METAZOAN PARASITES OF *Conorhynchos conirostris* (VALENCIENNES, 1840), AN ENDEMIC SILURIFORM FISH OF THE SÃO FRANCISCO BASIN, BRAZIL

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ABSTRACT:- BRASIL-SATO, M. DE C.; SANTOS, M. D. DOS. **Metazoan parasites of *Conorhynchos conirostris* (Valenciennes, 1840) an endemic siluriform fish of the São Francisco basin, Brazil.** [Parasitos metazoários de *Conorhynchos conirostris* (Valenciennes, 1840), peixe siluriforme endêmico da bacia do São Francisco, Brasil.] *Revista Brasileira de Parasitologia Veterinária*, v. 14, n. 3, p. 160-166, 2005. Departamento de Biologia Animal, Universidade Federal Rural do Rio de Janeiro (UFRRJ), Km 7 da BR 465, CP 74539, Seropédica, RJ, Brazil, 23890-970. E-mail: mcbsato@ufrrj.br.

Specimens of *Conorhynchos conirostris* (Valenciennes, 1840) (Osteichthyes: Siluriformes) were collected from the upper São Francisco River (18º12'32''S, 45º15'41''W) in the municipality of Três Marias, Minas Gerais, Brazil, to investigate their parasitofauna. Of the 24 pirá fish collected, 8 were male (33.3%) and 16 were female (66.7%). Of this total, 12 were parasitized (50%). Nine species of parasites were found: *Helobdella* sp., *Creptotrema creptotrema* Travassos, Artigas & Pereira, 1928; *Palaeocryptogonimus claviformis* Szidat, 1954; metacercariae of *Clinostomum* sp. and of *Austrodiplostomum compactum* (Lutz 1928); *Procamallanus (Spirocamallanus)* sp. (young specimen); larvae of Anisakidae and *Rhabdochona* sp.; and *Neoechinorhynchus* sp. (young specimen). *Helobdella* sp. had the highest prevalence, followed by *C. creptotrema* (most abundant) and *P. claviformis*. *Creptotrema creptotrema* was dominant species in the parasite community of *C. conirostris*. There was no influence of the host size and sex on the prevalence and abundance of parasites, with *P. claviformis* being found only in male hosts. With the exception of *Procamallanus (Spirocamallanus)* sp., this was the first record of these parasites in *C. conirostris.*

KEY WORDS: *Conorhynchos conirostris*, leeches, helminths, anisakid larvae, biodiversity.

RESUMO

Espécimes de *Conorhynchos conirostris* (Valenciennes, 1840) (Osteichthyes: Siluriformes) foram coletados no alto São Francisco (18º12'32''S, 45º15'41''W), município de Três Marias, Minas Gerais, Brasil, para a investigação de sua parasitofauna. Dos 24 pirás coletados, oito eram machos (33,3%) e dezesseis eram fêmeas (66,7%). Do total, doze estavam parasitados (50%). Nove espécies de parasitos foram encontradas: *Helobdella* sp., *Creptotrema creptotrema* Travassos, Artigas & Pereira, 1928; *Palaeocryptogonimus claviformis* Szidat, 1954; metacercárias de *Clinostomum* sp. e de *Austrodiplostomum compactum* (Lutz 1928), *Procamallanus (Spirocamallanus)* sp. (espécime jovem), larvas de Anisakidae e de *Rhabdochona* sp. e *Neoechinorhynchus* sp. (espécime jovem). *Helobdella* sp. apresentou a prevalência mais elevada seguida de *C. creptotrema* (mais abundante) e *P. claviformis*. *Creptotrema creptotrema* foi dominante na comunidade parasitária de *C. conirostris*. Não houve influência do tamanho e do sexo dos hospedeiros sobre a prevalência e a abundância dos parasitos, sendo que *P. claviformis* foi encontrada somente em hospedeiros machos. Com exceção de *Procamallanus (Spirocamallanus)* sp., os parasitos são registrados pela primeira vez em *C. conirostris.*

PALAVRAS-CHAVE: *Conorhynchos conirostris*, leeches, helmintos, larvas de Anisakidae, biodiversidade.

