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Parasitological assessment in hybrids Serrasalmidae fish farmed in Brazil

Avaliação parasitológica em híbridos de peixes Serrasalmidae cultivados no Brasil

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Abstract

This study evaluated the parasitic fauna of hybrids tambacu (*Colossoma macropomum* × *Piaractus mesopotamicus*) and patinga (*P. mesopotamicus* × *Piaractus brachypomus*) and their host-parasite-environment interaction during the warm and cold seasons in two fish farms located in the State of Mato Grosso do Sul, Brazil, South America. A total of 120 fish, being 60 fish for species (30 in the warm season and 30 in the cold season) were examined. Water quality was measured weekly to evaluate the interaction between environmental conditions and parasitism. Fifteen species of parasites were found: *Ichthyophthirius multifilis, Chilodonella hexasticha* e *Trichodina* sp. (Protozoa), *Henneguya piaractus, Myxobolus colossomatis* and *Myxobolus cuneus* (Myxozoa); *Anacanthorus penilabiatus, Mymarothecium boegeri, Mymarothecium viatorum*, and *Notozothecium janauachensis* (Monogenoidea), *Goezia spinulosa* and *Goezia* sp. (Nematoda), *Echinorhynchus jucundus* (Acanthocephala), and *Dolops carvalhoi, Lernaea cyprinacea* (Crustacea). The ciliate protozoan *I. multifilis* and monogenoids were the most prevalent parasites in the cold and warm seasons for both hybrids in the different culture systems intensive production and sportive fishing. It was observed that the serrasalmid hybrids are more susceptible to parasites and harbor high diversity of parasites in relation to the parental species *C. macropomum, P. mesopotamicus* and *P. brachypomus*, with a predominance of ectoparasites.

Keywords: Fish farming, hybridization, helminths, protozoans, water quality.

Resumo

Este estudo avaliou a fauna parasitária de híbridos tambacu (*Colossoma macropomum × Piaractus mesopotamicus*) e patinga (*P. mesopotamicus × Piaractus brachypomus*) e a interação hospedeiro-parasito-ambiente durante as estações quente e fria em duas pisciculturas localizadas no estado de Mato Grosso do Sul, Brasil, América do Sul. Foram examinados um total de 120 peixes, sendo 60 peixes por espécie. A qualidade da água foi mensurada semanalmente para avaliar a interação entre condições ambientais e parasitismo. Foram encontradas quinze espécies de parasitos: *lchthyophthirius multifiliis, Chilodonella hexasticha e Trichodina* sp. (Protozoa); *Henneguya piaractus, Myxobolus colossomatis* e *Myxobolus cuneus* (Myxozoa), *Anacanthorus penilabiatus, Mymarothecium boegeri, Mymarothecium viatorum* e *Notozothecium janauachensis* (Monogenoidea), *Goezia spinulosa* e *Goezia* sp. (Nematoda), *Echinorhynchus jucundus* (Acanthocephala), *Dolops carvalhoi eLernaea cyprinacea* (Crustacea). O protozoário ciliado *I. multifiliis* e helmintos monogenoides foram os parasitos mais prevalentes nas estações fria e quente para ambos os híbridos nos diferentes sistemas de cultivo, produção intensiva comercial e pesca esportiva. Observou-se que os híbridos de serrasalmídeos são mais suscetíveis aos parasitos e abrigam alta diversidade parasitária em relação às espécies parentais, *C. macropomum, P. mesopotamicus e P. brachypomus* em ambiente de cultivo com predominância de ectoparasitos.

Palavras-chave: Piscicultura, hibridização, helmintos, protozoários, qualidade da água.

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Introduction

Since the 1980s, several hybrids of fish of the family Serrasalmidae have been produced and grown commercially in Brazil and are prominent in the production of continental fish (Porto-Foresti et al., 2013). For this, native species are crossed with each other to produce hybrids with desirable characteristics for commercialization such as improved performance in aquaculture (Hashimoto et al., 2012) resistance to environmental changes (Martins et al., 2002) and parasites infestation (Jerônimo et al., 2016). The hybrids of greatest interest in the Central region of the country are the tambacu, resulting from the cross induced between tambaqui females (*Colossoma macropomum* Cuvier, 1818) and pacu males (*Piaractus mesopotamicus* Holmberg, 1887) and the patinga or papi, resulting from the cross between pacu females (*P. mesopotamicus*) and pirapitinga males (*Piaractus brachypomus* Cuvier, 1818) (Porto-Foresti et al., 2013). These are responsible for 42.212 tons of all freshwater fish production in the country (IBGE, 2017).

The increase in industrial production of native and hybrid freshwater fish in Brazil is concentrated in the central and northern regions of the country (Jerônimo et al., 2012). However, this increase implies cultivation following the intensive production model, which aims at high stocking density and maximum production. The adoption of this practice favors the development of infectious and parasitic diseases causing economic losses (Martins et al., 2002; Jerônimo et al., 2016; Ferreira et al., 2019). Considering this, the relevance of studies that evaluate the pathogen-host-environment interaction in species with potential for cultivation and commercialization has increased considerably in order to guarantee the improvement in the health conditions of the fish and consequently the production (Jerônimo et al., 2015).

Few parasitological studies have addressed the pathogen-host-environment interaction of hybrid fish in commercial production systems. In the northern region of Brazil, in fish farming in the State of Amapá, the poor water quality contributed to the high parasitism of the tambacu hybrid by ectoparasites (Silva et al., 2013). In addition, in a study of parasitic fauna of the tambatinga hybrid (*C. macropomum* × *P. brachypomus*) in different fish farms in this region, Dias et al. (2015) reported that the occurrence of ectoparasites was favored by poor quality management and sanitary conditions of the properties, whereas the presence of endoparasites was owing to the supply of nurseries along with water from natural water bodies. In the cultivation of a tambatinga hybrid in a net cages, Pinheiro et al. (2015) observed moderate levels of ectoparasite infestation with low species richness.

In view of the scarcity of health studies with hybrids fish, the present study aimed to evaluate parasitism in hybrid fish and the relationship during a production cycle in the Central Brazilian region.

