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Infection by *Austrodiplostomum compactum* metacercariae in fish from the Nova Avanhandava reservoir, Tietê river, São Paulo State, Brazil

Jaciara Vanessa Krüger Paes*, Edmir Daniel Carvalho and Reinaldo José Silva

Instituto de Biociências, Universidade Estadual Paulista "Julio de Mesquita Filho", Distrito de Rubião Júnior, s/n, 18618-970, Botucatu, São Paulo, Brazil. *Author for correspondence. Email: jvkpaes@gmail.com

ABSTRACT. This study aimed to evaluate the infection by the *Austrodiplostomum compactum* metacercariae in fishes from the Nova Avanhandava Reservoir, low Tietê river, São Paulo State, Brazil. The parasites were collected from eye (aqueous and vitreous humor), fixed in AFA solution and stained with carmine. The morphometric analysis was performed using a computerized system for analysis of images QWin Lite 2.5 (Leica). Prevalence, mean intensity of infection and abundance of infected fish were calculated. Of the 22 species of fish registered, five were infected by metacercariae: *Hoplias malabaricus*, *Metynnis maculatus*, *Plagioscion squamosissimus*, *Satanoperca pappaterra* and *Schizodon nasutus*. Of the 627 fish evaluated, 34% were infected. A higher prevalence was observed in *P. squamosissimus* and *S. pappaterra*. *Schizodon nasutus* and *M. maculatus* are new hosts reported for *A. compactum* metacercariae.

Key words: Diplostomidae, trematode, new host, eye, parasite.

RESUMO. Infecção por metacercárias de *Austrodiplostomum compactum* em peixes do Reservatório de Nova Avanhandava, rio Tietê, Estado de São Paulo, Brasil. Avaliou-se a infecção por metacercárias de *Austrodiplostomum compactum* em peixes do Reservatório de Nova Avanhandava, baixo rio Tietê, São Paulo, Brasil. Os parasitos foram coletados nos olhos (humor aquoso e vítreo), fixados com solução de AFA e corados com carmin. A análise morfométrica foi realizada por meio de sistema computadorizado de análise de imagem QWin Lite 2.5 (Leica). A prevalência, intensidade média de infecção e a abundância média dos peixes infectados foram calculadas. Das 22 espécies de peixes capturadas, cinco estavam infectadas por metacercárias: *Hoplias malabaricus*, *Metynnis maculatus*, *Plagioscion squamosissimus*, *Satanoperca pappaterra* e *Schizodon nasutus*. De um total de 627 peixes avaliados, 34% estavam infectados. Alta prevalência foi observada em *P. squamosissimus* e *S. pappaterra*. *Schizodon nasutus* e *M. maculatus* são novos hospedeiros registrados para metacercárias de *A. compactum*.

Palavras-chave: Diplostomidae, trematódeo, novos hospedeiros, olho, parasito.

Introduction

Digenetic helminthes of the family Diplostomidae are responsible for 'wormy cataract', also called diplostomiasis (MARTINS et al., 1999). The genus *Austrodiplostomum* Szidat and Nani, 1951 have been reported infecting several species of vertebrates (KOHN et al., 1995; RAMOS-RAMOS, 1995; YAMAGUTI, 1971; SCHOLTZ et al., 1995; NIEWIADOMSKA, 1996; NIEWIADOMSKA; LASKOWSKI, 2002). These parasites, when adults, are able to infect birds and mammals, and the metacercariae stage are found infecting fish and amphibians, which are intermediate hosts in their life cycle (AMATO et al., 2001; FLOWERS et al., 2004; OSTROWSKI-NÚÑEZ, 1982; RIETSCHER; WERDING, 1978). According to Eiras (1994), the

presence of this parasite in the eyes can cause blindness or reduced vision, making the fish susceptible to the predation that facilitates the transmission of the parasite to the definitive host.

Austrodiplostomum compactum (Lutz, 1928) metacercariae occur in the eyes of a wide variety of fish species (YAMADA et al., 2008). In Brazil, metacercariae infection by *A. compactum* has been registered in fish species in different hydrographic basins: *Plagioscion squamosissimus* (Heckel, 1840), *Hoplias* aff. *malabaricus* (Bloch, 1794), *Cichla monoculus* Spix, 1831 and *Cichla ocellaris* Bloch and Schneider, 1801, *Satanoperca pappaterra* (Heckel, 1840), *Crenicichla britskii* Kullander, 1982, *Cichlasoma paranaense* Kullander, 1983, *Geophagus brasiliensis* Quoy and Gaimard, 1824, *Serrasalmus maculatus* (Kner, 1858), *Hypostomus regani* (Thering, 1905), *Schizodon borellii* (Boulenger, 1900) and

Auchenipterus osteomystax Miranda-Ribeiro, 1918 (KOHN et al., 1995; MACHADO et al., 2005; MARTINS et al., 1999; 2002; NOVAES et al., 2006; PAES et al., 2003; PAVANELLI et al., 1997; SANTOS et al., 2002; YAMADA et al., 2007; YAMADA et al., 2008; ZICA et al., 2009).

