Lat. Am. J. Aquat. Res., 46(2): 377-382, 2018
DOI: 10.3856/vol46-issue2-fulltext-13

Research Article

Parasitism in gills of *Centropomus undecimalis* (Pisces, Centropomidae) from a protected area in São Luís, Maranhão, Brazil

Sildiane Martins Cantanhêde[1], Vívian Cristina Sodré Campos[1], Dayane Pestana Pereira[1] Almerinda Macieira Medeiros[1], Raimunda Nonata Fortes Carvalho Neta[2] Lígia Tchaicka[3] & Débora Martins Silva Santos[11]

[1]Laboratório de Morfofisiologia Animal, Departamento de Química e Biologia
Universidade Estadual do Maranhão, São Luís, MA, Brazil
[2]Laboratório de Biomarcadores e Organismos Aquáticos, Departamento de Química e Biologia
Universidade Estadual do Maranhão, São Luís, MA, Brazil
[3]Laboratório de Biodiversidade Molecular, Departamento de Química e Biologia
Universidade Estadual do Maranhão, São Luís, MA, Brazil
Corresponding author: Sildiane Martins Cantanhêde (sildymartins@hotmail.com)

ABSTRACT. Parasitism in fish can occur due to an imbalance in the environment-host-parasite relationship, usually associated with poor environmental quality. The present study aimed to determine the frequency of parasitism and histological alterations in the gills of *Centropomus undecimalis* from the Jansen Lagoon Ecological Park in São Luís, Maranhão, Brazil. Thirty-three specimens of *C. undecimalis* were collected. The first right gill arch was removed from each animal and fixed in 10% formalin for 24 h. These were then decalcified in 10% nitric acid, dehydrated in increasing concentrations of alcohol, clarified in xylene and impregnated with and embedded in paraffin. Five-μm thick sections were stained with hematoxylin and eosin for histologic description and to determine parasite frequency. The gill lesions of each fish were semi-quantitatively evaluated by calculating the histological alteration index (HAI). Structural alterations in the gills, such as epithelial hyperplasia and mucus, were associated with parasites in 48.4% of the fish. Three families of parasites from the Monogenean and Myxosporean groups were identified. The HAI average was 53.3, demonstrating that individuals collected from *C. undecimalis* had severe tissue modifications. Therefore, parasitism caused structural damages in the gill tissues of sampled fish, damages that can impair the organ's physiology, and harm the fish health.

Keywords: *Centropomus undecimalis***,** parasite, histopathology, snook, environment, pollution.

INTRODUCTION

Lesions in gills can be caused by chemical, physical and biological agents, ranging from environmental pollutants to various types of biological agents. These agents induce reactions of the body, such as circulatory disorders, proliferative, inflammatory and degenerative diseases, and congestion, telangiectasia, hemorrhages, and edemas, epithelial hyperplasia and mucous cells, inflammation and necrosis (Meyers & Hendricks, 1985; Roberts, 2001). Fish are susceptible to parasites, including various species groups which damage the gill tissue, such as *Ichthyophthirius multifiliis, Trichodina* sp., *Chilodonella* sp., *Henneguya* sp., *Dolops* sp., *Lernaea cyprinacea*, Monogeneans, Nematodes, and Ergasilidae (Schalch *et al*., 2006; Ueda *et al*., 2013). These parasites or symbionts are normally present in the environment or in the body of the fish, and when there is an imbalance in the environment-host-parasite relationship, outbreaks of disease occur (Martins *et al*., 2000, 2002). Fish diseases cause serious economic damage and often lead to high mortality rates. Monogenean parasites are notable etiological agents (Eiras *et al*., 2010). According to Adriano *et al*. (2005), these changes can partially impair the functions of the gills and consequently reduce respiratory capacity and ion exchange. Morphological parameters can be a useful tool for assessing the effects of environmental contamination on the quality of fish stocks. Among estuarine environment fish, the *Centropomus undecimalis* (Bloch, 1972) species is of great economic importance to the city of São Luís, Maranhão. It is

Corresponding editor: Enrique Dupré

commonly found in the Jansen Lagoon, a brackish water environment with a poor sewage infrastructure, and causes a reduction in the environmental quality of the lagoon, making the environment anoxic due to the high content of organic decaying matter, which has left the lagoon in a progressive state of eutrophication (Castro *et al*., 2002). In view of such impacts, the aim of the present study was to determine the frequency of parasitism and histological changes in the gills of *C. undecimalis* from the Jansen Lagoon Ecological Park in São Luís, Maranhão, Brazil.