Materials and Methods

Fish and study site

For this study, 120 fish and their host-parasite-environment interaction during the warm (october to march) and cold (may to august) seasons 2010 and 2011 were evaluated. Sixty tambacus (30 in the warm season and 30 in the cold season) and 60 patingas (30 in the warm season and 30 in the cold season), were collected from an intensive system located in the municipality of Itaporã (22°20'58.50"S; 54 ° 46'52.51"W) and from a semi-intensive system in the municipality of Dourados (21°59'29.12"S; 54°48'30.00"W), both in the Grande Dourados region, State of Mato Grosso do Sul, Brazil.

Fish capture and parasitological analysis

The fish were, euthanized by collected using trawl deep anesthetic with 75 mg L⁻¹ clove oil (Ethics Committee 23080.0 29979/2009-05/CEUA/UFSC), and subsequently necropsied for parasitological analysis according to recommendations by Jerônimo et al. (2011). To quantify helminth parasites, marked Petri dishes were used and analyzed with the aid of a stereomicroscope (Zeiss[®], Pleasanton, USA), and the protozoa were quantified with the aid of the Sedgewick Rafter camera (Pyser-SGI[®], London, England).

Identification of parasites and parasitological indexes

The identification of the parasites found followed the methodology proposed by Moravec (1998), Eiras et al. (2006), Thatcher (2006), and Cohen et al. (2013). Taxonomic identification of *Lernea*'s copepodite parasites was

performed based on Boxshall et al. (1997), Avenant-Oldewage & Robinson (1996) and Kabata (1979). Parasitic indices such as prevalence and mean intensity were calculated according to Bush et al. (1997).

Physico and chemical parameters of water

During the seasons, the physical and chemical characteristics of the breeding nurseries were monitored weekly. Dissolved oxygen, temperature, hydrogen potential, and electrical conductivity were measured using the HANNA multiparameter (Hanna Instruments®, Inc., USA); transparency with the Secchi disk; and total iron, orthophosphate, and alkalinity by the titration method (APHA, 2005). Samples from the water outlet were collected to measure total ammonia, nitrite, and nitrate using a colorimetric kit (Alfakit®, São Paulo, Brazil).

Statistical analysis

Statistical analyses were performed using software IBM[®] SPSS[®] Statistics 18. To compare the prevalence of parasites between hybrids and between seasons, the non-parametric Fisher exact test was used. Biometric data, physico and chemical parameters of water quality and the intensity of infestation were assessed using the Mann-Whitney (*U*) non-parametric test. The level of significance considered for all tests was 5% (p < 0.05). In order to better understand the relationship of the seasons (warm and cold) in the variations of the eleven water quality parameters, Principal Component Analysis (PCA) was performed in the cultivation of each hybrid. The active variables assumed were temperature, dissolved oxygen, water transparency, orthophosphate and ammonia, since together with pH they are the most important for an aquaculture water quality routine. The others variables, alkalinity, total iron, conductivity, nitrite and nitrate, as well as the pH, were considered supplementary variables because they suffered little variation, besides being unusual in the routine of analyzing a fish production.

Results

The fish were parasitized by *Ichthyophthirius multifiliis* Fouquet 1876, *Chilodonella hexasticha* Kiernik, 1909 and *Trichodina* sp. (Protozoa); *Henneguya piaractus* Martins & Souza 1997, *Myxobolus colossomatis* Molnár & Bekési 1993, *Myxobolus cuneus* Adriano, Arana & Cordeiro, 2006 (Myxozoa), *Anacanthorus penilabiatus* Boeger, Husak & Martins, 1995, *Mymarothecium boegeri* Cohen & Kohn 2005, *Mymarothecium viatorum* Boeger, Piasecki & Sobeck 2002, and *Notozothecium janauachensis* Belmont, Jegu, Domingues & Laterça, 2004 (Monogenoidea), *Goezia* sp. and *Goezia spinulosa* Rasheed, 1965 (Nematoda), *Echinorhynchus jucundus*, Travassos, 1923 (Acanthocephala) *Dolops carvalhoi* Lemos de Castro, 1949 and *Lernaea cyprinacea* Linnaeus, 1758 (Crustacea).

The average weight and average total length of tambacu in the warm season were significantly higher than those in the cold season (p < 0.05). There was no significant difference in the length and weight of the patinga hybrids between seasons (p > 0.05). The average weight and average total length of the tambacu was significantly greater than the patinga hybrid between seasons (p < 0.05) (Table 1).

Table 1. Mean values ± standard deviation of weight and length of the tambacu (*Colossoma macropomum* × *Piaractus mesopotamicus*) and patinga (*P. mesopotamicus* × *P. brachypomus*) hybrids farmed in the state of Mato Grosso do Sul, Central-West Brazil, in the warm and cold seasons.

Parameters	Tambacu				Patinga		Total		
	Warm Season (n=30)	Cold Season (n=30)	р	Warm Season (n=30)	Cold Season (n=30)	р	Tambacu (n=60)	Patinga (n=60)	p
Weight (g)	1356.1 ± 313.8	950.8 ± 173.9	0.010	667.3 ± 147.1	832.7 ± 175.6	0.647	1141.0 ± 296.0	724.0 ± 173.0	0.001
Length (cm)	38.4 ± 3.7	37.1 ± 2.6	0.002	32.7 ± 2.7	33.2 ± 2.8	0.210	37.6 ± 2.4	32.5 ± 2.3	0.001

n = number of fish sampled; (p < 0.05) indicate significant difference between seasons in each facility.

The results indicate that the joint representation of the two main factors represented 68% of the variance for the cultivation of patinga hybrid. The cold season showed a strong correlation with dissolved oxygen and transparency, whereas the warm season was strongly correlated with high values of ammonia, orthophosphate, nitrate, nitrite, and temperature (Figure 1). In contrast, in the cultivation of the tambacu hybrid, the PCA accounted for 59% of the variance. The cold season was correlated with higher values of dissolved oxygen, transparency, and

pH, whereas the warm season was correlated with higher values of temperature, ammonia, orthophosphate, and electrical conductivity (Figure 2). However, the physico and chemical parameters of water quality, for both species and evaluated seasons, were within the recommended values for the cultivation of tropical fish (Table 2), although nitrite levels were significantly higher in the warm season for both evaluated species.