Since the occurrence of *A. compactum* metacercariae was previously reported in the Tietê river (NOVAES et al., 2006; PAES et al., 2003) and considering the diversity of fish in the Tietê basin (VIDOTTO; CARVALHO, 2007) and the lack of parasitological studies in this environment highly influenced by human actions, the aim of this study was to evaluate the occurrence of these metacercariae in fish from the Santa Bárbara river in the Nova Avanhandava reservoir, low Tietê river, São Paulo State, Brazil.

Material and methods

The study was conducted in the Santa Bárbara river, municipality of Buritama, São Paulo State, Brazil, a major tributary of the Nova Avanhandava reservoir, the penultimate hydroelectric plant of the mid Tietê river, whose dam is located at the geographic coordinates 21°07'S and 50°17'W (Figure 1).

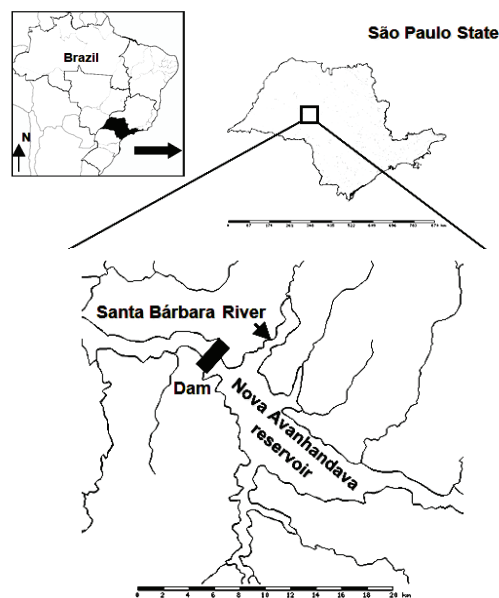


Figure 1. Map of Brazil highlighting São Paulo State and the place of study at the Santa Bárbara river, in the Nova Avanhandava reservoir, low Tietê river, municipality of Buritama, Brazil.

Fish were collected for this study on July 31, September 9, October 15 and December 18, 2003 using a gillnet of nylon monofilament, installed in several parts of the study area. Fish were identified and

conferred on the basis of Reis et al. (2003) and Graça and Pavanelli (2007). For the determination of the hosts for *A. compactum*, each sampled fish was macroscopically examined for detection of the metacercariae in the ocular globe (aqueous and vitreous humor). When a parasite was observed in a fish species, the eyes of all specimens of this species were analyzed in a stereomicroscope.

The parasites collected were processed according to Amato et al. (1991). The morphometric analysis was performed using a computerized system for analysis of images QWin Lite 2.5 (Leica) in a sample of 21 metacercariae from *P. squamosissimus*. The morphometric results (in micrometers) are presented as minimum-maximum (mean) values.

The identification of the metacercariae was according to Niewiadomska (2002a; 2002b), Kohn et al. (1995), Santos et al. (2002), Novaes et al. (2006), and Zica et al. (2009). Voucher specimens were deposited in the Coleção Helmintológica of the Departamento de Parasitologia, Instituto de Biociências, Universidade Estadual Paulista, Botucatu, São Paulo State, Brazil.

Prevalence, mean intensity of infection and abundance of infected fish was calculated according to Bush et al. (1997).