INTRODUCTION

There are some 150 fish species found in the São Francisco Basin (SATO; GODINHO, 1999), with many of these being endemic (MENEZES, 1996), as is the case of *Conorhynchos conirostris* (Valenciennes, 1840) (FERRARIS, 2003), chosen as a symbolic species (SATO, 1999). Known by the popular

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name of pirá or pirá-tamanduá (BRITSKI et al., 1984; FERRARIS, 2003), these fish feed mainly on mollusks along with the larvae and pupae of insects, worms and microcrustaceans (IHERING, 1933; AZEVEDO; VIEIRA, 1940). It has been classified by Alvim (1999) as an aquatic invertivore. It reaches a total length of 100cm and body weight of 13kg (SATO, 1999). It is a migratory fish (SATO; GODINHO, 2003) and is on the official list of endangered fauna of the state of Minas Gerais (LINS et al., 1997) and the national list of endangered fish and aquatic invertebrates, issued by the Ministry of the Environment (2004).

There is growing interest in inventorying Brazil's biodiversity to detect systems whose species and areas are at risk of disappearing without knowledge of the existing taxa and interrelationships with the biome. This alone justifies the need for studies seeking to catalog and/or describe taxa at specific and/or higher levels, deposit specimens of recognized taxa and estimate the groups of organisms of the great Brazilian biomes, such as the Caatinga (scrublands), Cerrado (savanna) and Pantanal (marshlands), which have been poorly studied (LEWINSOHN; PRADO, 2002). Another priority is to estimate the endemism, mainly in our continental aquatic systems (ROCHA, 2002). The São Francisco River Basin is mainly located in Cerrado and Caatinga regions, two of the biomes little studied in Brazil. Our studies of the fish parasitology of the upper São Francisco demonstrate an intricate relationship among the various species of aquatic invertebrates (parasites, intermediate hosts and vectors) and vertebrates (mainly fishes and birds). Expanded icthyoparasitology (BRASIL-SATO; PAVANELLI, 1998; 1999; 2004; BRASIL-SATO, 2003) will contribute to the knowledge of the biodiversity of this aquatic system, considered of high endemic level. In this context, the present study expands the knowledge of the parasite fauna of endemic fishes in the upper São Francisco region (BRASIL-SATO, 2002; BRASIL-SATO; SANTOS, 2003; SANTOS; BRASIL-SATO, 2004). In this study, we also evaluate the parasite community component in relation to the size and sex of the hosts, to assist better understanding of the biology of *C. conirostris*.

MATERIAL AND METHODS

Twenty-four specimens of *C. conirostris* were collected from the upper São Francisco, downstream from the Três Marias Dam (18º12'32''S, 45º15'41''W), in the municipality of the same name, by fisherman from the Companhia de Desenvolvimento dos Vales do São Francisco e do Parnaíba (CODEVASF), using nets and hooks. Of this total, 16 were captured between September and December 1998 and seven between October 2003 and March 2004.

Of the 24 pirá specimens collected, eight were male, with a total length of 51.8±3.5cm (47.0-56.5cm) and body weight of 1193.4±260.5g (828-1530g), and 16 were female, with length of 54.6±12.1cm (37.0-78.0cm) and weight of 1571.7±1015.6g (346-3860g).

The parasites encountered were collected, fixed, stained and mounted on slides according to Amato et al. (1991) and Eiras et al. (2000).

The statistical analyses applied to the parasite infrapopulations followed Zar (1996). The data normality was previously verified. We used the chi-square test (c^2) with a 2x2 contingency table to evaluate the parasite prevalence in relation to the hosts' sex, and Student's *t* test to analyze the possible difference in size and weight between the male and female hosts and parasite abundance in relation to host sex. We did not evaluate the parasite intensity due to the reduced number of individuals (24 fish). We employed Spearman's rank correlation coefficient (r_s) to detect possible correlations between the parasite abundance and total host length. Finally, we calculated the dominance and respective frequency, shared dominance and mean relative dominance of the parasite species according to Rohde et al. (1995). The ecological terminology in the parasitological data followed Bush et al. (1997) and we only carried out statistical analysis for the parasite species with prevalence above 10% (BUSH et al., 1990). All the statistical tests were at the $P<0.05$ level of significance. We deposited voucher specimens of the parasites in the Coleção Helmintológica do Instituto Oswaldo Cruz (CHIOC), Rio de Janeiro, Brazil, and deposited voucher specimens of *C. conirostris* in the Coleção de Peixes do Museu de Zoologia da Universidade de São Paulo, São Paulo, Brazil (MZUSP 87223).