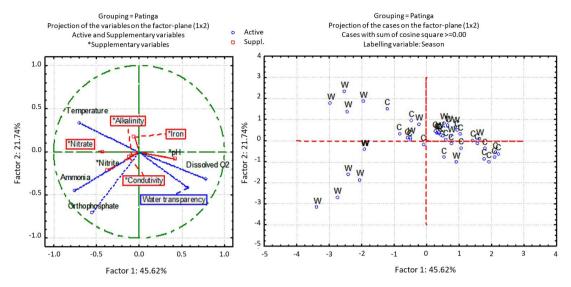


Figure 1. Principal Components Analysis (PCA) of the water quality parameters measured in the pond where patinga (*Piaractus mesopotamicus × Piaractus brachypomus*) were collected. C = cold season; W = warm season.

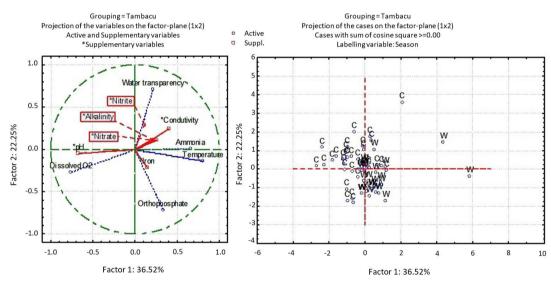


Figure 2. Principal Components Analysis (PCA) of the water quality parameters measured in the pond where tambacu (*Colossoma macropomum* × *Piaractus mesopotamicus*) were reared. C = cold season; W = warm season.

Myxozoa parasites were more prevalent (66.7%) in the warm season for the tambacu hybrid (p = 0.0006), whereas in the patinga hybrid, there was no effect of the season on the level of parasitic infection (p > 0.05). The parasitic prevalence did not differ between the different hybrids, regardless of the cold or hot season (p > 0.05) (Table 3).

Among crustacean parasites, copepodites of *L. cyprinacea* were more prevalent (p = 0.0033) in the patinga hybrid in the warm season; however, in the tambacu hybrid, the highest prevalence of this parasite was in the cold season (p = 0.0009). In the adult stage of *L. cyprinacea*, a higher prevalence in the warm season was observed in the tambacu hybrid, whereas for the patinga hybrid was not observed. *Dolops carvalhoi* was more prevalence in the patinga hybrid in the warm season (p = 0.0002), whereas in the tambacu hybrid was no found significant difference (p > 0.05). The prevalence of infestation/infection by Monogenoidea, and Nematoda were not influenced by the warm or cold season, or by the type of host (p > 0.05) (Table 3).

Table 2. Water quality parameters (mean values ± standard deviation) from ponds of the tambacu (*Colossoma macropomum* × *Piaractus mesopotamicus*) and patinga (*P. mesopotamicus* × *P. brachypomus*) hybrids farmed in the state of Mato Grosso do Sul, Central-West Brazil, in the warm and cold seasons.

Parameters —	Taml	pacu	Patinga			
Parameters -	Warm Season	Cold Season	Warm Season	Cold Season		
Total Alkalinity (mg L ⁻¹)	48.57 ± 11.65	50.68 ± 7.43	40.69 ± 6.77	40.74 ± 5.28		
рН	7.07 ± 0.59	7.27 ± 0.30	7.91 ± 1.37	7.09 ± 0.42		
Conductivity (µS cm ⁻¹)	62.21 ± 12.01	63.56 ± 8.21	31.00 ± 4.55	35.95 ± 9.27		
Dissolved Oxygen (mg L ⁻¹)	5.39 ± 1.56	6.76 ± 1.06	6.33 ± 1.07	7.41 ± 1.55		
Temperature (°C)	25.10 ± 3.44	19.87 ± 3.01	24.93 ± 3.27	20.44 ± 3.23		
Transparency (cm)	16.54 ± 2.92	18.38 ± 5.52	21.32 ± 5.11	25.70 ± 9.75		
Ammonia (mg L ⁻¹)	0.19 ± 0.12	0.12 ± 0.06	0.25 ± 0.22	0.14 ± 0.12		
Total iron (mg L ⁻¹)	0.26 ± 0.41	0.37 ± 0.60	0.29 ± 0.28	0.20 ± 0.41		
Nitrate (mg L ⁻¹)	0.42 ± 0.46	0.30 ± 0.45	0.24 ± 0.99	0.29 ± 1.17		
Nitrite (mg L ⁻¹)	0.18 ± 0.03ª	0.04 ± 0.08^{b}	0.20 ± 0.07^{a}	0.03 ± 0.03^{b}		
Orthophosphate (mg L ⁻¹)	0.66 ± 0.36	0.33 ± 0.36	0.16 ± 0.34	0.03 ± 0.14		

Different letters indicate indicate significant difference between seasons in each facility.

Table 3. Prevalence of parasites (%) in the tambacu (*Colossoma macropomum × Piaractus mesopotamicus*) and patinga (*P. mesopotamicus × P. brachypomus*) hybrids farmed in the state of Mato Grosso do Sul, Central-West Brazil, in the cold and warm seasons.

		Tambacu			Patinga			Total		
Parasites	Cold Season (n=30)	Warm Season (n=30)	р	Cold Season (n=30)	Warm Season (n=30)	р	Tambacu (n=60)	Patinga (n=60)	р	Site of infection
CILIOPHORA										
lchthyophthirius multifiliis	83.3	100	0.0522	76.7	100	0.0105	91.7	88.3	0.7623	Gill and Tegument
<i>Trichodina</i> sp.	3.3	0	0.5000	3.3	3.3	0.7542	1.7	3.3	0.5000	Gill and Tegument
Chilodonella hexasticha	10	3.3	0.6120	10	20	0.0471	6.7	10	0.2950	Gill
MIXOSPOREA	20	66.7	0.0006	66.7	53.3	0.4296	48.3	60	0.0997	Gill, liver and kidney
Henneguya piaractus	+	+		+	+		+	+		
Myxobolus colossomatis	+	+		+	+		+	+		
Myxobolus cuneus	+	+		-	-		+	-		
MONOGENOIDEA	100	93.3	0.4915	100	100	0.5000	96.6	100	0.4958	Gill and Tegument
Anacanthorus penilabiatus	+	+		+	+		+	+		
Mymarothecium boegeri	+	+		-	-		+	+		
Mymarothecium viatorum	-	-		+	+		-	-		
Notozothecium janauachencis	+	+		-	-		+	+		
NEMATODA	3.3	0	0.500	16.7	0	0.0522	1.7	8.3	0.2068	Stomach
<i>Goezia</i> sp.	+	+		+	+		+	+		
Goezia spinulosa	+	+		-	-		+	-		
ACANTHOCEPHALA	73.3	96	0.0256	43.3	43.3	0.6026	85	43.3	0.0000	Stomach and Intestine
Echinorhyncus jucundus	+	+		+	+		+	+		
CRUSTACEA										Gill and Tegument
Dolops carvalhoi	36.7	16.7	0.1432	3.3	46.7	0.0002	26.7	25	0.5000	Tegument
Lernaea cyprinacea (copepodids)	56.7	13.3	0.0009	20	60	0.0033	35	40	0.7063	Gill and Nostril
Lernaea cyprinacea	53.3	86.7	0.0101	*	*	*	70	*		Tegument

