Digenea, Nematoda, Cestoda, and Acanthocephala, parasites in Potamotrygonidae (Chondrichthyes) from the upper Paraná River floodplain, states of Paraná and Mato Grosso do Sul, Brazil.

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Abstract: The present paper represents the first study on the endoparasitic fauna of *Potamotrygon falkneri* and *P. motoro* in the upper Paraná River floodplain. Fishes were collected by fishing rod and gillnetting in different stations of the floodplain, from March, 2005 to September, 2006. Parasites were sampled, fixed and preserved according to specialized literature. About half of the analyzed fish were parasitized by at least one of the following species of endoparasites: *Clinostomum complanatum, Genarchella* sp. and *Tylodelphys* sp. (metacercaria) (Digenea); *Acanthobothrium regoi, Rhinebothrium paratrygoni, Paroncomegas araya* and *Potamotrygonocestus travassosi* (Cestoidea); *Brevimulticaecum* sp. (larva), *Cucullanus* sp., *Echinocephalus* sp. and *Spinitectus* sp. (Nematoda); and *Quadrigyrus machadoi* (Acanthocephala). Some species were already registered in Chondrichthyes and others were previously recorded in Osteichthyes from the study area. The study listed ten new records of parasites in the host *P. falkneri*, one new record in the host *P. motoro* and five new records in the locality upper Paraná River.

Introduction

Stingrays of the family Potamotrygonidae are the only elasmobranches permanently adapted to the freshwater environment, inhabiting rivers of South America that discharge into the Atlantic Ocean and Caribbean Sea (Compagno and Cook 1995). According to Lovejoy et al. (2006), this group appears to have entered South America since the isolation of the continent by the Cretaceous opening of the Atlantic. Such taxa display a diversity of biogeographic and phylogenetic patterns, having members of endemic Neotropical freshwater radiations with distributions that encompass multiple river basins.

Potamotrygonidae is formed by three genera: *Paratrygon* Duméril, 1865, *Plesiotrygon* Rosa, Castello & Thorson, 1987, and *Potamotrygon* Garman, 1877. The two formers are monospecific, while *Potamotrygon* has approximately 26 valid species (Carvalho et al. 2003). Fifteen of these species have parasitological records, including the species of the present study, considering several regions: Salobra River (Paraguay River Basin, state of Mato Grosso, Brazil) (Rego 1979); vicinity of Corumbá (state of Mato Grosso do Sul, Brazil) (Brooks and Amato 1992); Amazonas River (Amazonas River Basin, Brazil) (Campbell et al. 1999); Orinoco River delta (Orinoco River Basin, Venezuela) (Marques and Brooks 2003); upper Madre de Dios River (Manu River Basin, Peru) (Reyda and Olson 2003).

In general, the species of potamotrygonids exhibit distribution restricted to a single basin or fluvial system - P. falkneri is restricted to Paraná-Paraguay Basin (Carvalho et al. 2003) and P. *motoro* is present in Amazonas River, Orinoco River and La Plata system (Araújo et al. 2004). In area. only the studv two species of Potamotrygonidae are registered: Potamotrygon falkneri Castex & Maciel, 1963 and P. motoro (Müller & Henle, 1841). These two species moved upstream from the middle to the upper Paraná River when the reservoir of Itaipu Hydroelectric Power Plant flooded Sete Quedas Falls, which was a physical barrier for fishes (Agostinho et al. 2004).

Pavanelli et al. (2004) summarized data concerning more than 80 identified species of parasites from 57 species of fishes in the region, disclosing great biodiversity. However, detailed

studies on the parasitic fauna of potamotrygonids were never performed in the study area. The identification of new hosts and new localities for parasites contribute to the knowledge of local biodiversity as well as for the understanding of the evolution of parasites and their hosts. The present study aimed to perform a survey of the endoparasites of potamotrygonids from the upper Paraná River floodplain, and constitutes the register of new occurrences. The study area is part of the upper Paraná River floodplain, Paraná River Basin, limit between the states of Paraná and Mato Grosso do Sul, near the city of Porto Rico (state of Paraná, 22°43' S, 53°10' W). The region is characterized as a fluvial floodplain, with periodic floods that result in the flooding of the marginal areas in the state of Mato Grosso do Sul. The area was characterized in details by Thomaz et al. (2004) (Figure 1).