RESULTS

Of the 24 pirás collected, 12 (50%) were parasitized by at least one species of parasite. Five (20.8%) were parasitized by one species, five (20.8%) with two species, one (4.1%) with three and one (4.1%) with four species. Nine species of parasites were encountered: one of Hirudinea (*Helobdella* sp., nine specimens, CHIOC 35410, 36580); four of Digenea (*Creptotrema creptotrema* Travassos, Artigas & Pereira, 1928, 116 specimens, CHIOC 36573a-b, 35581, *Palaeocryptogonimus claviformis* Szidat, 1954, eight specimens, metacercariae of *Clinostomum* sp., three specimens, CHIOC 36574 and metacercariae of *Austrodiplostomum compactum* (Lutz, 1928), 15 specimens, CHIOC 36571); three of Nematoda (*Procamallanus (Spirocamallanus)* sp., one young specimen, CHIOC 36572, larvae of Anisakidae, eight specimens, CHIOC 36579a-b and larvae of *Rhabdochona* sp., two specimens, CHIOC 36582) and one of Acanthocephala (*Neoechinorhynchus* sp., one young specimen, CHIOC 36575).

The prevalence was higher for *Helobdella* sp., followed by *C. creptotrema* and *P. claviformis*, with *C. creptotrema* most abundant (Table 1). *Creptotrema creptotrema* was the dominant species, followed by *Helobdella* sp. and *P. claviformis* in the parasite community of *C. conirostris* (Table 2).

The total length and body weight of the hosts did not vary significantly between males and females (t=0.64, p=0.53; t=1.03, p=0.31, respectively), having been analyzed together.

The prevalence and abundance of *Helobdella* sp. and *C. creptotrema* did not differ significantly between male and

Table 1. Prevalence (P) expressed as a percentage, amplitude of intensity (AI), mean intensity (MI), mean abundance (MA) and place of infection/infestation (PI) of the metazoan parasites of *Conorhynchos conirostris* (Valenciennes, 1840) of the São Francisco River, Três Marias, Minas Gerais, Brazil.

Parasite	P (%)	AI	MI	MA	PI
Hirudinea					
Helobdella sp.	20.8	1-3	1.8±1.09	0.37±0.05	Gills and mouth
Digenea					
Creptotrema creptotrema	16.7	7-44	29.0±15.7	4.83±0.66	Anterior and middle intestine
Paleocryptogonimus claviformis	16.7	1-3	2.0±1.15	0.33±0.04	Middle and posterior intestine
Clinostomum sp. (metacercariae)	8.3	1-2	1.5±0.71	0.12±0.03	Gills
Austrodiplostomum sp. (metacercariae)	8.3	1-14	7.5±9.19	0.62±0.38	Gills and eyes
Nematoda					
Anisakidae (larvae)	8.3	1-7	4.0±4.24	0.33±0.06	Mesentery
Procamallanus (Spirocamallanus) sp. (young)	4.2	-	1.0	0.04±0.20	Anterior ntestine
Rhabdochona sp. (larvae)	4.2	-	2.0	0.08±0.02	Mesentery
Acanthocephala					
Neoechinorhynchus sp. (young)	4.2	-	1.0	0.04±0.008	Anterior intestine

Table 2. Dominance (Frequency of dominance), shared dominance and mean relative dominance of the metazoan parasites of *Conorhynchos conirostris* (Valenciennes, 1840) of the São Francisco River, Três Marias, Minas Gerais, Brazil.