Results

In this study, 22 species of fish were recorded in the Nova Avanhandava reservoir: *Acestrorhynchus lacustris* (Lütken, 1875) (n = 1), *Apareiodon affinis* (Steindachner, 1879) (n = 19), *Astyanax altiparanae* Garutti and Britski, 2000 (n = 356), *Astyanax fasciatus* (Cuvier, 1819) (n = 1), *Brycon orbignyanus* (Valenciennes, 1850) (n = 2), *Cichla kelberi* (Kullander and Ferreira, 2006) (n = 7), *Cichla piquiti* (Kullander and Ferreira, 2006) (n = 3), *Crenicichla britskii* (n = 2), *Hoplias malabaricus* (n = 7), *Hoplosternum littorale* (Hancock, 1828) (n = 1), *Hypostomus iheringii* (Regan, 1908) (n = 3), *Leporinus friderici* (Bloch, 1794) (n = 5), *Metynnis maculatus* (Kner, 1858) (n = 298), *Moenkhausia intermedia* Eigenmann, 1908 (n = 14), *Plagioscion squamosissimus* (n = 213), *Prochilodus lineatus* (Valenciennes, 1836) (n = 1), *Rhamdia quelen* (Quoy and Gaimard, 1824) (n = 8), *Satanoperca pappaterra* (Heckel, 1840) (n = 25), *Schizodon nasutus* Kner, 1859 (n = 84), *Serrasalmus maculatus* (n = 68), *Steindachnerina insculpta* (Fernández-Yépez, 1948) (n = 18) and *Triportheus nematurus* (Kner, 1858) (n = 5).

The presence of metacercariae of *A. compactum* was confirmed in five fish species in the Santa Bárbara river: *P. squamosissimus*, *S. nasutus*, *S. pappaterra*, *H. malabaricus*, and *M. maculatus*. *Plagioscion squamosissimus* (90%) and *S. pappaterra* (60%) presented

the highest prevalence, mean intensity of infection and mean abundance (Table 1).

Table 1. Parasitism by *Austrodiplostomum compactum* in the eyes of fish from the Nova Avanhandava reservoir, low Tietê river, municipality of Buritama, São Paulo State, Brazil.

Fish species	N	P (%)	MII	MA
<i>Plagioscion squamosissimus</i> ^a	213	90.1	20.8	18.718
<i>Schizodon nasutus</i>	84	4.8	1.2	0.059
<i>Satanoperca pappaterra</i> ^a	25	60	6.4	3.840
<i>Hoplias malabaricus</i>	7	14.3	9	1.286
<i>Metynnis maculatus</i> ^a	298	0.33	1	0.003

N - number of collected fish; P - prevalence; MII - mean intensity of infection; MA - mean abundance; ^a introduced species (AGOSTINHO et al., 2007; LANGEANI et al., 2007).

The metacercariae (Figure 2, Table 2) of *A. compactum* presented a foliaceous body, slightly concave in the ventral face and with a small conical segment in the posterior region; a small subterminal oral sucker, two lateral pseudosuckers in the anterior region; oval pharynx, short esophagus, intestinal caeca ending near the posterior region; oval holdfast (tribocytic organ). Gland cells occupying most of anterior region, extending from the beginning of intestinal caeca to the anterior region of the tribocytic organ and small gonads in the posterior region, after the tribocytic organ, were observed.

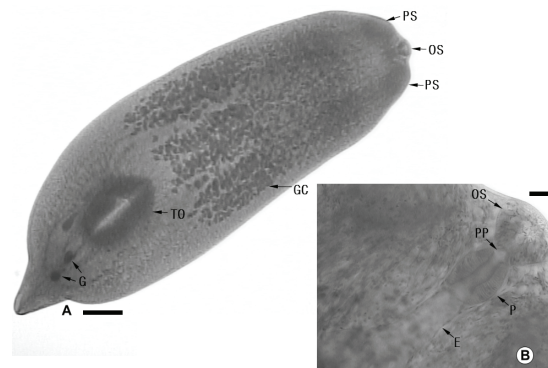


Figure 2. *Austrodiplostomum compactum* (Diplostomidae) metacercariae found in the eyes of *Plagioscion squamosissimus* (Scianidae). A: overview metacercariae, ventral view (Bar = 200 µm), with oral suckers (OS), pseudosuckers (PS), gland cells (GC), tribocytic organ (TO) and gonads (G); B: detail of anterior region of the body, ventral view (Bar = 30 µm), showing oral suckers (OS), prepharynx (PP), pharynx (P) and esophagus (E).

Discussion

Among the species found infected by *A. compactum* in the present study, *P. squamosissimus*, *S. pappaterra* and *H. malabaricus* had previously been recorded as hosts for this digenetic metacercariae (PAVANELLI et al., 2000; MACHADO et al., 2005; PAVANELLI et al., 2006). However, *S. nasutus* and *M. maculatus* are new hosts recorded for *A. compactum* metacercariae.

The morphological and morphometrical data of *A. compactum* metacercariae from Nova Avanhandava reservoir are in accordance with other morphological studies (Table 2) for this species (KOHN et al., 1995; SANTOS et al., 2002; NOVAES et al., 2006; ZICA et al., 2009).

