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Parasitic infections in pirarucu fry, *Arapaima gigas* Schinz, 1822 (Arapaimatidae) kept in a semi-intensive fish farm in Central Amazon, Brazil

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ABSTRACT

Studies regarding parasite fauna in farmed fish are of great relevance to the knowledge of the parasites species, allowing interference in their proliferation in order to avoid epizooties and consequently, economical losses. This study was designed to investigate the prevalence and intensity of parasites in fry *Arapaima gigas* maintained in ponds of a semi-intensive fish farm in Amazonas State, Brazil. On necropsy, 96.0% of *A. gigas* were found parasitized by *Dawestrema cycloancistrioides*, *Dawestrema cycloancistrium* (Monogenoidea), *Trichodina* sp., *Ichthyobodo* sp. (Protozoa), *Camallanus tridentatus*, *Terranova serrata*, *Goezia spinulosa* (Nematoda) and *Argulidae*. However, *D. cycloancistrium*, *D. cycloancistrioides* and *Trichodina fariai* were the parasites of greatest intensity. This study is the first to report parasitic infections in farmed *A. gigas* and the results indicated a high rate of infection that might lead to important changes in the health of the hosts.

Key words: Amazon, *Arapaima gigas*, culture, infections, parasites

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Introduction

The *Arapaima gigas,* known as pirarucu, is an Osteoglossiforme fish with double breathing, which is only found in South America, and inhabits the Amazon River drainage, the western Orinoco and the river systems of the Guianas (QUEIROZ and CRAMPTON, 1999). This fish can reach up to three meters in length and 200 kg in mass (QUEIROZ and CRAMPTON, 1999) and is a much appreciated species with great acceptance on the Brazilian Amazonian market, being regarded as a food fish of the highest quality. Hence, it is one of the most important species for the development and improvement of intensive aquaculture in the Amazonian region (ONO et al., 2003 and 2004). Despite its great importance to the Amazonian people, little is known about the parasitic infections of farmed *A. gigas*, because these studies have been carried out, in general, in wild fish.

The presence of 20 species of the parasites for pirarucu have been reported. Three parasites species are Monogenoideas, the *Dawestrema cycloancistrium* (Price and Nowlin, 1967), *D. cycloancistrioides* (Kritsky, Boeger and Thatcher, 1985) and *D. punctatum*, (Kritsky, Boeger and Thatcher, 1985). Six species are Nematoda, the *Goezia spinulosa* Diesing, 1939; *Philometra senticosa* Baylis, 1927; *Camallanus tridentatus* Drasche, 1884; *Gnatostoma gracilis* Diesing, 1838; *Rumai rumai* Travassos, 1960; *Terranova serrata* Drasche, 1884. Two species are Acantocephala, the *Polyacanthorhynchus macrorhynchus* Diesing, 1851 and *Polyacanthorhynchus rhopalorhynchus* Diesing, 1851. Two species are Cestoda, *Schizochoerus liguloides* Diesing, 1850 and *Nesolecithus janicki* Poche, 1922. Three species are Trematoda, the *Caballerotrema brasiliense* Prudhoe, 1960, *C. arapaimensee*, Thatcher, 1980 and *Himasthla piscicola* Stunkard, 1960. Other two species are Branchiura, the *Dolops discoidalis* Bouvier, 1899 and *Argulus* sp., while one other species is Copepoda, the *Ergasilus* sp. and another is Pentastomidae, the *Sebekia* sp. (BAYLIS, 1927; KRITSKY et al., 1985; THATCHER, 1980 and 2006; GOMES, 2006).

Modern fish farming with high stock fish densities and intensive production units provide ideal conditions for the invasion and persistence of a range of pathogens and parasites (MARTINS et al., 2002; PIAZZA et al., 2006; LEMOS et al., 2007). Infections by these disease-causing agents reduce the survival of fish, causing economical losses to farmers. Hence, fish susceptibility to parasites is a constant concern of farmers who need to decrease these problems and increase production. The increment of parasitic infections in the culture environment has also been associated with the low quality of water and inadequate management. All these environmental factors have been responsible for high infection by monogenean (72.9%), *Piscinoodinium pillulare* (43.2%), *Henneguya piaractus* (34.2%), *Ichthyophthirius multifiliis* (23.4%) and *Laernea cyprinacea* (11.9%) in fish cultivated in the Southeast region of Brazil (MARTINS et al., 2002). However, parasitic infection rates in farmed fish in the Brazilian Northern Region, especially for the *A. gigas*, are yet unknown.

