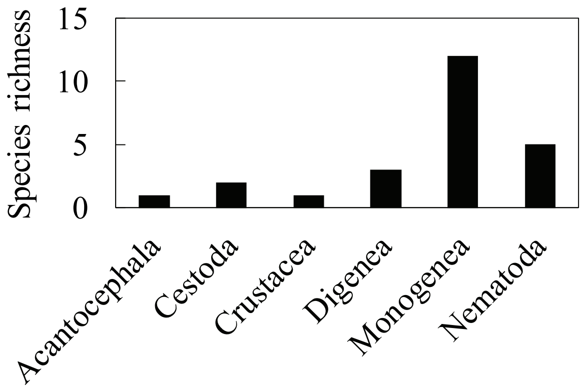

Figure 1. Species richness of fish parasites according to taxonomic group from the Amambai River, Upper Paraná River, Brazil.

Table 1. Species and total number of parasites recorded, their prevalence (%) and their hosts.

Parasites	PF	P (%)	Total n	Hosts
Amphocleitrhum paraguayensis	4	8.33	322	*Prochilodus lineatus* *Pseudoplatystoma corruscans*
Anacanthorus biscupidatus	1	2.08	3	*Salminus brasiliensis*
Anacanthorus contortus	1	2.08	3	*Salminus brasiliensis*
Anacanthorus daulometrus	1	2.08	13	*Salminus brasiliensis*
Anacanthorus douradensis	3	6.25	148	*Salminus brasiliensis*
Annulotrematoides bryconi	1	2.08	3	*Salminus brasiliensis*
Choanoscolex abscisus	1	2.08	81	*Pseudoplatystoma corruscans*
Cladorchidae gen. sp.	1	2.08	1	*Pterigoplychthys ambroseti*
Cosmetocleithrum bubocirrus	4	8.33	251	*Pterigoplychthys ambroseti* *Pterodoras granulosus*
Cuculanus sp.	1	2.08	2	*Sorubim lima*
Dactilogiridae gen. sp.	2	4.17	11	*Pseudoplatystoma corruscans* *Salminus hilarii*
Dadaytrema oxycephala	3	6.25	43	*Pterodoras granulosus*
Ergasilus sp.	3	6.25	10	*Prochilodus lineatus* *Salminus brasiliensis* *Serrasalmus marginatus*
Jainus iocenses	2	4.17	41	*Salminus brasiliensis*
Metacercaria	1	2.08	1	*Schizodon borelli*
Mymarothecium sp.	1	2.08	2	*Pterigoplychthys ambroseti*
Monticellia belavistensis	1	2.08	6	*Pterodoras granulosus*
Neoechynorhynchus curemai	4	8.33	31	*Prochilodus lineatus*
Procamallanus inopinatus	2	4.17	2	*Rhaphiodon vulpinus* *Serrasalmus marginatus*
Rhabdochona acuminata	1	2.08	9	*Auchenopterus osteomistax*
Rhabdochona sp.	1	2.08	2	*Prochilodus lineatus*
Rondonia rondoni	4	8.33	20320	*Pterigoplychthys ambroseti* *Pterodoras granulosus*
Tereancistrum sp.	1	2.08	1	*Pterigoplychthys ambroseti*
Vancleaveus janauacaensis	2	4.16	208	*Pterigoplychthys ambroseti* *Pterodoras granulosus*

PF: Individuals of parasitized fish; P (%): Prevalence (%).

Only three digeneans were encountered in the present study, *Dadaytrema oxycephala* and Cladorchidae as adults, and an unidentified metacercaria in larval form. Acanthocephalans were found only in *Prochilodus lineatus.*

There are more than 300 described species of monogeneans in Brazil, mainly belonging to two families, Dactylogyridae and Gyrodactylidae (Takemoto, Luque, Bellay, Longhini, & Graça, 2013). In the present study, we found only species of the family Dactylogyridae, which were collected from the gill arches of hosts. It is estimated that more than 95% of monogeneans are ectoparasites found on the gills or cutaneous regions of fish (Euzet & Combes, 1998). It appears as though most monogeneans have a high degree of parasitic specificity, occurring only on one host, or a few hosts that are phylogenetically very closely related (Cone & Kurt, 1982).