(+) presence of the parasite; (-) absence of the parasite; (*) not observed; n = number of fish sampled; (p < 0.05) indicate significant difference between seasons for each parasite in each facility.

A higher mean intensity of infestation by the ciliated protozoan *I. multifiliis* (p = 0.001), Monogenoidea (p = 0.01), and copepodites of *L. cyprinacea* (p = 0.007) was observed in the tambacu hybrid in the warm season; furthermore, the average intensity of infection by Acanthocephala in the tambacu hybrid was higher in the warm season (p = 0.002) (Table 4). In the patinga hybrid, a higher mean infestation intensity was observed for the *L. cyprinacea* copepodites in the warm season. When compared to the average infestation/infection intensity among hybrids, no significant difference was observed for the identified parasites.

Table 4. Mean intensity (average ± standard deviation, minimum and maximum values in parenthesis) of parasitism in the tambacu (*Colossoma macropomum × Piaractus mesopotamicus*) and patinga (*P. mesopotamicus × P. brachypomus*) hybrids farmed in the state of Mato Grosso do Sul, Central-West Brazil, in the cold and warm seasons.

	Tambacu			Patinga			Total		
Parasites	Cold Season	Warm Season	р	Cold Season	Warm Season	р	Tambacu	Patinga	р
CILIOPHORA									
lchthyophthirius multifiliis	515 ± 396 (170 – 1650)	2681 ± 2874 (233 – 16253)	0.001	3145 ± 3244 (127 – 13650)	3318 ± 3968 (160 - 15333)	0.936	1678 ± 2371 (170 – 16253)	3220 ± 3541 (127 – 1533)	0.143
Trichodina sp.	177 ± 177 (177 – 177)	0	0.622	153 ± 153 (153 – 153)	470 ± 470 (470 – 470)	0.177	177 ± 177 (177 – 177)	312 ± 224 (153 – 470)	0.403
MONOGENOIDEA	11.0 ± 10.2 (1 – 43)	97.6 ± 81.9 (1 – 285)	0.010	78.9 ± 70.5 (3 – 314)	83.7 ± 42.4 (18 – 172)	0.267	54.4 ± 72.4 (1 – 285)	81.3 ± 57.7 (3 – 314)	0.159
NEMATODA	2.0 ± 2.0 (2 – 2)	0	0.189	3.4 ± 1.5 (2 – 5)	0	0.612	2.0 ± 2.0 (2 - 2)	3.4 ± 1.5 (2 – 5)	0.083
ACANTHOCEPHALA	4.6 ± 3.4 (1 – 17)	12.8 ± 2.2 (1 – 27)	0.002	2.9 ± 3.3 (1 – 13)	5.3 ± 6.8 (1 – 24)	0.810	4.4 ± 3.4 (1 – 17)	4.0 ± 5.3 (1 – 24)	0.171
CRUSTACEA									
Dolops carvalhoi	46.6 ± 41.0 (1 – 180)	12.0 ± 10.5 (1 – 34)	0.465	1.0 ± 1.0 (1 – 1)	0	0.103	1.0 ± 1.0 (1 – 1)	1.0 ± 1.0 (1 – 1)	0.060
<i>Lernaea cyprinacea</i> (copepodids)	1.0 ± 0.0 (1 – 4)	56.5 ± 48.2 (1 – 214)	0.007	1.8 ± 0.8 (1 – 3)	8.8 ± 1.4 (1 – 42)	0.001	46.0 ± 48.5 (1 – 214)	7.0 ± 9.5 (1 – 42)	0.154

(p < 0.05) indicate significant difference between seasons for each parasite in each facility.

Discussion

The environmental stress associated with fluctuations in water quality parameters such as temperature, oxygen levels, pH and levels of nitrogen compounds can increase fish susceptibility to parasitic infections in the farming environment (Schalch & Moraes, 2005; Santos et al., 2013; Lizama et al., 2007). The strong correlation observed in the warm season with high values of ammonia, orthophosphate, nitrate, nitrite and temperature, justifies the greater intensity of monogenoids and *I. multifiliis* parasites in the tambacu hybrid, environmental conditions that favored the development of these parasites. Since parasitism by monogenoids, *P. pillulare* and myxosporids is favored by high annual temperatures in tambacu hybrids (Schalch & Moraes, 2005). Studies also show that the population dynamics of monogenoids is influenced by water temperature, directly affecting the reproduction and survival of these parasites (Blažek et al., 2008).

High levels of nitrogenous compounds can induce chronic toxicity leading to reduced growth and fish tolerance to disease (Lizama et al., 2007). With the increase in water temperature, the metabolic rate of fish increases, which contributes to high levels of excretion of nitrogenous compounds (Pereira & Mercante, 2005). In addition, the increase in these compounds is mainly related to the high cultivation density, low water renewal rate (Lizama et al., 2007). The increase in the decomposition rate is also an important factor, which can be proven by the greater correlation of electrical conductivity in the warm season in the cultivation of tambacu hybrid, corroborating the results of the present study. Nitrite toxicity is greater in environments with lower oxygen concentration (Boyd, 1998). Although a higher content of this intermediate compound was observed in the hot season in the cultivation of both hybrids, the oxygen levels in the different cultivation systems did not suffer sudden variations during the cultivation period, which contributed to the maintenance of adequate conditions for the development of the hybrids fish in good health.