It is emphasized that, in *P. squamosissimus*, parasitic levels were considerably higher. This species, from the Amazon Basin, was introduced by the Energy Company of São Paulo (CESP), in reservoirs under its responsibility from 1966 to 1973 (TORLONI et al., 1993). According to Pojmanska and Chabros (1993), the prevalence of diplostomid in introduced fish is higher when compared with that of indigenous fish. This finding was also observed by Machado et al. (2005), who found higher rates of infection in fish introduced into the floodplain of the upper Paraná river, and *P. squamosissimus* species with the highest prevalence and intensity of infection. These authors suggest that probably *A. compactum* was introduced along with *P. squamosissimus*, which justified its higher prevalence and intensity of infection. However, native species may have been acting as intermediate hosts, to which the adaptation of metacercariae of *A. compactum* would thus justify the low prevalence rates and intensity of infection in these fish.

The introductions, depending on the fish species, can promote reductions of the native stocks or local extinctions, as result of several factors including dissemination of pathogens and parasites (AGOSTINHO; JÚLIO JUNIOR, 1996). The infection with *A. compactum* metacercariae in several fish species shows that the low specificity of the parasite favors its spread, infecting other species of native fish, and the parasite appears to have found appropriate ecological conditions for its parasite cycle (snails, fish and birds) in many aquatic ecosystems (MACHADO et al., 2005; RUIZ; AGUILAR, 2005).

In this reservoir, the elements necessary for the biological cycle of the parasite are present. Studies have demonstrated the occurrence of gastropod mollusks *Biomphalaria* sp. Preston, 1910 (FRANÇA et al., 2007) and also birds of the genus *Casmerodius* Gloger, 1842 and *Phalacrocorax* Brisson, 1760. In this environment, *H. malabaricus*, *M. maculatus*, *P. squamosissimus*, *S. pappaterra* and *S. nasutus* serve as an intermediate host for *A. compactum*. However, we postulate that this helminth adapted better to *P. squamosissimus* and *S. pappaterra*, and that *S. nasutus* and *M. maculatus* are sporadic secondary intermediate hosts or even an accidental infection case, also suggesting a low specificity of this parasite.

Table 2. Morphometrical data of *Austrodiplostomum compactum* (Diplostomidae) metacercariae.

Host	Kohn et al. (1995)	Santos et al. (2002)	Novas et al. (2006)	Zica et al. (2009)	Present study	Total variation for the species
	<i>Plagioscion squamosissimus</i>	<i>Plagioscion squamosissimus</i>	<i>Cichla ocellaris</i>	<i>Geophagus brasiliensis</i>	<i>Hypostomus regani</i>	<i>Plagioscion squamosissimus</i>
Number of specimens	14	20	20	5	10	21
Length	1470-2740 (2170)	880-1840 (1434)	960-2480 (1462)	1584-1947 (1800)	1570-2281 (1988)	1301-2386 (1911)
Width	600-1180 (970)	400-792 (611)	560-960 (711)	537-706 (642)	543-864 (756)	482-854 (678)
Length of conical segment	41-97 (77)	-	-	-	-	-
Oral sucker						
Length	41-97 (77)	44-90 (65)	40-98 (68)	45-83 (59)	69-102(91)	51-87 (71)
Width	56-116 (79)	40-64 (52)	30-98 (56)	54-77 (68)	75-99 (84)	51-92 (73)
Lateral pseudosucker (1)						
Length	-	-	-	-	93-148 (132)	-
Width	-	-	-	-	68-157 (118)	-
Lateral pseudosucker (2)						
Length	-	-	-	-	78-168 (131)	-
Width	-	-	-	-	85-146 (119)	-
Prepharynx						
Length	-	2-10 (6)	4-20 (9)	-	-	-
Width	-	10-24 (17)	16-38 (23)	-	-	-
Pharynx						
Length	64-94 (83)	50-64 (62)	44-98 (68)	53-73 (61)	57-85 (73)	49-84 (69)
Width	45-79 (60)	40-60 (49)	38-78 (52)	50-64 (56)	57-80 (64)	45-74 (58)
Esophagus						
Length	-	-	-	-	86-139 (111)	54-101 (72)
Width	-	-	-	-	-	29-53 (39)
Tribocytic organ						
Length	326-650 (507)	200-600 (285)	200-496 (308)	422-434 (428)	287-414 (373)	205-554 (401)
Width	251-500 (370)	160-232 (182)	120-320 (180)	220-319 (258)	178-310 (243)	127-347 (246)

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