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Parasitic infections of *Piaractus mesopotamicus* and hybrid (*P. mesopotamicus* x *Piaractus brachypomus*) cultured in Brazil

Infecções parasitárias de *Piaractus mesopotamicus* e do híbrido (*P. mesopotamicus* x *Piaractus brachypomus*) cultivados no Brasil

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Abstract

This study evaluated the occurrence of parasitic infections in the “pacu” fish *Piaractus mesopotamicus* and the “patinga” hybrid (*P. mesopotamicus* x *Piaractus brachypomus*) in the northwest of São Paulo State, Brazil. Fish from the following three fish farms were evaluated every two months: A, a hatchery and larviculture farm (n = 16 pacu / n = 19 patinga), B, a growout farm (n = 35 patinga) and C, a fee-fishing property (n = 28 pacu / n = 7 patinga). Thirty-five fish from each property were collected from February 2010 to February 2011 and subjected to parasitological analysis. The parasites found were the following: *Mymarothecium viatorum*, *Anacanthorus penilabiatus*, *Notozothecium janauachensis* (Dactylogyridae, Monogenea), *Trichodina* spp., *Ichthyophthirius multifiliis*, *Chilodonella* sp. (Protozoa), *Myxobolus* spp., *Henneguya* spp. (Myxozoa), *Rondonia rondoni*, *Contracaecum* sp. (Nematoda), and *Dolops carvalhoi* (Crustacea). Of the fish examined, 62.9% from “A” and 100% from “B” and “C” were infested with at least one parasite species. Pacu fish (n = 44) showed a higher susceptibility to *Anacanthorus penilabiatus* infestations, whereas patinga (n = 61) were more susceptible to *Mymarothecium viatorum* ($p < 0.05$). Appropriate fish handling (nutrition, transport and storage), in conjunction with monitoring of water quality, can reduce the stress to which the farmed fish are exposed and is essential for pathogen control.

Keywords: Handling, helminths, protozoans, storage control, water quality.

Resumo

Este estudo avaliou a ocorrência de infecções parasitárias em pacus *Piaractus mesopotamicus* e do híbrido patinga (*P. mesopotamicus* x *P. brachypomus*) procedentes da região Noroeste do Estado de São Paulo, Brasil. Peixes de três pisciculturas foram avaliados bimestralmente: A - Reprodução e Larvicultura (n = 16 pacus / n = 19 patingas), B- Engorda n = 35 patingas), e C- Pesque-pague (n = 28 pacus / n = 7 patingas). Trinta e cinco peixes de cada propriedade foram coletados de fevereiro de 2010 a fevereiro de 2011, para análises parasitológicas. Os parasitas encontrados foram: *Mymarothecium viatorum*, *Anacanthorus penilabiatus*, *Notozothecium janauachensis* (Dactylogyridae, Monogenea), *Trichodina* spp., *Ichthyophthirius multifiliis*, *Chilodonella* sp. (Protozoa), *Myxobolus* spp., *Henneguya* spp. (Myxozoa), *Rondonia rondoni*, *Contracaecum* sp. (Nematoda), and *Dolops carvalhoi* (Crustacea). Dentre os peixes analisados, 62,9% de “A” e 100% de “B” e “C” estavam infectados/infestados por pelo menos uma espécie de parasita. Pacus (n = 44) apresentaram maior suscetibilidade a infestações por *Anacanthorus penilabiatus*, e patingas (n = 61), por *Mymarothecium viatorum* ($p < 0.05$). A profilaxia e cuidados durante o manejo (alimentação, transporte e estocagem), associados ao monitoramento da qualidade de água reduzem o estresse o qual os peixes cultivados estão submetidos, sendo medidas imprescindíveis para o controle de patógenos.

Palavras-chave: Manejo, helmintos, protozoários, controle de estocagem, qualidade de água.

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Introduction

The significant increase in aquaculture in Brazil and around the world has increased the importance of studies of parasites and other pathogens of aquatic organisms, especially in those hosts that have the potential for production and sale (LUQUE, 2004). The monitoring of parasites and other fish pathogens is an important part of prophylactic management both for production and for public health, especially due to the zoonotic potential of some of these organisms (PAVANELLI et al., 2008).