Parasite	Dominance (%)	Shared dominance	Relative dominance mean ± standard deviation
Helobdella sp.	2 (8.3)	1	0.15±0.34
Creptotrema creptotrema	4 (16.7)	0	0.16±0.37
Paleocryptogonimus claviformis	2 (8.3)	1	0.10±0.29

Table 3. Analysis of the influence of the sex of *Conorhynchos conirostris* (Valenciennes, 1840) of the São Francisco River, Três Marias, Minas Gerais, Brazil, on the parasite prevalence (χ^2) and abundance (*t*) and the total length of these hosts on the parasite abundance (r_s).

Parasite	Statistical Test*					
	χ^2	*p*	*t*	*p*	r_s	*p*
Helobdella sp.	0.03	0.84	1.30	0.20	-0.10	0.61
Creptotrema creptotrema	0.15	0.69	0.29	0.77	-0.37	0.07
Paleocryptogonimus claviformis	-	-	-	-	-0.06	0.77

*Chi-square: χ^2; Student's t: *t*; Spearman's rank correlation coefficient: r_s; level of significance: $p<0.05$

female hosts. There was no correlation between the abundance of these parasites and the total length of their hosts. Regarding *P. claviformis*, all were found to be infecting male hosts and there was no significant correlation of the parasite abundance and the size of these fish (Table 3).

DISCUSSION

From the results obtained, *C. conirostris* is part of the intermediate host community, in which metacercariae and larvae of Nematoda are distributed. Various fish species are intermediate hosts of the metacercariae of *Diplostomum (A.) compactum* (used in this article as *Austrodiplostomum compactum* according to Niewiadomska, 2002) and of *Clinostomum* sp. in South American limnic systems, especially in Brazil (THATCHER, 1993, KOHN et al., 1995, PAVANELLI et al., 1998, AMATO et al. 2001, MARTINS et al., 2002, BRASIL-SATO, 2003 and BRASIL-SATO; PAVANELLI, 2004). In the upper São Francisco, *C. conirostris* expands the list of hosts. In the São Francisco Basin, the prevalence of *Austrodiplostomum compactum* was comparatively higher in *Pimelodus maculatus* La Cepède, 1803 than in other fish species, such as *C. conirostris*. The indication by Brasil-Sato and Pavanelli (2004) that *P. maculatus* plays an important role in transmitting these larvae to the definitive hosts (birds),

among other fishes of the São Francisco River, gains strength. In counterpart, the prevalence of *Helobdella* sp. was the highest of the parasite community of *C. conirostris*, a result indirectly related to its malacophage habit. It is probable that the pirás facilitate infestation of leeches through contact with the sediment while feeding on snails, since according to Sidall and Burreson (1995), these leeches feed on the hemolymphs of Oligochaetes and winkles. Hirudinea also have been found in *Franciscodoras marmoratus* (Reinhardt, 1874), endemic to the São Francisco River, with comparatively low prevalence and intensity of infestation (SANTOS; BRASIL-SATO, 2004) in relation to the levels found in the present study.

Creptotrema creptotrema presented considerable prevalence and the mean intensity and abundance values were higher in *C. conirostris*, while in *P. maculatus* of the São Francisco, these indices were lower among endoparasites (BRASIL-SATO; PAVANELLI, 2004). Once again, the presence of mollusks in the diet may have favored the establishment of these Digenea in *C. conirostris*. Since the prevalence and abundance of *C. creptotrema* did not differ between male and female hosts, and the abundance was not correlated with total length, this suggests that sex and size do not influence the feeding habits of *C. conirostris*. The parasite fauna of fishes of the upper São Francisco under study presents some evidence on the relationship between diet and types of parasite communities. If a diet of mollusks structures a parasite community based on species of Digenea (with mature specimens) such as in *C. conirostris*, a diet including a more diverse spectrum of items favors the establishment of a parasite community made up of various types, such as cestodes, nematodes and acanthocephalans, which normally use arthropods (adult insects and insect larvae, copepods, etc.) and other invertebrates as intermediate hosts. *Pimelodus maculatus* has been found to be a definitive host of a heterogeneous parasite community, whose species have considerable indices (BRASIL-SATO; PAVANELLI, 1998, 1999, BRASIL-SATO, 2003). *Franciscodoras marmoratus*, an endemic omnivore has a predilection for arthropods and its endoparasitic fauna basically consists of cestodes and acanthocephalans, with no species of Digenea (SANTOS; BRASIL-SATO, 2004). Obviously, a fish species' food source is a noteworthy aspect, and although perhaps not the only or most important one, it definitely has an effect on the structure and regulation of these parasite communities. This is exactly why parasitological studies need to be encouraged, since they demonstrate the intricate relationships among organisms in systems and the need to conserve biodiversity.