Parasitological assessment of hybrids Serrasalmidae

Ectoparasitic species were found to have a high prevalence of infestation in both studied hybrids. The proliferation of *I. multifiliis* is strongly related of water temperatures above 24°C, when the parasite's life cycle is favored and completed rapidly (Martins et al., 2015). However, this parasite has been reported in the tambacu hybrid at average water temperature of 30 °C in nurseries in properties with low oxygen levels, and poor nutritional and sanitary conditions (Silva et al., 2013). This is consistent with the observations in the present study of a higher prevalence of *I. multifiliis* infestation in the warm season in both hybrids. This fact can be explained as being due to the production environment having an accumulation of organic matter which is an ideal condition for greater proliferation of ciliated protozoan parasites (Martins et al., 2015). These parasites are good indicators of the quality of the cultivation environment because they usually infest the gills and integument and are exposed to the environment throughout their monoxenic life cycle (Tavares-Dias et al., 2014; 2015). Despite the high prevalence observed, no clinical signs of ichthyophthiriasis were found.

This is the first report of *C. hexasticha* for Serrasalmidae hybrids However, the infestation was less than that observed in *P. mesopotamicus* (Pádua et al., 2013), which justifies the non-occurrence of clinical signs owing to parasitic infestation. *Trichodina* sp. are reported as parasitizing *C. macropomum* (Fujimoto et al., 2019), *P. mesopotamicus* (Franceschini et al., 2013), and their hybrids tambatinga (Dias et al., 2015) and patinga (Jerônimo et al., 2012; Franceschini et al., 2013). In addition, there are occurrences of *T. heterodentata* and *T. colisae* in *P. mesopotamicus* (Jerônimo et al., 2012). The reduction in dissolved oxygen levels and high levels of eutrophication may favor the reproduction of *Trichodina* sp. (Maciel et al., 2018), which supports the results of the present study having a low prevalence of infestation by *Trichodina* sp., attributed to the fact that the water quality parameters were within the necessary conditions for the cultivation of Serrasalmidae hybrids (Boyd, 1998).

Myxosporids are cosmopolitan parasites and can infect different organs in fish (Lom & Dyková, 2006). In juveniles of tambaqui, *M. colossomatis* was observed in the fins, gills, heart, and intestinal membrane (Molnár & Békesi, 1993) gills and skin (Santos et al., 2013), while *M. cuneus* was found parasitizing *P. mesopotamicus* in the gallbladder, urinary bladder, gills, spleen, fins, surface of the head, liver, and heart (Adriano et al., 2006). Similar studies also report the presence of parasites of *Myxobolus* sp. in *C. macropomum* blood (Maciel et al., 2011). Infections in the branchial system caused by myxosporids result in direct or indirect damage to the health of their hosts (Videira et al., 2016; Araújo et al., 2018). Despite the moderate prevalence of myxosporid parasitism in the present study, the hosts did not manifest clinical signs of the disease in unfavorable seasons. This corroborates the observations of Silva et al. (2019) who described *Myxobolus* sp. as causing asymptomatic disease in *Astyanax* aff. *bimaculatus*. Studies have described parasitic infections by *Henneguya* sp. in *P. mesopotamicus* (Sant'Ana et al., 2012; Videira et al., 2016), *C. macropomum* (Schalch & Moraes, 2005), and the tambacu hybrid (Martins et al., 2002).

Monogenean *M. boegeri* was initially registered in the gills of *C. macropomum* (Cohen & Kohn, 2005), but it also infests the gills of the tambatinga hybrid (Cohen & Kohn, 2009; Dias et al., 2015; Pinheiro et al., 2015) and the tambacu hybrid as observed in the present study. The monogenoid *N. janauachensis* was registered in the gills of *C. macropomum* and in the tambacu (Silva et al., 2013) and tambatinga (Dias et al., 2015) hybrids, a fact that supports the results of the present study in which this particular parasite was present in the tambacu hybrid but absent in the patinga hybrid. *A. penilabiatus* and *M. viatorum* were found in *P. mesopotamicus* (Jerônimo et al., 2014) and *P. brachypomus* (Cohen & Kohn, 2009), and in the tambacu (Silva et al., 2013) and patinga (Franceschini et al., 2013) hybrids. However, in this study, parasitism by *M. viatorum* was observed only in a patinga hybrid; such variations can be attributed to the specificity of the host, which varies between parasitic species (Whittington et al., 2000).

In South America, five species of the nematode *Goezia* Zeder, 1800 are known: *G. spinulosa*, *Goezia intermedia* Rasheed, 1965, *Goezia brasiliensis* and *Goezia brevicaeca* Moravec, Kohn & Fernandes, 1994, and *Goezia leporini* Martins & Yoshitoshi, 2003. In the present study, their prevalence was low, occurring only in the cold season for both hybrids, and not manifesting any clinical signs. Larvae of the nematode *Goezia* sp., were found in *P. mesopotamicus* and several other species of fish (Kohn et al., 2011). However, there have been no reports in the patinga and tambacu hybrids. Thus, this study contributes to expanding knowledge regarding hosts capable of hosting this species of helminth.

The acanthocephalan *E. jucundus* was more prevalent in the tambacu hybrid in the warm season, whereas in the patinga hybrid, the level of infection was lower, which shows greater susceptibility of the tambacu hybrid to this parasite. Species of *Echinorhynchus* are common in natural environments and fish farms where intermediate hosts are present (Thatcher, 2006). However, in the cultivation of the tambacu hybrid, the sanitary conditions provided a higher prevalence of this group of endohelminths in the warm season. Thus, one can infer the possible host-parasite interaction by the *bottom-up* effect, as environments rich in organic matter increase primary production at higher

temperatures (Kratina et al., 2012) and such temperature variations are responsible for the regulation of all trophic chains. In this manner, arthropods that act as intermediate hosts (primary consumers) of the endohelminth cycle benefit from this environment, increasing their biomass and thereby increasing the resources for the development of the parasite in the aquatic culture environment. Thus, there is a greater possibility of contact between the final host and the intermediate host containing the infective form of the parasite.