Since the 1980s, Brazil has made great progress in the research and development of new growing techniques for fish, which have increased the number of species and strains of fish that can be reared in an aquaculture environment. For example, the fishes *Piaractus mesopotamicus*, popularly known as “pacu” (GENOVEZ et al., 2008), and the “patinga” hybrid, which is the result of interspecific hybridisation between a female (F1) of *P. mesopotamicus* and male (M1) of *Piaractus brachipomus*, known as “pirapitinga”, are now both commonly cultured fish in Brazil. Interspecific hybridisation has been used in fish farms to produce animals with better performance than the parental species (hybrid vigour), including improvements in characteristics such as better meat quality, lower feed conversion ratio, rapid growth and resistance to pathogens. However, few studies have been carried out to determine whether hybrids have such desirable characteristics compared to the parental species, which may influence the choice of species in production.

Poor management practices and pathogen infections are important causes of economic losses in aquaculture (PAVANELLI et al., 2008; URBINATI et al., 2010). Parasitic infections are more relevant in the Neotropical region because of the climatic characteristics of this region, which facilitate the rapid and continuous propagation of these pathogens (THATCHER; BRITES-NETO, 1994). Data regarding the epidemiology of pathogens in the production chain of aquaculture fish are scarce, hampering the implementation of cultivation and management techniques that can reduce the need for disease control (OSTRENSKY; BOEGER, 1998).

The aim of the present study was to evaluate the occurrence of parasitic infections in “pacu” fish and the “patinga” hybrid from three fish farms that represent different stages of the production chain in São Paulo State, Brazil.

Materials and Methods

Characterisation of fish farms

To evaluate parasite infection during each phase of pacu and patinga hybrid production, three fish farms were evaluated every two months. One farm was a hatchery, located in the municipality of Estrela d'Oeste (20° 16' 17.26" S 50° 23' 30.24" W), which produces larvae and juveniles of *P. mesopotamicus*, the patinga hybrid and “tambaqui” fish (*Colossoma macropomum*). The hatchery provides juveniles (20-30 g) for growout farms. Fish are stocked at a density of 0.4 kg/m² in ponds with an area of 6,000 m² and

a depth of 2 m with continuous water flow. The second farm was a growout farm that is located in the municipality of Cosmorama (20° 27' 59.70" S 49° 46' 06.65" W) and provides fish (600-800 g) to fee-fishing farms. The patinga hybrid is the major species produced, followed by Nile tilapia (*Oreochromis niloticus*). The pond is 12,500 m² in area and 3 m in depth and is supplied with a water flow rate of 5.10⁻³ m³/s. Fish are stocked at a density of 1.2 kg/m². The fee-fishing farm is located in the municipality of Votuporanga (20° 24' 30.31" S 49° 58' 05.33" W). It contains a pond with different species and their hybrids, including “pacu”, the “patinga” hybrid, “piaçu” fish (*Leporinus macrocephalus*), and Nile tilapia, which are kept at an unknown density with nightly aeration. This pond has an area of 6,000 m² and a depth of 2.8 m, and its water flow rate is 3.10⁻³ m³/s. There is no effective control of the stocking density due to the high marketing of the fish and the absence of harvesting to perform fallowing; however, the estimated density is approximately 1.7 kg/m². The fishes are fed commercial diet once a week, and during the day, fishermen usually offer bait (corn, bran, etc.) to attract fish.

Parasitological procedures

Fish were collected between February 2010 and February 2011 in a total of seven bimonthly samples from each fish farm.

Five fish were collected bimonthly, totalling 105 fish (35 fish/fish farm), and were caught either with nets or with hook and line before being isolated and transported to the laboratory, where they were weighed (g), measured (cm), and analysed to verify the presence of cysts or ectoparasites on the body surface. For qualitative analysis of ciliate protozoa and myxosporidians, fish skin scrapes were performed using a glass slide, and the collected material was immediately examined under a light microscope (PAVANELLI et al., 2008). Imprints and smears of tissues from the kidney, liver and spleen were also made in order to assess the presence of myxosporidian spores.

After the skin scraping of the fish, gills were collected and fixed in alcohol for the later identification and counting of monogenoids. For the study of sclerotised structures of the haptor (anchor, bars, and hooks) and the copulatory complex and vagina, samples were clarified with Hoyer's and Gray and Wess's media (EIRAS et al., 2006). All organs were analysed with a stereoscopic microscope. The collected nematodes were fixed in an alcohol-formaldehyde-acetic acid (AFA) solution, preserved in 70% ethyl alcohol, and were cleared with phenol for identification purposes (EIRAS et al., 2006).

The identification of parasites was based primarily on Thatcher (2006), Moravec (1998), and scientific articles. Morphological and morphometrical analyses were performed using a computerised system for image analysis with differential interference contrast (DIC) - LAS V3 (Leica Application Suite). Voucher helminth species were deposited in the Coleção Helmintológica (CHIBB) of the Departamento de Parasitologia, Instituto de Biociências, Universidade Estadual Paulista (UNESP), municipality of Botucatu, São Paulo State, Brazil.