Therefore, this paper provides information on the prevalence and intensity of parasites in farmed *A. gigas* fry in Central Amazonia. This contribution is needed for disease research and management programs that could serve as a tool to monitor and analyze the causes and trends in disease occurrences and epidemiology in Northern Brazil, mainly in the Amazonas State. The development of techniques for the control of parasites depends on the epidemiological and sanitary diagnosis of the culture establishments, so it may be possible to intervene in an efficient way on the process and consequently improve the quality of the fish produced.

Materials and methods

Fish and culture. A hundred and ten *A. gigas* fry (17.1 ± 4.5 cm of length) were collected monthly, from May to August 2006, from a semi-intensive fish farm in the municipality of Manacapuru, Amazonas State, Brazil, for parasitological analyses.

Parasitological analyses. After the fish were captured, their skin, fins, mouth and eyes were screened for the verification of macroscopic lesions. For parasitological exams of their stomachs and intestines, the organs were removed and placed in Petri plates containing distilled water and then examined. A single scratching was made between their caudal and lateral fins for the quantification of parasites in the tegument. All the gills were removed for the quantification of Monogenoidea. The methodology employed for the collection and fixation of the parasites was the one described by EIRAS et al. (2000). A field of the lamina of each fish and fresh collected material were used to estimate the quantity of trichodinids in the tegument and gills. For the quantification of Monogenea in the tegument, the total mucus collected was used. All the parasites were quantified with the help of a stereomicroscopic for the evaluation of the parasitic indices, according to the recommendations of BUSH et al. (1997). The parasite identification followed THATCHER's recommendation (2006).

Physical-chemistry parameters of the water. In each collection, from May to August 2006, the potential hydrogen (pH) was measured with digital equipment (WTW pH meter model D-812). The temperature, electric conductivity, dissolved oxygen (OD) and turbidity were also measured with digital equipment (YSI-55). Nitrate and nitrite concentrations in the culture ponds were determined (BOYD and TUCKER, 1992), as well as the total phosphorus (CLESCERI et al., 1998). The levels of total ammonia were determined by colorimetric methodology (VERDOUW et al., 1978) with absorbance readings in spectrophotometer (Amersham Pharmacia Biotech, model Novaspec II).

Statistical Analysis. The correlation between the total length of the fish and the quantity of Monogenoidea in its gills was tested through simple linear regression with non- logarithmized numbers and a confidence interval of 95% ($P<0.05$) was assumed.

Results

Variations in the physical and chemical parameters of the water from the culture ponds of *A. gigas* fry are presented in Table 1.

Table 1. Physical-chemical parameters of the water from the ponds of *A. gigas* fry

Parameters	Minimum-Maximum
Dissolved oxygen (mg/L)	4.0-6.0
pH	5.1-6.6
Temperature (^{o}C)	28.0-32.0
Conductivity (μS/cm)	7.3-29.5
Turbidity (μT)	5.6-86.9
Total phosphorus (mg/L)	0.40-0.78
Ammonia (mg/L)	0.10-0.16
Nitrite (mg/L)	0.001-0.003
Nitrate (mg/L)	0.08-0.17

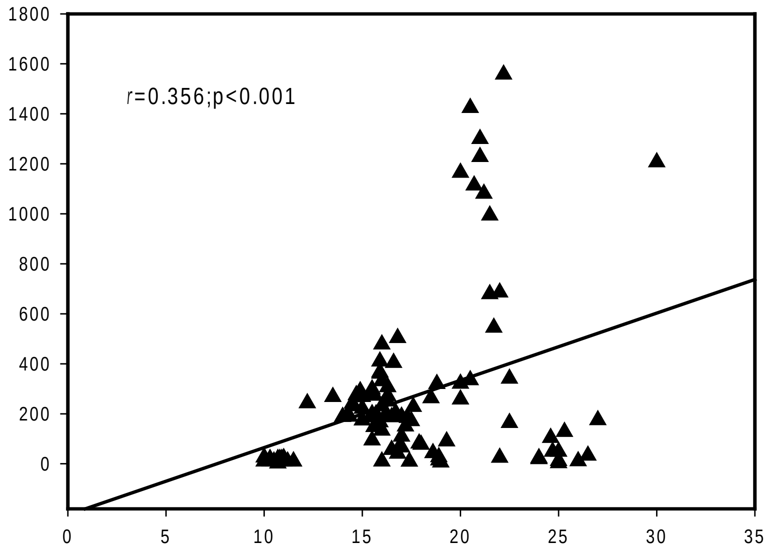

Fig. 1. Correlation between the number of Monogenoidea in the gills and the total length in cultivated *A. gigas* fry (n = 104)