In *C. conirostris* there was similar parasitism of *C. creptotrema* and *P. claviformis*. It is probable that the intermediate hosts of *P. claviformis* are mollusks or associated invertebrates that share the same type of environment as *C. creptotrema*. On the other hand, *P. claviformis* was found only in male hosts, without significant correlation between its abundance and fish size. Although this occurrence may have been purely accidental, one cannot discard the possibility of selective feeding habits between the sexes. Unfortunately, the parasite fauna of aquatic vertebrates and invertebrates and those infecting non-aquatic animals in nearby areas of the upper São Francisco – as in the majority of Brazilian continental aquatic systems – is still not sufficiently known to understand the main parasite cycles and structure of parasite transmission.

Pinto et al. (1974) collected *Procamallanus* (*Spirocamallanus*) sp. from the stomach of *Conostome conirostris* (Val.) (=*Conorhynchos conirostris* according to Reis et al., 2003) from the São Francisco River in the municipality of Pirapora, Minas Gerais, constituting the only record of parasitism in this fish. After observing the specimen deposited by Pinto et al. (1974) (CHIOC 28629), we found it impossible to visualize the diagnostic characteristics or decide on its developmental stage. The occurrence of these nematodes in *C. conirostris* in the São Francisco Basin is ratified, although in the present study the only specimen (still young) found leaves doubt on the use of this fish as a definitive host. In the upper São Francisco, *Procamallanus (Spirocamalllanus)* spp. has been found in various fishes: Characidae, Pimelodidae and mainly among the Anastomidae (BRASIL-SATO, 2003). This is also the case for the water systems related to the Rio de la Plata Basin (KOHN; FERNANDES, 1987, PAVANELLI et al., 1997). Although the occurrence of young nematodes and acanthocephalans has been inexpressive and apparently accidental, the infection must have occurred by ingestion of arthropods by the pirás.

The parasitism by larval nematodes indicates that *C. conirostris* is a fish at the intermediate level of the food chain. The larvae of Anisakidae, widely distributed in continental fishes such as *C. conirostris*, guarantee transmission to their definitive hosts. These larvae have similar morphology to that found by Moreira (1994) in various fish species in the Três Marias reservoir, in the São Francisco Basin, identified as *Heterotyphlum* sp., and by Martins et al. (2000) in *Plagioscion squamosissimus* (Heckel 1840) from the Volta Grande reservoir, in the Rio Grande Basin in Minas Gerais, identified as *Thynnascaris* sp. The larvae present a excretory pore adjacent to the nerve ring, long ventricular appendix and relatively undeveloped intestinal caecum. According to Hartwich (1975), the presence of interlabia in adult nematodes defines the species in *Thynnascaris* Dolffus, 1933 (a junior synonym of *Hysterothylacium* (Rudolphi, 1802) according to Deardorff; Overstreet (1980) and currently classified by some researchers in Rhaphidascarididae *sensu* Fagerholm (1991)) and separates them from the species allocated in *Heterotyphlum* Spaul, 1927 (included in the group *Contracaecum* Railliet & Henry, 1912, *sensu lato*, according to Myers (1975) and classified in Anisakidae). *Hysterotylacium* sp. has been found in various fishes of the Paraná River, including in *P. squamosissimus* (MORAVEC et al., 1993). According to Anderson (2000), the larvae in the third stage (L3) have rudimentary labia but not interlabia and a tooth-like structure ventro-lateral to the oral opening, present in the labium. In fact, Moreira (1994) did not