Water analysis of the fish ponds

Water samples were obtained bimonthly from each pond at the time of host collection. Physical and chemical parameters of the water, such as dissolved oxygen and temperature, were evaluated *in situ* using “YSY - Mod.50” multisensor equipment, and the transparency of the water was measured with a Secchi Disk. The pH, ammonia and nitrite concentrations were obtained using the colorimetric kit AlfaKit® and compared with parameters defined by Ostrensky and Boeger (1998) and Urbinati et al. (2010).

Data analysis

Prevalence (P), mean abundance (MA) and mean intensity of infection (MII) of parasites were calculated according to Bush et al. (1997). Data from MII and MA are presented as the mean \pm standard error followed by the range in parentheses.

The proportion of infected fish was compared to the totality of *P. mesopotamicus* and the hybrid, regardless of the property sampled, using the proportion test (Z-test). The intensity of infection and abundance were compared using the non-parametric Mann-Whitney test (U-test) in accordance with the data distribution (non-normal).

Based on the data for standard length and total weight, the condition factor was calculated according to Le Cren (1951) and was then correlated with the parasite abundance of the fish from the three properties sampled using Spearman's correlation test (r_s) in accordance with the data distribution ($p < 0.05$). Statistical tests were performed using SigmaStat 3.1 (Systat Software Inc., California, USA).

Results

None of the fish collected showed clinical signs of parasitic infections (see PAVANELLI et al., 2008).

In the hatchery, we collected 16 *P. mesopotamicus* and 19 hybrid patinga. In the growout property, all 35 of the collected specimens were hybrids, and in the fee-fishing property, 28 *P. mesopotamicus* and seven hybrids were collected (Table 1).

During the parasitological analysis of fish from these three properties, we found the following parasites: *Myxosporidium viatorum* (CHIBB: 068L, 069L, 070L); *Anacanthorus penilabiatus* (CHIBB: 071L, 072L, 073L); *Notozothecium janauachensis*

(CHIBB: 074L) (Dactylogyridae); third stage larvae (L3) of the anisakid nematode *Contracaecum* sp. (CHIBB 6986) and adults of the nematode *Rondonia rondoni* (CHIBB 6988); the ciliate protozoan *Ichthyophthirius multifiliis*; *Chilodonella* sp.; *Trichodina* spp.; myxosporidians *Henneguya* spp. and *Myxobolus* spp.; and the branchiura crustacean *Dolops carvalhoi* (CHIBB 6987).

Of the fish analysed from the hatchery, 62.9% were parasitised with at least one parasite species. Considering all fish analysed from this property (n=35), the Dactylogyridae monogenoids were the most prevalent parasites (prevalence [P] = 51.4%), and *A. penilabiatus* (P = 64.3%) and *M. viatorum* (P = 35.7%) were the identified species. The prevalence of the other parasites was 37.1% for ciliate protozoans of the genus *Trichodina* (P = 37.1%) and 20.0% and 14.3% for myxosporidians of the genera *Myxobolus* and *Henneguya*, respectively. Only 2.9% of the analysed fish were infected with the protozoan ciliate *I. multifiliis*. The prevalence, mean intensity of infection and mean abundance of parasites observed in pacus and hybrids are shown separately in Table 2.

A higher intensity of infection with *A. penilabiatus* (41.4 \pm 15.6 [0.8-111.6] versus 2.1 \pm 0.6 [0.1-5.9]; $p = 0.03$) and a higher mean abundance of *Trichodina* spp. (133.3 \pm 96.9 [0-1498.0] versus 14.8 \pm 14.4 [0-275.0]; $p = 0.04$) were observed in *P. mesopotamicus* compared to hybrids from the hatchery. There was no significant difference with respect to the levels of parasitism of other parasite species when compared to the rates observed in *P. mesopotamicus* and the hybrid (Table 2).

In the growout system, 100.0% of hybrids were parasitised with at least one parasite species. The Dactylogyridae monogenoids were the most prevalent parasites (P = 100.0%). *M. viatorum* was found at the highest prevalence (P = 100.0%) and was followed by *A. penilabiatus* (P = 88.6%). Furthermore, the hybrid had a higher mean intensity of infection and mean abundance of *M. viatorum* (MII and MA = 261.0 \pm 57.2 [1.0-1331.0]). *Trichodina* spp. also exhibited high prevalence and mean intensity of infection (P = 25.7%; MII = 22.9 \pm 1.0 [1.0-94.0]) (Table 3).