Dolops carvalhoi was found parasitizing in the body surface, fins, and gills of several species of wild and captive fish (Thatcher, 2006). Studies show that they cause diseases in *P. mesopotamicus, C. macropomum,* and in the tambacu hybrid (Schalch et al., 2009); however, this is the first record of this parasite in a patinga hybrid. In the present study, the lernaeids were also observed during periods of high temperature in the cultivation of the tambacu hybrid; in contrast, copepodites, the initial phase of their life cycle, were found to be more prevalent in the cold season for the tambacu hybrid and in the warm season for the patinga hybrid. A study has shown that high parasitism by these parasites causes epizooty in larvae of *C. macropomum* leading to important economic losses (Delgado et al., 2011). Although fish in this study showed high parasitic infestation, mortality did not occur as they were adult fish in good body condition. According to Thatcher (2006), this group of parasites cause high mortality, especially in young or small fish.

The complex environment-parasite interaction is what determines the occurrence of infestation/infection, where the parasite initially tries to establish itself in the host while the latter resists infestation/infection through its defense mechanisms. Therefore, the host's susceptibility and resistance will determine whether the infestation/ infection is established (Tavares-Dias et al., 2017). In this case, possibly the warm season provided a favorable environment for the development and proliferation of parasites, besides allowing greater susceptibility of the host to infestations. The hybrids harbored species of parasites common to both parental species; hybridization between species affected the specificity of the host. This fact may represent an important ecological issue owing to interspecific hybridization (Jerônimo et al., 2016). However, the role of hybridization in fish in the specificity of the host is not yet clear as there may be changes in the parasitic fauna owing to the adaptation of the host (Simková et al., 2013). In addition, the levels of infestations by such parasites are, in general, mainly influenced by the management and environmental conditions of the crop (Silva et al., 2013; Tavares-Dias et al., 2014, 2015).

Conclusions

In fish farms, serrasalmid fish hybrids are more susceptible and harbor a high diversity of parasites when compared to the parental species. The parasites *C. hexasticha* and *Goezia* sp., and *D. carvalhoi* were recorded for the first time in the hybrid fish. The prevalence and intensity of infection are strictly related to the environment during the fish production cycle.

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References

Adriano EA, Arana S, Cordeiro NS. *Myxobolus cuneus* n. sp. (Myxosporea) infecting the connective tissue of *Piaractus mesopotamicus* (Pisces: Characidae) in Brazil: histopathology and ultrastructure. *Parasite* 2006; 13(2): 137-142. http://dx.doi.org/10.1051/ parasite/2006132137. PMid:16800122.

American Water Works Association, Water Environmental Federation – APHA. *Standard Methods for the Examination of Water and Wastewater*. 21st ed. Washington: APHA; 2005.

Araújo RS, Corrêa F, Sousa FB, Ramos ABMA, Sindeaux JL No, Matos ER. Ocorrência de *Myxobolus* sp. (Myxozoa) em *Thoracocharax stellatus* (Kner, 1858) (Characiformes) em um igarapé da floresta amazônica, Pará, Brasil. *Braz J Aquat Sci Technol* 2018; 21(1): 16-20. https://doi.org/10.14210/bjast.v21n1.11102.

Parasitological assessment of hybrids Serrasalmidae

Avenant-Oldewage A, Robinson J. Aspects of the morphology of the parasitic copepod *Lernaea cyprinacea* Linnaeus, 1758 and notes on its distribution in Africa. *Crustaceana* 1996; 69(5): 610-626. http://dx.doi.org/10.1163/156854096X00628.

Blažek R, Jarkovský J, Koubková B, Gelnar M. Seasonal variation in parasite occurrence and microhabitat distribution of monogenean parasites of gudgeon *Gobio gobio* (L.). *Helminthologia* 2008; 45(4): 185-191. http://dx.doi.org/10.2478/s11687-008-0037-9.

Boxshall GA, Montú MA, Schwarzbold A. A new species of *Lernaea* L. (Copepoda: Cyclopoida) from Brazil, with notes on its ontogeny. *Syst Parasitol* 1997; 37(3): 195-205. http://dx.doi.org/10.1023/A:1005899405901.

Boyd CE. *Water quality for pond aquaculture*. Auburn: International Center for Aquaculture and Aquatic Environments/Alabama Agricultural Experiment Station/Auburn University; 1998.

Bush AO, Lafferty KD, Lotz JM, Shostak AW. Parasitology Meets Ecology on Its Own Terms: Margolis et al. Revisited. *J Parasitol* 1997; 83(4): 575-583. http://dx.doi.org/10.2307/3284227. PMid:9267395.

Cohen SC, Justo MCN, Kohn A. South American Monogenoidea parasites of fishes, amphibians and reptiles. Rio de Janeiro: Oficina de Livros; 2013.

Cohen SC, Kohn A. A new species of *Mymarothecium* and new host and geographical records for *M. viatorum* (Monogenea: Dactylogyridae), parasites of freshwater fishes in Brazil. *Folia Parasitol (Praha)* 2005; 52(4): 307-310. http://dx.doi.org/10.14411/ fp.2005.042. PMid:16405294.

Cohen SC, Kohn A. On Dactylogyridae (Monogenea) of four species of characid fishes from Brazil. *Check List* 2009; 5(2): 351-356. http://dx.doi.org/10.15560/5.2.351.

Delgado PM, Delgado JPM, Arenas JV, Orbe RI. Infestación masiva por *Perulernaea gamitanae* (Crustacea: Cyclopoida: Lernaeidae) en juveniles de gamitana, cultivados en la Amazonia peruana. *Vet Mex* 2011; 42(1): 59-64.

Dias MKR, Neves LR, Marinho RGB, Pinheiro DA, Tavares-Dias M. Parasitismo em tambatinga (*Colossoma macropomum x Piaractus brachypomus*, Characidae) cultivados na Amazônia, Brasil. *Acta Amazon* 2015; 45(2): 231-238. http://dx.doi.org/10.1590/1809-4392201400974.

Eiras JC, Takemoto RM, Pavanelli GC. Métodos de estudo e técnicas laboratoriais em parasitologia de peixes. Maringá: EDUEM; 2006.

Ferreira LC, Cruz MG, Lima TBC, Serra BNV, Chaves FCM, Chagas EC, et al. Antiparasitic activity of *Mentha piperita* (Lamiaceae) essential oil against *Piscinoodinium pillulare* and its physiological effects on *Colossoma macropomum* (Cuvier, 1818). *Aquaculture* 2019; 512: 734343. http://dx.doi.org/10.1016/j.aquaculture.2019.734343.