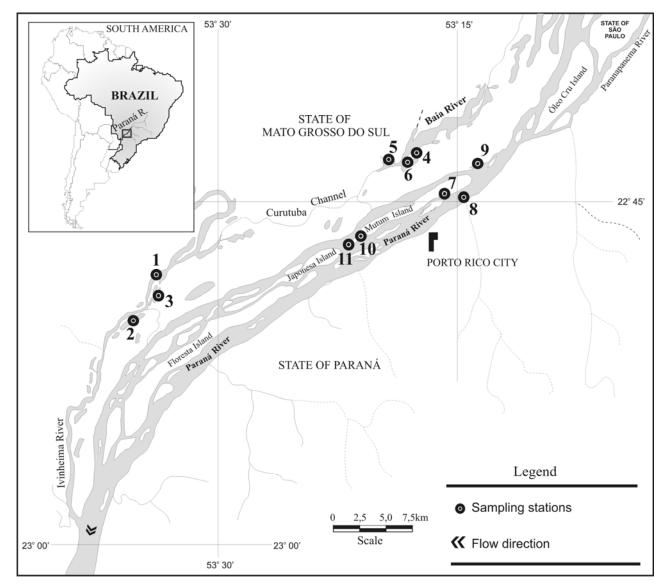


Figure 1. Upper Paraná River floodplain. Sampling stations: 1-Ivinheima River; 2-Ventura Lagoon; 3-Patos Lagoon; 4-Fechada Lagoon; 5-Guaraná Lagoon; 6-Baía River; 7-Pau Véio Backwater; 8-Paraná River; 9-Garças Lagoon; 10-Osmar Lagoon; 11-Mutum Island.

Materials and Methods

Fish were collected by fishing rod in the south margin of Mutum Island, from March 2005 to September 2006 (*P. falkneri*) and from April 2005 to December 2006 (*P. motoro*). Moreover, at the same time, fish were captured by gillnetting in different stations of Baía River, Ivinhema River and Paraná River (Figure 1). The samples were authorized by IBAMA (*Instituto Brasileiro do Meio Ambiente e dos Recursos Naturais Renováveis*), authorizations 003/2005 and 137/2006.

Sampling, fixation and preservation of parasites were according to Eiras et al. (2006). Identification of the parasites followed Szidat (1969), Rego and Dias (1976), Brooks et al. (1981), Fábio (1983), Moravec (1998), Campbell et al. (1999), Gibson et al. (2002), and Marques et al. (2003). The taxonomic classification follows Moravec (1998) for nematodes, Bush et al. (2001) for cestodes, and Gibson et al. (2002) for digeneans and acanthocephalans. Voucher specimens were deposited at *Coleção Helmintológica do Instituto Oswaldo Cruz*, numbers 36969 to 36982.

Results and Discussion

Among the 47 specimens of *P. falkneri* collected, 22 presented at least one species of endoparasites (46.8 %) and of the 19 especimens analyzed of *P. motoro*, 11 were parasitized (57.9 %). One hundred and seventy five endoparasites were collected in *P. falkneri*, including adults and larvae. In *P. motoro*, 22 adult endoparasites were collected. Taxa, sites of infection, and levels of parasitism are shown in Table 1. In total, three species of digeneans, five species of cestodes, four species of nematodes and one species of acanthocephalan were recorded (Table 1).

Table 1. Parasites taxa, deposit numbers in CHIOC (*Coleção Helmintológica do Instituto Oswaldo Cruz*), infection sites (ES = external wall of stomach, S = stomach and SV = spiral valvae), number of parasitized fish (PF) and range of the number of parasites in infected hosts (R) in *Potamotrygon falkneri* and *P. motoro* collected in the upper Paraná River floodplain from March 2005 to September 2006 (*P. falkneri*) and from April 2005 to December 2006 (*P. motoro*).

Parasites taxa	CHIOC numbers	Site of infection	Parasitized fish					
			Potamotrygon falkneri			Potamotrygon motoro		
			PF	MI	R	PF	MI	R
Digenea								
Clinostomum complanatum (metacercaria)	36971	SV	1	1	1	_	_	—
Tylodelphis sp. (metacercaria)	36970	SV	1	8	8	_		_
Genarchella sp.	36969	S	2	1	1	_		_
Cestoidea								
Acanthobothrium regoi	36972	SV	6	5.3	1–19	7	2	1-7
Rhinebothrium paratrygoni	36973, 36974	SV	17	5.2	1–26	4	1.5	1-11
Paroncomegas araya	36975	SV	3	3	1–5	_		_
Potamotrygonocestus travassosi	36976	SV	1	4	4	_		_
Nematoda								
Brevimulticaecum sp. (larva)	36977	ES	1	16	16	_		_
Cucullanus sp.	36978	SV	1	3	3	_		_
Echinocephalus sp.	36979	SV	2	1	1	_		_
Spinitectus sp.	36980	SV	1	3	3	_		_
Acanthocephala								
Quadrigyrus machadoi	36981,36982	SV	3	2	1–3	1	2	2