At the fee-fishing property, 100.0% of the collected fish were also parasitised by at least one parasite species. The most prevalent parasites in the total sample (n = 35) were Dactylogyridae monogenoids (P = 97.1%) with a high prevalence of *M. viatorum* (58.0%) and *A. penilabiatus* (41.9%). Other parasites found were *N. janauachensis* (P = 14.3%), the ciliate protozoan *Chilodonella* sp. (P = 14.3%), and the crustacean *D. carvalhoi* (P = 3.6%). The pacu and hybrid results are shown separately in Table 4. The prevalence, mean intensity of infection and mean abundance of

Table 1. Weight (g), standard length (cm) and total length (cm) of fish collected from three fish farms: hatchery and larviculture, growout and fee-fishing farms in the northwest of São Paulo State, Brazil. (Values expressed as the mean \pm standard error).

Properties	Weight	Standard length	Total length
Hatchery and larviculture			
<i>Piaractus mesopotamicus</i> (n=16)	37.1 \pm 12.1 (12.0-139.1)	10.9 \pm 1.0 (7.8-18.5)	11.0 \pm 0.7 (8.1-17.8)
Hybrid (<i>P. mesopotamicus</i> x <i>P. brachypomus</i>) (n=19)	36.7 \pm 9.4 (5.9-160.5)	9.5 \pm 0.7 (6.3-17.2)	11.9 \pm 0.9 (7.5-20.5)
Growout			
Hybrid (<i>P. mesopotamicus</i> x <i>P. brachypomus</i>) (n=35)	1,252.6 \pm 85.7 (418.4-2,950.0)	32.3 \pm 0.7 (22.0-39.5)	37.2 \pm 0.7 (25.3-45.0)
Fee-fishing			
<i>Piaractus mesopotamicus</i> (n=28)	1,188.6 \pm 77.3 (529.4-2,355.0)	32.9 \pm 0.6 (25.8-39.5)	38.3 \pm 0.7 (29.7-48.0)
Hybrid (<i>P. mesopotamicus</i> x <i>P. brachypomus</i>) (n=7)	1,595.5 \pm 131.1 (941.4-2,104.9)	37.2 \pm 1.8 (31.0-45.5)	41.4 \pm 1.8 (36.5-51.0)

Table 2. Mean values ± standard error of the prevalence (P), mean intensity of infection (MII), mean abundance (MA) and sites of infection of the parasites of *Piaractus mesopotamicus* (n=16) and the “patinga” hybrid (*P. mesopotamicus* x *P. brachypomus*) (n=19) from the hatchery and larviculture farm, in the northwest of São Paulo State, Brazil.

Parasite	<i>Piaractus mesopotamicus</i>			Hybrid			Site of infection
	P (%)	MI	MA	P (%)	MI	MA	
Monogenea							Gills
<i>Anacanthorus penilabiatu</i> s	43.7	41.4±15.6* (0.8-111.6)	18.1±8.4 (0-111.6)	47.4	2.1±0.6* (0.1-5.9)	1.0±0.4 (0-5.9)	
<i>Mymarothecium viatorum</i>	31.2	11.2±3.6 (0.1-22.3)	3.5±1.7 (0-22.3)	52.6	11.6±3.6 (0.8-36.0)	6.1±2.3 (0-36.0)	
Protozoa							Gills/Skin(mucus)
<i>Trichodina</i> spp.	56.2	237.0±168.1 (1.0-1498.0)	133.3±96.9* (0-1498.0)	21.0	70.2±68.2 (1.0-275.0)	14.8±14.4* (0-275.0)	
<i>Ichthyophthirius multifiliis</i>	6.2	6.0	0.4±0.4 (0-6.0)	-	-	-	
Myxosporea							Kidney/Spleen/ Skin (mucus) Kidney/Spleen
<i>Henneguya</i> spp.	25.0	-	-	5.2	-	-	
<i>Myxobolus</i> spp.	18.7	-	-	21.0	-	-	

*Statistically significant difference (p < 0.05).

Table 3. Mean values ± standard error of the prevalence (P), mean intensity of infection (MII), mean abundance (MA) and site of infection of the parasites in the “patinga” hybrid (*P. mesopotamicus* x *P. brachypomus*) (n=35) from the growout farm in the northwest of São Paulo State, Brazil.