Franceschini L, Zago AC, Schalch SH, Garcia F, Romera DM, Silva RJ. Parasitic infections of *Piaractus mesopotamicus* and hybrid (*P. mesopotamicus x Piaractus brachypomus*) cultured in Brazil. *Rev Bras Parasitol Vet* 2013; 22(3): 407-414. http://dx.doi.org/10.1590/S1984-29612013000300015. PMid:24142174.

Fujimoto RY, Hide DMV, Paixão PEG, Abe HA, Dias JAR, Sousa NC, et al. Fauna parasitária e relação parasito-hospedeiro de tambaquis criados na região do Baixo São Francisco, nordeste do Brasil. *Arq Bras Med Vet Zootec* 2019; 71(2): 563-570. http:// dx.doi.org/10.1590/1678-4162-10306.

Hashimoto DT, Senhorini JÁ, Foresti F, Porto-Foresti F. Interspecific fish hybrids in Brazil: management of genetic resources for sustainable use. *Rev Aquacult* 2012; 4(2): 108-118. http://dx.doi.org/10.1111/j.1753-5131.2012.01067.x.

Instituto Brasileiro de Geografia e Estatística – IBGE. *Pesquisa Pecuária Municipal* [online]. Rio de Janeiro: IBGE; 2017 [cited 2020 Aug 18]. Available from: https://sidra.ibge.gov.br/Tabela/3940#resultado

Jerônimo GT, Franceschini L, Zago AC, Silva RJ, Pádua SB, Ventura AS, et al. Parasitos de Peixes Characiformes e seus Híbridos Cultivados No Brasil Parasites from characiform fishes and its hybrids farmed in Brazil. In: Tavares-Dias M, Mariano WS. *Aquicultura no Brasil: novas perspectivas*. São Carlos: Pedro & João Editores; 2015. p. 283-304.

Jerônimo GT, Marchiori NC, Pádua SB, Dias J No, Pilarski F, Ishikawa MM, et al. *Trichodina colisae* (Ciliophora: Trichodinidae): new parasite records for two freshwater fish species farmed in Brazil *Braz. J Vet Parasitol* 2012; 21(4): 366-371. http://dx.doi. org/10.1590/S1984-29612012005000008. PMid:23207983.

Jerônimo GT, Martins ML, Ishikawa MM, Ventura AS, Tavares-Dias M. Métodos para coleta de parasitos de peixes. *Circular Técnica Embrapa Macapá* 2011; 39(1): 1-6.

Jerônimo GT, Pádua SB, Bampi D, Gonçalves ELT, Garcia P, Ishikawa MM, et al. Haematological and histopathological analysis in South American fish *Piaractus mesopotamicus* parasitized by monogenean (Dactylogyridae). *Braz J Biol* 2014; 74(4): 1000-1006. http://dx.doi.org/10.1590/1519-6984.09513. PMid:25627614.

Jerônimo GT, Pádua SB, Ventura AS, Gonçalves ELT, Ishikawa MM, Martins ML. Parasitological assessment in the hybrid surubim (*Pseudoplatystoma reticulatum x P. corruscans*), with uncommon occurrence of Monogenea parasites. *Rev Bras Parasitol Vet* 2016; 25(2): 179-186. http://dx.doi.org/10.1590/S1984-29612016037. PMid:27334818.

Kabata Z. Parasitic copepoda of british fishes. Londres: The Ray Society; 1979.

Kohn A, Moravec F, Cohen SC, Canzi C, Takemoto RM, Fernandes BMM. Helminths of freshwater fishes in the reservoir of the Hydroelectric Power Station of Itaipu, Paraná, Brazil. *Check List* 2011; 7(5): 681-690. http://dx.doi.org/10.15560/7.5.681.

Kratina P, Greig HS, Thompson PL, Carvalho-Pereira TSA, Shurin JB. Warming modifies trophic cascades and eutrophication in experimental freshwater communities. *Ecology* 2012; 93(6): 1421-1430. http://dx.doi.org/10.1890/11-1595.1. PMid:22834382.

Lizama MAP, Takemoto RM, Ranzani-Paiva MJT, Ayroza LM, Pavanelli GC. Relação parasito-hospedeiro em peixes de pisciculturas da região de Assis, estado de São Paulo, Brasil. 1. *Oreochromis niloticus* (Linnaeus, 1757). *Acta Sci Biol Sci* 2007; 29(2): 223-231. http://dx.doi.org/10.4025/actascibiolsci.v29i2.594.

Lom J, Dyková I. Myxozoan genera: definition and notes on taxonomy, life-cycle terminology and pathogenic species. *Folia Parasitol (Praha)* 2006; 53(1): 1-36. http://dx.doi.org/10.14411/fp.2006.001. PMid:16696428.

Maciel PO, Affonso EG, Boijink CL, Tavares-Dias M, Inoue LAKA. *Myxobolus* sp. (Myxozoa) in the circulating blood of *Colossoma macropomum* (Osteichthyes, Characidae). *Rev Bras Parasitol Vet* 2011; 20(1): 82-84. http://dx.doi.org/10.1590/S1984-29612011000100018. PMid:21439240.

Maciel PO, Garcia F, Chagas EC, Fujimoto RY, Tavares-Dias M. Trichodinidae in commercial fish in South America. *Rev Fish Biol Fish* 2018; 28(1): 33-56. http://dx.doi.org/10.1007/s11160-017-9490-1.

Martins ML, Cardoso L, Marchiori NC, Benites de Pádua S. Protozoan infections in farmed fish from Brazil: diagnosis and pathogenesis. *Braz J Vet Parasitol* 2015; 24(1): 1-20. http://dx.doi.org/10.1590/S1984-29612015013. PMid:25909248.

Martins ML, Onaka EM, Moraes FR, Bozzo FR, Paiva AMFC, Gonçalves A. Recent studies on parasitic infections of freshwater cultivated in the state of São Paulo, Brazil. *Acta Sci Biol Sci* 2002; 24(4): 981-985.

Molnár K, Békési L. Description of a new *Myxobolus* species, *M. colossomatis* n. sp. from the teleost *Colossoma macropomum* of the Amazon River basin. *J Appl Ichthyology* 1993; 9(1): 57-63. http://dx.doi.org/10.1111/j.1439-0426.1993.tb00388.x.