see interlabia at the cephalic extremity of the larvae under scanning electron microscopy. Considering the proposal of Deardorff and Overstreet (1980) and the characteristics of the larvae found by Anderson (2000), also present in the larvae encountered by Moreira (1994), Martins et al. (2000) and in the present study, we suggest that they constitute the third stage of *Hysterothylacium* sp., but we prefer to maintain the identification of the larvae found in *C. conirostris* as larvae of Anisakidae. The prevalence and mean intensity, respectively, of the larvae found by Moreira (1994) were 2.3% and 1.0 in *P. maculatus*, 6.9% and 3.5 in *Tetragonopterus chalceus* Spix & Agassiz, 1829, 9.5% and 8.0 in *Pachyurus squamipennis* Agassiz, 1831, 15.8% and 1.7 in *Serrasalmus brandti* (Lütken, 1875), 37.6% and 3.3 in *Hoplias lacerdae* Miranda Ribeiro, 1908, 42.5% and 7.6 in *Acestrorhynchus lacustris* (Lütken, 1875), 51.1% and 3.2 in *A. britskii* Menezes, 1969 and 100% and 1.5 in *Salminus hilarii* Valenciennes 1850. Martins et al. (2000) recorded 44.1% and 1.7 (mean calculated by us with data from Table 1, p. 522) in *P. squamosissimus*. Comparing the parasite indices between the fish species allocated in Sciaenidae from the two locations, it can be observed that they were higher in the Volta Grande than in the Três Marias reservoir. The results obtained by Moreira (1994) show that the larvae use various fishes for successful transmission to their definitive hosts. These results also showed that in some fish species (Characidae) the prevalence of larvae was higher than that recorded by Martins et al. (2000) and that the mean intensity was higher in *P. squamosissimus* than in the fishes from the Três Marias reservoir. In *C. conirostris* the prevalence and mean intensity of the larvae is within the amplitude of variation of the indices recorded by Moreira (1994) and lower than those obtained by Martins et al. (2000). This variation may be due to the host types and also the place studied in the limnic system. In Characidae, the indices in general were higher than in the other species from different families (MOREIRA.1994). The fish examined in this study and those by Moreira (1994) were collected near the Três Marias dam (downstream and upstream, respectively). The higher prevalence values in the fish from the reservoirs indicate the possibility that this type of environment contributes more to the superdispersion of these larvae in the intermediate host community, and consequently in the transmission to the definitive hosts, supposedly regulated by abiotic characteristics of the season of the year they were collected. Martins et al. (2000) found a positive correlation between the prevalence of larvae and increased air temperature and rainfall in the region of the reservoir.

Because it *C. conirostris* is an endemic species that is in danger of extinction, the present parasite assay could not be more extensive. Nevertheless, through this sample, we have shown that *C. conirostris* is a host to some species of parasites, principally adult specimens of Digenea, probably because of the differentiated use of mollusks among the invertebrates. As an endangered species, in studies where it is necessary to stock these fish in tanks, in which the installation of invertebrate fauna is common, prophylactic handling measures should be considered in to avoid parasites such as Digenea and Hirudinea. Eradicating the mollusks that naturally colonize this environment could be an adequate measure, if none of them are part of the fish's diet. An evaluation of the real harm to the fish's health and to the objectives of the study or the degree of threat these parasites can cause to fish under cultivation would be necessary. Allowing the interactions to occur and conducting periodic examinations of the fish with careful handling would be laborious, but probably a conscientious decision.

In the present study, we increased to 22 the number of metazoan parasites collected from four species of fish endemic to the upper São Francisco River, but we did not study the ectoparasites of two of these hosts. The prevalence and abundance of parasites were not influenced by the host sex and the parasite abundance was not significantly correlated with the size of the hosts. With the exception of *Procamallanus (Spirocamallanus)* sp., the parasites have been registered for the first time in *C. conirostris*. *Helobdella* sp., *P. claviformis* and *Rhabdochona* sp. (larvae) constitute new occurrences in fishes of the upper São Francisco, an expansion of their known geographic distribution.

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