Parasite	P (%)	MII	MA	Site of infection
Monogenea				
<i>Anacanthorus penilabiatu</i> s	88.6	9.5±2.0 (0.2-44.0)	8.4±1.9 (0-44.0)	Gills
<i>Mymarothecium viatorum</i>	100.0	261.0±57.2 (1.0-1331.0)	261.0±57.2 (1.0-1331.0)	
Protozoa				Gills/Skin (mucus)
<i>Trichodina</i> spp.	25.7	22.9±11.0 (1.0-94.0)	5.9±3.2 (0-94.0)	
<i>Ichthyophthirius multifiliis</i>	14.3	4.2±2.7 (1.0-15.0)	0.6±0.4 (0-15.0)	
Myxosporea				
<i>Henneguya</i> spp.	17.1	-	-	Kidney/Skin (mucus)
<i>Myxobolus</i> spp.	5.7	-	-	Kidney

the pacu and patinga were not compared because the number of patinga collected was too low.

Of the *P. mesopotamicus* analysed from the fee-fishing property (n = 28), *A. penilabiatu*s and *M. viatorum* were the most prevalent parasites. The prevalence of *R. rondoni* and third-stage larvae (L3) of the anisakid nematode *Contracaecum* sp. was also high. No larvae of *Contracaecum* sp. were found encysted in the musculature of the fish (Table 4).

No correlation was observed between parasite abundance and the condition of the fish in all properties (p > 0.05).

Considering all analysed *P. mesopotamicus* (n = 44) and hybrids (n = 61), regardless of the property from which they were sampled (Table 5), no difference was observed with respect to the prevalence of parasites that occur in common in the three properties (*A. penilabiatu*s, *M. viatorum*, *Trichodina* spp., *I. multifiliis*, *Henneguya* spp., and *Myxobolus* spp.). *Piaractus mesopotamicus* had a higher mean intensity of infection by *A. penilabiatu*s compared with the hybrid (p = 0.012) (Table 5). The reverse was observed for the mean intensity of infection (p = 0.005) and mean abundance (p = 0.002) of *M. viatorum*, which were significantly higher in the

hybrids (Table 5). No influence of parasitism on the condition of either *P. mesopotamicus* or the hybrids was observed (p > 0.05).

At all properties, the water quality was acceptable for tropical fish production on the sampling dates, with the exception of some observed variations in the water temperature, the concentration of ionised ammonia (NH₄⁺) and the nitrite level (Table 6).

Discussion

For the three properties studied, the fish presented a high prevalence of Dactylogyridae monogenoids, and the prevalence was particularly high in fish from the growout (P = 100.0%) and fee-fishing (P = 97.1%) properties. This finding may be associated with the fact that these parasites have a monoxenic cycle and reproduce quickly, especially in environments where there is a high density of hosts, facilitating their development cycle (PAVANELLI et al., 2008; TAKEMOTO et al., 2004). The presence of ectoparasites in the ponds and cages of fish farms is also directly related to water quality and management practices (MORAES; MARTINS, 2004). Thus, the confinement of fish in ponds, cages and tanks

Table 4. Mean values \pm standard error of the prevalence (P), mean intensity of infection (MII), mean abundance (MA) and site of infection of the parasites of the *Piaractus mesopotamicus* (n=28) and “patinga” hybrid (*P. mesopotamicus* x *P. brachypomus*) (n=7) from the fee-fishing farm in the northwest of São Paulo State, Brazil.