Moravec F. Nematodes of freshwater fishes of the Neotropical Region. Czech Republic: Academia Praha; 1998.

Pádua SB, Martins ML, Carraschi SP, Cruz C, Ishikawa MM. *Trichodina heterodentata* (Ciliophora: Trichodinidae): a new parasite for *Piaractus mesopotamicus* (Pisces: Characidae). *Zootaxa* 2012; 3422(1): 62-68. http://dx.doi.org/10.11646/zootaxa.3422.1.4.

Pádua SB, Martins ML, Carrijo-Mauad JR, Ishikawa MM, Jerônimo GT, Dias-Neto J, et al. First record of *Chilodonella hexasticha* (Ciliophora: Chilodonellidae) in Brazilian cultured fish: A morphological and pathological assessment. *Vet Parasitol* 2013; 191(1-2): 154-160. http://dx.doi.org/10.1016/j.vetpar.2012.07.030. PMid:22902259.

Pereira LPF, Mercante CTJ. A amônia nos sistemas de criação de peixes e seus efeitos sobre a qualidade da água. Uma revisão. *Bol Inst Pesca* 2005; 31(1): 81-88.

Pinheiro DA, Santos EF, Neves LR, Tavares-Dias M. Ectoparasitos em híbrido tambatinga provenientes de piscicultura em tanquerede no estado do Amapá (Brasil). *Bol Inst Pesca* 2015; 41(2): 409-417.

Porto-Foresti F, Hashimoto DT, Senhorini JA, Foresti F. Hibridização em piscicultura: monitoramento e perspectivas. In: Baldisserotto B, Gomes LC, editors. *Espécies nativas para piscicultura no Brasil*. Santa Maria: Editora UFSM; 2013. p. 589-601.

Sant'Ana FJF, Oliveira SL, Rabelo RE, Vulcani VAS, Silva SMG, Ferreira JA Jr. Surtos de infeccão por *Piscinoodinium pillulare* e *Henneguya* spp. em pacus (*Piaractus mesopotamicus*) criados intensivamente no Sudoeste de Goiás. *Pesq Vet Bras* 2012; 32(2): 121-125. http://dx.doi.org/10.1590/S0100-736X2012000200005.

Santos EF, Tavares-Dias M, Pinheiro DA, Neves LR, Marinho RGB, Dias MKR. Parasitic fauna of tambaui *Colossoma macropomum* (Characidae) farmed in cages in the State of Amapá, eastern Amazon. *Acta Amazon* 2013; 43(1): 107-114. http://dx.doi.org/10.1590/ S0044-59672013000100013.

Schalch SHC, Moraes FR. Distribuição sazonal de parasitos branquiais em diferentes espécies de peixes em pesque-pague do município de Guariba-SP, Brasil. *Braz J Vet Parasitol* 2005; 14(4): 141-146.

Schalch SHC, Soares VE, Morais FR. Praziquantel, levamisol e diflubenzuron no controle de *Dolops carvalhoi* (Crustacea: Branchiura) e *Anacanthorus penilabiatus* (Monogenea: Dactylogyridae) em *Piaractus mesopotamicus* Holmberg, 1887 (Osteichthyes: Characidae). *Braz J Vet Parasitol* 2009; 18(1): 53-59. http://dx.doi.org/10.4322/rbpv.01801010.

Silva MF, Sousa-Henrique DD, Messias-Luz N, Borralho LS, Oliveira JD, Sindeaux-Neto JL, et al. *Myxobolus* sp. (Myxozoa; Myxosporea) causing asymptomatic parasitic gill disease in *Astyanax* aff. *bimaculatus* (Characiformes; Characidae) in the Tocantins river basin, amazon region, Brazil. *Rev Bras Parasitol Vet* 2019; 28(4): 739-743. http://dx.doi.org/10.1590/s1984-29612019041. PMid:31390431.

Silva RM, Tavares-Dias M, Dias MWR, Dias MKR, Marinho RGB. Parasitic fauna in hybrid tambacu from fish farms. *Pesq Agropec Bras* 2013; 48(8): 1049-1057. http://dx.doi.org/10.1590/S0100-204X2013000800034.

Parasitological assessment of hybrids Serrasalmidae

Simková A, Dávidová M, Papousek I, Vetesník L. Does interspecies hybridization affect the host specificity of parasites in cyprinid fish? *Parasit Vectors* 2013; 6(1): 95. http://dx.doi.org/10.1186/1756-3305-6-95. PMid:23587287.

Tavares-Dias M, Dias-Júnior MB, Florentino AC, Silva LM, Cunha AC. Distribution pattern of crustacean ectoparasites of freshwater fish from Brazil. *Braz J Vet Parasitol* 2015; 24(2): 136-147. http://dx.doi.org/10.1590/S1984-29612015036. PMid:26154954.

Tavares-Dias M, Oliveira MSB, Gonçalves RA, Neves LR. Parasitic diversity of a wild *Satanoperca jurupari* population, an ornamental cichlid in the Brazilian Amazon. *Acta Amaz* 2017; 47(2): 155-162. http://dx.doi.org/10.1590/1809-4392201602514.

Tavares-Dias M, Oliveira MSB, Gonçalves RA, Silva LMA. Ecology and seasonal variation of parasites in wild *Aequidens tetramerus,* a Cichlidae from the Amazon. *Acta Parasitol* 2014; 59(1): 158-164. http://dx.doi.org/10.2478/s11686-014-0225-3. PMid:24570063.

Thatcher VE. Amazon parasites. 2nd ed. Pensoft, Sofia: Moscow; 2006.

Videira M, Velasco M, Malcher CS, Santos P, Matos P, Matos E. An outbreak of myxozoan parasites in farmed freshwater fish *Colossoma macropomum* (Cuvier, 1818) (Characidae, Serrasalminae) in the Amazon region, Brazil. *Aquacult Rep* 2016; 3: 31-34. http://dx.doi.org/10.1016/j.aqrep.2015.11.004.

Whittington ID, Cribb BW, Hamwood TE, Halliday JA. Host-specificity of monogenean (Platyhelminth) parasites: a role for anterior adhesive areas? *Int J Parasitol* 2000; 30(3): 305-320. http://dx.doi.org/10.1016/S0020-7519(00)00006-0. PMid:10719124.