Phylum Platyhelminthes Class Trematoda Rudolphi, 1808 Subclass Digenea Carus, 1863 Order Strigeida (La Rue, 1926) Superfamily Clinostomoidea Lühe, 1901 Family Clinostomidae Lühe, 1901 Genus *Clinostomum* Leidy, 1856 *Clinostomum complanatum* Rudolphi, 1814

According to Szidat (1969), *Clinostomum complanatum* (= *Clinostomum marginatum* Rudolphi, 1819) (metacercaria) is one of the most common species of Clinostomidae and is distributed in the tropical and subtropical regions of Americas. Also according to Szidat (1969), metacercariae of *Clinostomum* are found encapsulated in several South American cichlids and free in the killifish *Neofundulus paraguayensis* (Eigenmann & Kennedy, 1903) (Cyprinodontiformes). In the upper Paraná River floodplain, *C. complanatum* was already registered encysted in *Gymnotus carapo* Linnaeus, 1758 (Gymnotiformes), *Parauchenipterus galeatus* (Linnaeus, 1766) (Siluriformes), *Hoplosternum littorale* (Hancock, 1828) (Siluriformes), and *Loricariichthys platymetopon* Isbrücker & Nijssen, 1979 (Siluriformes) (Pavanelli et al. 2004). Dias et al. (2003) studied the life cycle of *C. complanatum* in the same region, identifying the mollusk *Biomphalaria peregrina* (Orbigny, 1835) (Mollusca, Gastropoda) as first intermediate host and the Cocoi heron *Ardea cocoi* Linnaeus, 1766 (Aves, Ciconiiformes) and the Neotropical cormorant *Phalacrocorax brasilianus* (Gmelin, 1789) (Aves, Pelecaniformes) as main definitive hosts. There are no previous records of *C. complanatum* parasitizing potamotrygonids.

Superfamily Hemiuroidea Looss, 1899 Family Derogenidae Nicoll, 1910 Subfamily Halipeginae Poche, 1926 Genus *Genarchella* Travassos, Artigas & Pereira, 1928 *Genarchella* sp.

The genus *Genarchella* parasitizes anterior portions of the digestive tract (usually stomach) of freshwater fishes of Central and South Americas (Gibson 2002). According to Brooks and Amato (1992), there are records of *Genarchella overstreeti* (= *Paravitellotrema overstreeti* Brooks, Mayes & Thorson, 1981) in *Potamotrygon magdalenae* (Valenciennes, 1865) from Magdalena River (Colombia). *Genarchella fragilis* Lunaschi, 1990 was registered parasitizing *Pimelodus ornatus* Kner, 1858 (Siluriformes) in Paraná River by Fernandes and Kohn (2001). This is the first record of *Genarchella* sp. parasitizing *P. falkneri*.

Superfamily Diplostomoidea Poirier, 1886 Family Diplostomidae Poirier, 1886 Subfamily Diplostominae Poirier, 1886 Genus *Tylodelphis* Diesing, 1850 *Tylodelphis* sp. (metacercaria)

In the studied region, *Tylodelphys* sp. (metacercaria) was already registered parasitizing *Cyphocharax nagelli* (Steindachner, 1881) (Characiformes) (Pavanelli et al. 2004), *Gymnotus carapo* (Gymnotiformes) (Isaac et al. 2004), *Prochilodus lineatus* (Valenciennes, 1837) (Characiformes) (Lizama et al. 2005), and *Leporinus lacustris* Amaral Campos, 1945 (Characiformes) (Guidelli et al. 2006). There are no previous records of *Tylodelphys* parasitizing potamotrygonids.

Class Cercomeridea Brooks, O'Grady & Glen, 1985 Subclass Cercomeromorphae Bychowsky, 1937 Infraclass Cestodaria Monticelli, 1891 Cohort Cestoidea Rudolphi, 1808 Subcohort Eucestoda Southwell, 1930 Order Tetraphyllidea Carus, 1863 Family Onchocothriidae Braun, 1900 Genus Acanthobothrium van Beneden, 1848 Acanthobothrium regoi Mayes, Brooks & Thorson, 1981