Parasite	<i>Piaractus mesopotamicus</i>			Hybrid			Site of infection
	P (%)	MI	MA	P (%)	MI	MA	
Monogenea							Gills
<i>Anacanthorus penilabialis</i>	96.4	17.5 \pm 3.7 (0.4-7.6)	16.9 \pm 3.6 (0-61.7)	100.0	5.8 \pm 1.8 (1.8-16.0)	5.8 \pm 1.8 (1.8-16.0)	
<i>Mymarothecium viatorum</i>	96.4	30.5 \pm 6.5 (0.6-107.0)	29.4 \pm 6.4 (0-107)	100.0	6.5 \pm 2.0 (2.1-17.9)	6.5 \pm 2.0 (2.1-17.9)	
<i>Notozothecium janauachensis</i>	-	-	-	14.3	2.0	0.3 \pm 0.3 (0-2.0)	
Protozoa							Gills/Skin
<i>Trichodina</i> spp.	14.3	1.5 \pm 0.3 (1.0-2.0)	0.2 \pm 0.1 (0-2.0)	14.3	1.0	0.1 \pm 0.1 (0-1.0)	
<i>Ichthyophthirius multifiliis</i>	3.6	19.0	0.7 \pm 0.7 (0-19.0)	-	-	-	
<i>Chilodonella</i> sp.	-	-	-	14.3	2.0	0.3 \pm 0.3 (0-2.0)	
Myxosporea							Gills/Skin/Kidney
<i>Henneguya</i> spp.	14.3	-	-	42.8	-	-	
<i>Myxobolus</i> spp.	10.7	-	-	14.3	-	-	Kidney/Spleen
Nematoda							Intestine
<i>Rondonia rondoni</i>	89.3	11874.4 \pm 2451.1 (675.0-49476.0)	10602.1 \pm 2295.2 (0-49476.0)	57.1	2905.0 \pm 1097.4 (1344.0-6138.0)	1660.3 \pm 829.8 (0-6138.0)	
<i>Contracaecum</i> sp. (larvae L3)	28.7	24.9 \pm 16.4 (1.0-136.0)	7.1 \pm 4.9 (0-136.0)	14.3	17.0	2.4 \pm 2.4 (0-17.0)	Mesentery/ Visceral cavity
Crustacea							Skin
<i>Dolops carvalhoi</i>	3.6	1.0	0.03 \pm 0.03 (0-1.0)	-	-	-	

Table 5. Mean values \pm standard error of prevalence (P), mean intensity of infection (MI) and mean abundance (MA) of the parasites found in common in *Piaractus mesopotamicus* (n=44) and the “patinga” hybrid (*P. mesopotamicus* x *Piaractus brachypomus*) (n=61) from the hatchery and larviculture, growout and fee-fishing farms in the northwest of São Paulo State, Brazil.

Parasites	<i>Piaractus mesopotamicus</i>			Hybrid		
	P (%)	MI	MA	P (%)	MI	MA
Monogenea						
<i>Anacanthorus penilabialis</i>	77.3	22.4 \pm 4.5* (0.4-111.6)	17.3 \pm 3.8 (0-111.6)	77.0	7.5 \pm 1.5* (0.1-44.0)	5.8 \pm 1.2 (0-44.0)
<i>Mymarothecium viatorum</i>	72.7	27.5 \pm 5.6* (0.2-107.3)	19.9 \pm 4.5* (0-107.3)	85.2	178.7 \pm 41.7* (0.8-1331.0)	152.3 \pm 36.4* (0-1331.0)
Protozoa (Ciliophora)						
<i>Trichodina</i> spp.	29.5	164.5 \pm 118.5 (1.0-1498.0)	48.6 \pm 35.9 (0-1498.0)	22.9	34.8 \pm 19.9 (1.0-275.0)	8.0 \pm 4.8 (0-275.0)
<i>Ichthyophthirius multifiliis</i>	4.5	12.5 \pm 6.5 (6.0-19.0)	0.6 \pm 0.4 (0-19.0)	8.2	4.2 \pm 2.7 (1.0-15.0)	0.3 \pm 0.2 (0-15.0)
Myxosporea						
<i>Henneguya</i> spp.	18.0	-	-	16.4	-	-
<i>Myxobolus</i> spp.	13.6	-	-	11.5	-	-

*Statistically significant difference ($p < 0.05$).

favours the proliferation of these parasites and facilitates their spread primarily due to the proximity between hosts and the accumulation of organic matter, and this causes great economic losses (FLORES-CRESPO et al., 1992; MARTINS et al., 2000)

In this study, *P. mesopotamicus* had higher rates of infestation with *A. penilabialis*, while hybrids were infested more frequently

with *M. viatorum* than *A. penilabialis*, suggesting that these fish have a higher susceptibility to these monogenoids. *Anacanthorus penilabialis* is a parasite that was previously reported in *P. mesopotamicus*, and *M. viatorum* was previously reported in *P. brachypomus* (BOEGER et al., 1995, 2002; PAMPLONA-BASILIO et al., 2001; COHEN; KOHN, 2005). As the hybrid

Table 6. Analysis of water quality in the samplings conducted between February 2010 and February 2011 at the hatchery and larviculture, growout and fee-fishing farms in the northwest of São Paulo State, Brazil. (Values expressed as the mean \pm standard error).