A new species of Monogenea, *Mymarothecium,* was reported for *Pterygoplichthys ambroseti.* This is the first report of a *Mymarothecium* infecting a species of Siluriformes, with the only previously known hosts being characids, including *Piaractus brachypomus*, *Piaractus mesopotamicus* and *Colossoma macropomum* (Cohen & Kohn, 2005; Leão, São-Clemente, & Cohen, 2015).

The nematode *Procamallanus inopinatus* was found on *Serrasalmus marginatus* and *Raphiodon vulpinus*, which is a new parasite record for the latter. Nematodes in general have low parasitic specificity (Takemoto & Lizama, 2010) and are common in freshwater fish. They can be found both in the larval stage, usually encysted, and as adults (Portz et al., 2013), and on practically any fish organ. According to Eiras, Takemoto, and Pavanelli (2010), this species of nematode has been identified in more than 51 different species of fish in Brazil.

The nematode *Cucullanus* sp. was found in the pimelodid *Sorubim lima,* which is a new record for that host. Nematodes of the genus *Cucullanus* also seem to have low parasitic specificity, and have been reported from other hosts from other localities (Azevedo, Abdallah, & Luque, 2011; Kohn et al., 2011; Yamada & Takemoto, 2013). According to Giese, Furtado, Lanfredi, & Santos (2010), 16 species of *Cucullanus* have been described from Brazil.

Nematodes of the genus *Rabdochona* were found parasitizing *Prochilodus lineatus* and are principally parasites of freshwater fish (Moravec, 2010). The present study represents a new parasite record for this host. The species *Rabdochona acuminate* was the only component of the parasite fauna of *Auchenipterus osteomistax,* representing 100% of the specimens collected. This nematode was reported from *A. osteomistax* by Tavernari et al. (2009) and in other hosts in other studies, such as *Astyanax fasciatus*, *A. bimaculatus* and *Geophagus brasiliensis* (Paraguassú & Luque, 2007), *Cichla kelberi* (Santos-Clapp & Brasil-Sato, 2014) and *Brycon amazonicus* and *B. melanopterus* (Ribeiro, Ueda, Pavanelli, & Takemoto, 2016).

Rondonia rondoni was found in *Pterodoras granulosus* and *Pterigoplichthys ambrosetti*, with it being a new parasite record for the latter. Studying *P. granulosus* on the Paraná River, Paraná State, and Ivinhema River, Mato Grosso do Sul State, Dias, Furuya, Pavanelli, Machado, and Takemoto (2004) reported high intensities of parasitic infection by this species, with approximately 13,168.82 parasites per fish analyzed. Campos, Takemoto, Fonseca, and Moraes (2009) encountered similar results with *Piaractus mesopotamicus*. Brasil-Sato & Santos (2003) reported finding this nematode in specimens of *Myleus micans,* in which they were obstructing the intestinal lumen of all the fish and filling all of the intestinal compartments with great intensity. According to Dias et al. (2004), no sign of apparent injury or damage to the host was found in any of these studies, indicating that the existing host-parasite relationship is not new, and the hosts have acquired resistance to the parasite, and the parasite has adapted to the host. Thatcher (2006) suggested that the relationship of these nematodes with the hosts is commensal in nature, since they are not fixed in the intestinal wall and are actively swimming in the intestine of the fish. This great abundance of nematodes is due to endogenous multiplication of the parasite, and infection of other hosts, such as *P. ambrosetti*. Infection occurs via ingestion of larvae or even adults present in the water column after having been excreted in the feces of infected fish, of which they comprise about 50% due to the huge number of parasites occupying the intestine. According to Azevedo et al. (2011), the high species richness of parasites of *P. granulosus* (five species) can be explained by the variety of foods consumed by this known omnivore, and the ease of ingestion of intermediate hosts and contamination via the food chain.

Digeneans are endoparasites that live in the intestine, but can be found in the visceral cavity and subcutaneously (Thatcher, 2006). *Dadaytrema oxycephala* has been reported in *mesopotamicus* in São Paulo State; the Miranda, Paraguai and Aquidauana rivers in Mato Grosso do Sul State (Campos et al., 2009; Kohn et al., 2011); in *Myleus mican*si in the São Francisco River, Minas Gerais State (Brasil-Sato & Santos, 2003); and in *granulosus* and *Pseudoplatystoma corruscans* in the Paraná River, Paraná State (Kohn et al., 2011). In the present study, the digenean family Cladorchidae was found in *P. ambrosetti*. Only one digenean larval form, the metacercaria, was found in *Serrasalmus marginatus*, with no occurrences in other hosts. The pathogenic significance of digeneans in fish is much more pronounced with infections by metacercaria than with adults because the metacercaria can encyst in any tissue or organ except cartilage or bone, and thus weaken the host (Thatcher, 2006).