According to Ivanov (2005), more than 150 species of *Acanthobothrium* were described parasitizing the digestive tract of elasmobranches, being only four species described parasitizing freshwater elasmobranches: *Acanthobothrium quinonesi* Mayes, Brooks & Thorson, 1978 in *Potamotrygon magdalenae*, from Magdalena River (Colombia); *Acanthobothrium amazonensis* Mayes, Brooks & Thorson, 1978 in *P. circularis* (Müller & Henle, 1841) from Itacuai River (Amazonas River Basin, Brazil); *Acanthobothrium regoi* in *P. hystrix* (Müller & Henle, 1841) from Orinoco River Delta (Venezuela) and *Acanthobothrium terezae* Rego & Dias, 1976 in *Potamotrygon motoro* from Salobra River (upper Paraguay River Basin, Brazil). *Acanthobothrium regoi* was described parasitizing the spiral valvae (intestine) of *P. hystrix* in Orinoco River (Brooks et al. 1981). Brooks and Amato (1992) also registered species of this taxon parasitizing *P. motoro* in the city of Corumbá (state of Mato Grosso do Sul, Brazil). There are no records of *A. regoi* parasitizing another species of potamotrygonid in the studied region.

Genus Potamotrygonocestus Brooks & Thorson, 1976

Potamotrygonocestus travassosi Rego, 1979

Potamotrygonocestus travassosi was described parasitizing the spiral valvae (intestine) of *Potamotrygon orbignyi* (Castelnau, 1855) in the Amazonas River (Maicuru, state of Pará, Brazil). Brooks et al. (1981) recorded the parasite in the delta of Orinoco River (Venezuela). Marques et al. (2003) in their redescription of the species also recorded *Paratrygon ayereba* (Müller & Henle, 1841), *Potamotrygon constellata* (Vaillant, 1880) and *Potamotrygon motoro* as new hosts, and the lower Amazonas River (state of Pará, Brazil) and Paraná Januacá (state of Amazonas, Brazil) as new localities. The present study registered *P. travassosi* for the first time parasitizing *P. falkneri* and it is the first record of the parasite in the upper Paraná River.

Family Phyllobothriidae Braun, 1900 Genus *Rhinebothrium* Linton, 1890 *Rhinebothrium paratrygoni* Rego & Dias, 1976

Rhinebothrium paratrygoni was described parasitizing the spiral valvae of the intestine of *P. ayereba* in the region of Salobra (upper Paraguay River Basin, Brazil). Later it was found in *Potamotrygon hystrix* and *Potamotrygon reticulatus* (Günther, 1880) in the delta of the Orinoco River (Venezuela), in *P. falkneri* in Paraná River Basin (near Hohenau, Paraguay) (Brooks et al. 1981) and in *P. motoro* in Corumbá (state of Mato Grosso do Sul, Brazil). In the present study, *R. paratrygoni* was found in *P. falkneri* and *P. motoro*, being this the first register for those species in the studied region.

Order Trypanorhyncha Diesing, 1863

Family Eutetrarhynchidae Guiart, 1927 Genus *Paroncomegas* Campbell, Marques & Ivanov, 1999

Paroncomegas araya (Woodland, 1934)

Paroncomegas araya corresponds to the cestode originally described as *Tentacularia araya* in *P. motoro* from Amazonas River (Brazil), and to *Eutetrarhynchus araya* described in *P. motoro* (Salobra River). In their work, Campbell et al. (1999) also registered as additional hosts *Potamotrygon reticulatus* (Garman, 1980) and *P. falkneri*, and they added the delta of Orinoco River (Venezuela), Paraná River (Paraguay) and Puerto Reconquista (Santa Fé, Argentina), respectively, as new occurrence localities. In the present study, *P. araya* was only found in *P. falkneri*, also the first register for the upper Paraná River floodplain. Besides the four species of adult cestodes, one larva was found in one specimen of *P. falkneri*, with 38.8 long by 12.2 width. The specimen could not be identified because of its early stage of development.

Phylum Nematoda

Class Rhabditea Inglis, 1983 Order Ascaridida Skrajabin & Schulz, 1940 Superfamily Ascaridoidea Railliet & Henry, 1915 Family Acanthocheilidae Wülker, 1929 Genus *Brevimulticaecum* Mozgovoy, 1951 *Brevimulticaecum* sp. larvae of Moravec, Prouza & Royero, 1997

A larva of *Brevimulticaecum* sp. was registered for the first time parasitizing potamotrygonids by Rego (1979) in *Paratrygon motoro*, Salobra River (upper Paraguay River Basin, Brazil). The specimens presented herein resemble the ones described by that author and were also found in the same site (stomach) in *P. falkneri*.

Family Cucullanidae Cobbold, 1864 Genus *Cucullanus* sp. Müller, 1777 *Cucullanus* sp.