Parameters	Hatchery and larviculture	Growout	Fee-fishing	Reference values
Temperature ($^{\circ}\text{C}$)	25.7 \pm 1.5 (19.8-29.5)	25.8 \pm 1.6 (19.6-31.2)	26.4 \pm 1.5 (20.4-30.1)	20-30*
Transparency (cm)	27.0 \pm 3.9 (18.0-43.0)	32.8 \pm 4.4 (13.0-45.0)	36.7 \pm 2.4 (26.0-44.0)	25-45*
O ₂ D* (mg L ⁻¹)	5.8 \pm 0.5 (4.0-7.2)	3.9 \pm 0.5 (2.1-5.5)	4.7 \pm 0.8 (1.9-7.6)	3-10†
Total ammonia NH ₄ ⁺ (mg L ⁻¹)	0.3 \pm 0.15 (0-1.2)	0.4 \pm 0.2 (0-1.8)	0.6 \pm 0.3 (0-1.8)	<0.6*
Nitrite (mg L ⁻¹)	0.06 \pm 0.02 (0-0.2)	0.2 \pm 0.1 (0-0.7)	0.13 \pm 0.09 (0-0.7)	<0.3*
pH	7.5 \pm 0.1 (7.0-8.0)	7.3 \pm 0.2 (7.0-8.0)	7.3 \pm 0.09 (7.0-7.5)	6-8*,†
Toxic ammonia NH ₃ (mg L ⁻¹)	0.0076 \pm 0.004 (0-0.03)	0.01 \pm 0.007 (0-0.05)	0.01 \pm 0.007 (0-0.04)	<0.2*

*Ostrensky and Boeger (1998) and †Urbinati et al. (2010); reference values for the species production in general and specifically for the production of *Piaractus mesopotamicus*, respectively.

presented with a high infestation of *M. viatorum* only, it is possible that it has greater susceptibility to this monogenoid, the cause of which is unknown. Moreover, due to the monogenoid exhibiting specificity for the host species, confinement also favours the spread of these parasites, thus reinforcing the observations at the growout property.

Notozothecium janauachensis is a natural parasite of *C. macropomum* gills and has been described in specimens collected in Amazonas State, Brazil (BELMONT-JÉGU et al., 2004). This is the first occurrence of *N. janauachensis* in a patinga hybrid, and this finding may be related to the fact that fish from different species, including *C. macropomum*, are confined in the same pond, thus increasing the chances of erratic infestations. This fact may explain the low prevalence of this parasite in the sample. Moreover, fishes of the genera *Colossoma* and *Piaractus* are phylogenetically closely related (COHEN; KOHN, 2009), which could facilitate monogenoid infestations of the same species.

With respect to infestations caused by ciliate protozoa, and trichodinids of the genus *Trichodina* specifically, infections were higher in the hatchery system ($P = 37.1\%$). These protozoa have a monoxenic life cycle, which facilitates their transmission, especially in fish farms with a high stocking density.

On the sampling dates, all observed fish farms exhibited water of acceptable quality for the production of *P. mesopotamicus*, based on the conditions suggested by Ostrensky and Boeger (1998) and Urbinati et al. (2010). However, the water parameters may change during the year because the renewal systems were limited in the sampled properties, which can influence variations in the water's physical-chemical characteristics, especially its transparency, temperature, nitrogen compound concentration (ammonia and nitrite) and dissolved oxygen. In addition, in the fee-fishing property, sanitary breaks are not performed regularly, favouring nutrient accumulation and an increase in the decomposition process at the bottom of the pond, as also observed by Lizama et al. (2007) in fish farms in São Paulo State, Brazil. These alterations can contribute to an increase in parasitic diseases in fish farms.

The high density of fish observed in the growout and fee-fishing properties, which is related to the type of activity performed in these properties, also favours the accumulation of organic matter and nitrogen compounds in water. The supply of commercial foods in large quantities promotes the increased fertilisation of ponds and the accumulation of organic matter, which is often observed in growout (LIZAMA et al., 2007) and fee-fishing systems where

feed management is performed improperly, thus facilitating infections/infestations by pathogens. Interaction with fish from other species can cause stress, particularly to fish kept in captivity and in high stocking density (VAL et al., 2004). This contributes to the higher rates of parasitism observed in these properties.

Regarding the myxosporidians, *P. mesopotamicus* collected from the hatchery showed a higher prevalence of *Henneguya* spp. ($P = 25.0\%$) and *Myxobolus* spp. ($P = 18.7\%$) when compared with the results observed in samples from the other properties. Some species of fish parasites can affect a specific range of developmental periods (larvae/post-larval/juvenile/adult) more severely. In the case of the myxosporidians, infections usually occur during the juvenile stage, as these parasites live on the cartilaginous tissue, including cartilage close to the brain, causing lesions and destroying nerves in the brain related to motor coordination (PAVANELLI et al., 2008).