The cestode *Monticellia belavistensis* was first described from *Pterodoras granulosus* by Pavanelli, Machado, Takemoto, and Santos (1994). According to these authors, the low number of parasites they found could be explained by the fact that *P. granulosus* is not piscivorous. This observation becomes even more evident when comparing its parasitic index with those obtained from piscivorous fish, especially pimelodids, which are the most intensely parasitized cestode hosts (Rego & Pavanelli, 1992). According to Kennedy (1994), insect larvae ingested by a host could function as a substitute for fish transmission, in particular the second intermediate or paratenic host, thereby allowing the completion of the life cycle of the species. The cestode *Choanoscolex abscisus* was observed to be dominant in *P. corruscans*. According to Rego (2002), proteocephalid cestodes are the major helminth found in this species and have been reported in the genus *Pseudoplatystoma* by Rego (2002), Campos, Fonseca, Takemoto, and Moraes (2008) and Ribeiro, Lizama, and Takemoto (2014), among others. The presence of adult cestodes in *P. corruscans* indicates that these hosts are part of the top level of the food chain, where they feed on fish, which are used as intermediate hosts in the life cycle of proteocephalids, and, in general, support parasitism by adult cestodes because they only remove the food necessary for their survival (Pavanelli et al., 2008).

In the present study, acanthocephalans were found only in *Prochilodus lineatus*. The parasite *Neoechinorhynchus curemai* had been previously found in *Prochilodus scrofa* (Noronha, 1984), however, it demonstrates a certain degree of specificity since most studies found it in the species *Prochilodus lineatus* (Ranzani-Paiva, Silva-Souza, Pavanelli, & Takemoto, 2000; Martins, Moraes, Fujimoto, Onaka, & Quintana, 2001; Santos et al., 2005; Lizama, Takemoto, & Pavanelli, 2005; 2006; Belo et al., 2013). Acanthocephalans are exclusive to the digestive tract of vertebrates (Eiras et al., 2010), and are considered pathogenic because they are endowed with a proboscis bearing hooks that pierce the intestinal wall causing lesions (Thatcher, 2006). Despite being comprised of a small number of species, this group is highly diversified compared to other metazoan parasite groups (Santos et al., 2013). This phylum has experienced great adaptive success and can parasitize all vertebrate classes and has a wide geographic distribution, occurring in marine, freshwater and terrestrial ecosystems on every continent (Kennedy, 1994). Acanthocephalans generally have biological cycles that include a wide variety of possible hosts, including invertebrates, fish, amphibians, birds and mammals, but the cycles always maintain the same pattern of transmission (Eiras et al., 2010).

Fish of natural populations have a more diversified diet than those in manmade lakes and reservoirs, which can positively affect the composition of endoparasites because the diet can include numerous animals that act as intermediate and/or paratenic hosts. Thus, the population of parasites with direct (monoxenous) life cycles, such as monogeneans, can increase or become prevalent in a community because of the ease of completing its life cycle in a single host, the ease of transmission and the reduction of host immune response (Mackenzie, 1999).

The diversity of species in parasite communities is the result of, among other factors, interactions between evolutionary history and ecology of hosts, and is associated with the diversity of intermediate and definitive hosts. According to Kennedy (1994), the species richness of parasites within a community at a site is considered a reflection of the number of host species present and the capacity for transmission and infection of the intermediate and definitive hosts in these environments. Furthermore, fish that have longer lives, and consequently longer exposure to parasites, facilitate the accumulation of parasites in the host (Takemoto et al., 2009).

Conclusion

The results presented here suggest that the density of populations of different host species may be the main determinant of species richness of parasites in the Amambai River. If we assume that each species of host can house an average of 10 species of parasites, and the number of host species in the Amambai River is vastly greater (data to be published), we can expect that the parasite biodiversity of this river is far from being even minimally known.

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