A hundred and ten specimens of pirarucu *A. gigas* were examined. In the tegument, the greatest intensity and occurrence observed were of *Trichodina* sp. (Protozoa), followed by Monogenoidea *D. cycloancistrium*. However, in the gills there was greater intensity and occurrence of Monogenoidea *D. cycloancistrioides* and *D. cycloancistrium*, followed by *Trichodina* sp. (Table 2).

Table 2. Parasitic indices in examined *A. gigas* fry from a semi-intensive fish farm from the municipality of Manacapuru, Amazonas State, Brazil

Parasites	Examined Fish	Parasitized Fish	Prevalence (%)	Intensity of parasites	Range of parasites	Mean intensity
Skin						
Monogenoidea	110	40	36.4	253	1-20	n. d.
Trichodina sp.	110	76	69.0	2330	10-50	n. d.
Argulidae	110	01	0.9	n. d.	n. d.	n. d.
Ichthyobodo sp.	110	01	0.9	n. d.	n. d.	n. d.
Gills						
Monogenoidea	100	96	96.0	26906	1-1558	280
Trichodina sp.	110	43	39.1	640	10-50	n. d.
Argulidae	110	03	2.7	n. d.	n. d.	n. d.
Ichthyobodo sp.	110	21	19.1			
Intestinal tract						
Nematoda						
Terranova serrata	110	3	2.7	17	3-10	5,6
Camallanus tridentatus	110	5	4.5	23	2-10	4,6
Goezia spinulosa	110	6	5.4	8	3-4	1,3
Larvae type IV	110	2	1.8	9	3-6	4,5

Among the species of *A. gigas* parasites, the number of Monogenoidea in the gills was positively correlated to the total length of fish (Fig. 1) and this correlation can be described by the equation Length = 16.018 + (0.00470 * Monogenoidea).

Nematoda *Camallanus tridentatus* (Drashe, 1884), *Goezia spinulosa* (Diesing, 1939), *Terranova serrata* (Drasche, 1884) and type IV larvae (Table 2) were found in the intestines and/or stomachs of *A. gigas*.

Discussion

Even though in the ponds of cultivated *A. gigas* the physical and chemical parameters of the water were within the expected range of values for tropical species, several parasites were found. However, the increment of parasitic infections in artificial environments has been associated with low quality of water and inadequate management (MARTINS et al., 2002; SILVA-SOUZA et al., 2006; PIAZZA et al., 2006).

In semi-intensively cultivated pirarucu fry, there was a high prevalence of Monogenoidea of the *Dawestrema* and *Trichodina* gender, followed by *Argulidae*, *Ichthyobodo* sp. and Nematoda. Parasites were also reported in other fish species cultivated in Brazil (MARTINS et al., 2002; PIAZZA et al., 2006; LEMOS et al., 2007). However, in the skin of *A. gigas* the higher prevalence and intensity were of *Trichodina* sp., followed by Monogenoidea, while in the gills it was the opposite. Parasites with a direct life cycle, such as the Monogenoidea and the protozoan parasites, are more often found in sluggish environments, since these environments are favorable to their spread (PAVANELLI et al., 2004; SILVA-SOUZA et al., 2006). Therefore, the high prevalence and intensity of infection by Monogenoidea and *Trichodina* sp. protozoan are due to these parasites' predilection for this kind of environment.