Under culture conditions, infections caused by myxosporidians can be facilitated by the accumulation of organic matter and the presence of intermediate hosts (oligochaetes) at the bottom of the ponds. Some species of myxosporidians have been described in *P. mesopotamicus*, such as *Henneguya lutzi*, *Henneguya piaractus*, *Henneguya pellucida*, *Myxobolus colossomatis* and *Myxobolus cuneus* (ADRIANO et al., 2006; EIRAS et al., 2008), but this is the first report of the occurrence of *Henneguya* sp. and *Myxobolus* sp. parasitising the patinga hybrid.

With respect to infections caused by nematodes in the fish of the fee-fishing property, the prevalence, mean intensity of infection and mean abundance of *R. rondoni* were considerably higher than those observed for the *Contracaecum* sp. larvae, especially in *P. mesopotamicus*. *Rondonia rondoni* is a monoxen parasite (MORAVEC, 1998) and can occur at high levels of infection, but it does not seem to cause significant damage to its hosts at a histological level (MARTINS; URBINATI, 1993). However, the high level of infection can lead to cases of intestinal obstruction, which can block the passage of food in the digestive tract (THATCHER, 2006; PARRA et al., 1997). High levels of *R. rondoni* in *P. mesopotamicus* were also recorded by Kohn et al. (1985) and Parra et al. (1997).

The Anisakidae nematodes are parasites of aquatic organisms, such as fish, marine mammals and piscivorous birds. Adult nematodes of the genus *Contracaecum* infect the intestines of piscivorous birds and marine mammals (MORAVEC, 1998). In fish, third stage larvae (L3) are located on visceral serosa and can migrate to muscles, where they encyst and, if ingested, are a

potential risk to human health because they may cause a zoonotic disease known as anisakiasis (TAVARES; LUQUE, 2006). The transmission of this disease is associated with the consumption of raw or undercooked fish meat containing third stage larvae of these parasites (the infective form) (TAVARES; LUQUE, 2006; THATCHER, 2006).

In the present study, larvae of *Contracaecum* sp. were not found encysted in the analysed fish muscles. This finding is relevant in view of the importance of this species in commercial fishing, and these fish were collected in an establishment that sells fish meat. On the fee-fishing property, where these larvae were found, regular fallowing is not performed. A sanitary break, followed by sanitisation using products such as lime, is one of the most effective ways to eliminate and control eggs, cysts, larvae and adults of parasites that may be present in the environment and to control the populations of some intermediate hosts, such as gastropods, annelids, insects and copepods (ROJAS, 2006). In addition, the presence of piscivorous birds around the pond allows the entry of new pathogens into the production system, especially endoparasites, which can complete their life cycle (LIZAMA et al., 2007). Thus, the use of protective screens around the ponds is an important measure for the prevention of this disease.

In this study, *P. mesopotamicus* and the patinga hybrid are described as new hosts for the third stage larvae of *Contracaecum* sp., thus widening the knowledge of the parasite-host relationship and geographic distribution of these helminth species. Some of the parasites were observed in all three farms sampled (*A. penilabiatus*, *M. viatorum*, *Trichodina* spp., *I. multifiliis*, *Henneguya* spp. and *Myxobolus* spp.), indicating epidemiological factors due to the transport of fish between the properties that form the production chain. The transport of fish between fish farms can act as a disseminator of agents with pathogenic potential (MARTINS et al., 2002). Moreover, it also acts as a stressful element, as the animals might be exposed to water with low oxygen or high ammonia concentrations and changes in temperature and pH, which predispose the fish to various infectious and parasitic diseases (MARTINS et al., 2002).

Despite the scarcity of data on the epidemiology of fish in the production chain, it is known that prophylactic management, which involves regular disinfection of ponds, disinfection of fish (eggs, juveniles and adults), sterilisation of instruments, quarantine, transport in controlled conditions, therapeutic baths, stocking controls associated with the monitoring of water quality in ponds and tanks of fish farms, and the training of fish farm workers regarding the importance of better handling practices, are essential measures to control pathogens. In the present study, the high stocking density observed in the growout and fee-fishing properties had a large influence on the rates of parasitism. In addition, in the fee-fishing property, the coexistence of different species of fish in the same pond influenced the rates of parasitism, as did the non-regular disinfection of ponds, which increased the contamination of the site with eggs and larvae of parasites and contributed to the presence of organisms that serve as intermediate hosts for parasites with heteroxenous life cycles (i.e., oligochaetes, molluscs, crustaceans, etc.).

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