The genus *Cucullanus* was registered in the upper Paraná river floodplain parasitizing *Serrasalmus* marginatus Valenciennes, 1837 (Characiformes), *Schizodon borelli* (Boulenger, 1900) (Characiformes), *Pseudoplatystoma corruscans* (Spix & Agassiz, 1829) (Siluriformes), *Pimelodus maculatus* Lacepède, 1803 (Siluriformes), *Hemisorubim platyrhynchos* (Valenciennes, 1840) (Siluriformes) and *Paulicea luetkeni* (= *Zungaro zungaro* Humboldt, 1821) (Siluriformes) (Pavanelli et al. 2004), being this study the first register of *Cucullanus* sp. parasitizing potamotrygonids.

Order Spirurida Chitwood, 1933 Superfamily Gnathostomatoidea Railliet, 1895 Family Gnathostomatidae Railliet, 1895 Genus *Echinocephalus* sp. Molin, 1858 *Echinocephalus* sp.

Deardorff et al. (1981) described *Echinocephalus daileyi* Deardorff, Brooks & Thorson, 1981 parasitizing the intestine of *Potamotrygon circularis* and *P. hystrix* in Letícia (Colombia), in the delta of Orinoco River (Venezuela) and Itacuai River (Amazonas Basin, Brazil). In the present study, only one specimen of the genus was found parasitizing *P. falkneri* (*Echinocephalus* sp.). There is no previous record of the genus *Echinocephalus* in Paraná River, neither parasitizing *P. falkneri*.

Superfamily Habronematoidea Chitwood & Wehr, 1932 Family Cystidicolidae Skrjabin, 1946 Genus *Spinitectus* Fourment, 1883 *Spinitectus* sp.

In the studied region, the genus *Spinitectus* was never recorded, even so there are registers of *Spinitectus asperus* Travassos, Artigas & Pereira, 1928 parasitizing Prochilodontidae and Anostomidae in Paraná River, in Emas Falls (Pirassununga, state of São Paulo, Brazil) and in the lower Paraná River (Argentina) (Moravec 1998). Still according to Moravec (1998), the genus parasitizes the digestive tract of marine and freshwater fishes and also amphibians. Due to the low number of specimens, the identification at species level was not possible. However, the genus could be identified by presenting cuticle with numerous transversal rings and by having a row of spines (Moravec 1998).

Phylum Acanthocephala

Class Eoacanthocephala Van Cleave, 1936

Order Gyracanthocephalida Van Cleave, 1936

Family Quadrigyridae Van Cleave, 1920

Genus Quadrigyrus Van Cleave, 1920

Quadrigyrus machadoi Fabio, 1983

The only Acanthocephala found in the present study was *Quadrigyrus machadoi*. This species was already registered in the upper Paraná River floodplain parasitizing *Hoplias* sp. (Characiformes: Erythrinidae) as adult and *Hemisorubim platyrhynchos* (Valenciennes, 1840) and *Cichla monoculus* Spix & Agassiz, 1831 (Perciformes) as larvae (Pavanelli et al. 2004). This is the first register of this species in potamotrygonidae from upper Paraná. According to Brooks (1992), the majority of the known parasites of Potamotrygonidae are restricted to this family of fish. However, four of eleven new records of parasites for Potamotrygonidae in the present study had already been registered in fishes of other families in the study region.

Conclusions

The following parasitic forms are recorded for the first time in Potamotrygon falkneri: Clinostomum complanatum (metacercaria), Genarchella sp., Tylodelphis sp. (metacercaria), Acanthobothrium regoi, Potamotrygonocestus travassosi. Brevimulticaecum sp. (larva), Cucullanus sp., *Echinocephalus* sp., *Spinitectus* sp., and Quadrigyrus machadoi. The present study also represents the first record of Q. machadoi parasitizing Potamotrygon motoro and A. regoi, Rhinebothrium paratrygoni, Paroncomegas arava, P. travassosi, and Echinocephalus sp. parasitizing fishes from the upper Paraná River.

Considering that the species *Clinostomum complanatum*, *Tylodelphis* sp. (metacercaria), *Brevimulticaecum* sp. (larva), *Cucullanus* sp., and *Spinitectus* sp. were found parasitizing only one specimen of *Potamotrygon falkneri*, it is soon to affirm that they were parasitizing this species. It is reasonable to think that *Potamotrygon falkneri* acted as an accidental host, once those species, or at least genera of parasites, are only known as parasites of other groups of fish (Osteichthyes) in the Paraná River Basin.

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