Helminthes are a kind of parasite found in 70% of sluggish environments, since these environments are very favorable to the proliferation of parasites with a direct life cycle (PAVANELLI et al., 2004; TAKEMOTO et al., 2004). Monogenoidea are usually considered specific hosts parasites (TAKEMOTO et al., 2004; GUIDELLI et al., 2006; THATCHER, 2006). This specificity seems to guarantee high rates of parasitic infection because the parasites can invest more in achieving high rates of abundance and less in immune system evasion, besides having better mechanisms for meeting the host (GUIDELLI et al., 2006). Three species of Monogenoidea are known to parasite the *A. gigas* from natural environments, *D. cycloancistrum*, *D. cycloancistrioides* and *D. punctatum* (KRITSKY et al., 1985). However, in *A. gigas* from a semi-intensive fish farms only *D. cycloancistrioides* and *D. cycloancistrium* were found. In the tegument, only *D. cycloancistrium* was found, while in the gills *D. cycloancistrioides* and *D. cycloancistrium* were found. Possibly, this preference by epidermis suggests interspecific competition in the organization of these monogeneans communities. According to WHITTINGTON et al. (2000) monogeneans are parasites that, once on the host epidermis, live in its products, e.g. mucus. Some of these products are ``attractants'' and they may be an inhospitable surface because of their immunological activity. The chemical composition of the fish skin is known to be species-specific, and a preliminary analysis of the chemistry of some monogenean adhesives indicated they are novel proteins that display some differences between parasite families and species skin parasites, which are known to be species-specific.

In *A. gigas*, the level of gill infection by *D. cycloancistrioides* and *D. cycloancistrium* increased along with the body growth of the hosts. Similarly, other studies have described the correlation between the host length and its parasite level (SASAL et al., 1999; MARQUES and CABRAL, 2007). The parasite richness of specific monogeneans correlates positively with host body size and specific monogeneans are found on larger fish (SASAL et al., 1999). Thus, hosts with a long life expectancy, such as the *A. gigas*, may provide a more stable environment, not subject to sudden changes. In addition, the increase in host size seems to be linked with the increase in available niches for colonization.

In the Camallanidae family the intermediary hosts are not obligatory. Species from the Camallanidae and Anisakidae families can use several paratenic or transport hosts until they reach their definitive host and complete their development. Therefore, fish can be either definitive or intermediate hosts for these parasites. *G. spinulosa, T. serrata* and *C. tridentatus* were found in the stomachs and/or intestines of *A. gigas* from semi-intensive cultures. However, the prevalence and intensity of these parasites were smaller than the ones described for this same host in the natural environment (GOMES et al., 2006). In addition, *P. rarus*, another Nematoda parasite known to infect the *A. gigas,* was not observed here. Therefore, the results suggest either a low frequency or the absence of infectious forms in the culture when compared to the natural environment.

To conclude, this study is the first to report parasitic infections in farmed *A. gigas*. The results indicated a high prevalence and abundance of monogenean *D. cycloancistrioides* and *D. cycloancistrium* and protozoa *Trichodina* sp. which might lead to important changes in the health status of farmed fish and cause epizooties, resulting in economic losses for aquaculture. Higher parasitic infection may be favored by the hot and humid climate of the Amazonian region, as well as by aggregation behavior that is common in fry *A. gigas*. Therefore, the high stock density during fish rearing facilitates the quick spread of parasites and severe disease outbreaks can occur in the fish farms.

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SAŽETAK

Istraživanje parazitske faune na farmama riba od velikoga je značenja za nove spoznaje o parazitskim vrstama jer omogućuje upletanje u njihovo razmnožavanje s ciljem sprječavanja pojave epizootija i smanjenja posljedičnih gospodarskih šteta. Ovo istraživanje poduzeto je u svrhu određivanja prevalencije i intenziteta parazita u mlađi vrste *Arapaima gigas* držane na poluintenzivnim jezerskim farmama u državi Amazona u Brazilu. Pri razudbi je ustanovljeno da je 96,0% *A. gigas* bilo pozitivno na parazite *Dawestrema cycloancistrioides*, *Dawestrema cycloancistrium* (Monogenoidea), *Trichodina* sp., *Ichthyobodo* sp. (Protozoa), *Camallanus tridentatus*, *Terranova serrata*, *Goezia spinulosa* (Nematoda) i *Argulidae*. Ipak, najčešće dokazane vrste parazita bile su *D. cycloancistrium*, *D. cycloancistrioides* i *Trichodina fariai*. U istraživanju se prvi put izvješćuje o parazitima u farmski držane *A. gigas*, a rezultati govore o visokoj stopi invadiranosti što može dovesti do znatnih poremećaja zdravlja domaćina.

Ključne riječi: Amazona, *Arapaima gigas*, kultura, paraziti