MATERIALS AND METHODS

Fish collection

Based on selection criteria of economic importance and significance as a bioindicator, the *Centropomus undecimalis* (Bloch, 1972) (Perciformes, Centropomidae) species, popularly known as snook fish, which is abundant in estuarine environments, was chosen. The specimens were collected by artisanal fishing techniques using cast nets from March to June 2012 in the Jansen Lagoon Ecological Park, which is characterized as a depression formed by brackish water. Located in a metropolitan area on the coast, it communicates with the sea through a channel and is considered as an almost salt water lake (Fig. 1). The fish were stored in sterilized plastic bags and immersed in ice until they were euthanized. Thirty-three specimens were collected.

Histopathological analysis

The first right branchial arch was removed from each animal, and fixed in 10% formalin for 24 h. These were subsequently decalcified in 10% nitric acid, dehydrated in increasing concentration of alcohols, clarified in xylene and impregnated with and embedded in paraffin. Five-µm thick sections were stained with hematoxylin and eosin (Luna, 1968) for histological description and to determine parasite frequency. The parasites were identified to family level following the guidelines of Rohde (1993), Noga (1996) and Thatcher (2006).

Histological alterations were semi-quantitatively evaluated for each fish by calculating the histological alteration index (HAI), in accordance with Poleksic & Mitrovic-Tutundzic (1994), based on the severity of each lesion and classified in progressive stages of tissue damage using the formula $HAI = 1 \times \Sigma\ I + 10 \times \Sigma\ II + 100 \times \Sigma\ III$, where I: stage I alterations, II: stage II alterations, III: stage III alterations (Table 1).

RESULTS

The gill arch and gill filament structures of *C. undecimalis* are the same as those from the gills of other teleost fishes (Fig. 2a). The fish caught were apathetic with excessive mucus production on the body surface and in the gills. Of the fish collected, 50% had parasitic alterations associated with alterations in the gill structure (Fig. 2b).

The main lesions observed were epithelial hyperplasia with interlayer space filling, congestion, and necrosis. In the *C. undecimalis* specimens, excess mucus was observed on the gills and Myxosporean cysts were located in the central part of the filament. Several monogeneans were scattered among the lamellae; in such cases, there was epithelial hyperplasia and mucus (Figs. 3-4).

The histological alterations observed in the gills and associated with the parasites were classified according to the degree of severity and impairment of gill function, and are shown in Table 2. The lifting of the lamellar epithelium was observed in all the gills analyzed (100%), followed by lamellar epithelium hyperplasia (96.9%), excessive proliferation of the filament epithelium cells causing complete fusion of various lamellae (82%) and vascular congestion (60%). The presence of parasites was observed in 50% of the collected fish. Hyperplasia of the mucous cells was also observed, as well as lamellar epithelium disruption, uncontrolled thickening of the proliferative tissue and aneurysms of various sizes in the lamellae.

The histopathological alteration index (HAI) per individual ranged from 3 to 138. The average HAI value obtained was 53.3, demonstrating that the gills of the individuals of the *C. undecimalis* species collected exhibited severe modifications in the tissue.

DISCUSSION

The microscopic tests identified the presence of parasites on the gills of *C. undecimalis* specimens of the Monogenean group (Van Beneden, 1858) Monopisthocotylea. According to Rohde (1993), Monopisthocotylea is considered a subclass of the Monogenean group, capable of parasitizing the gills of teleost fish and infecting a variety of microenvironments. It can move quickly, feed on epithelial cells and has a haptor that forms a single fixing unit. The parasites of the Monopisthocotylea class observed are part of the Ancyrocephalinae (Bychowsky, 1937), Capsalidae (Baird, 1853) and Dactylogyroidea (Bychowsky, 1933) families.

Fish with monogenean and other gill parasite infections exhibit epithelial hyperplasia and mucous cells in response to this aggression, with the latter resulting in increased mucus production. This acts as a defense element thanks to its antimicrobial properties, through the action of lysozymes, antibodies and low

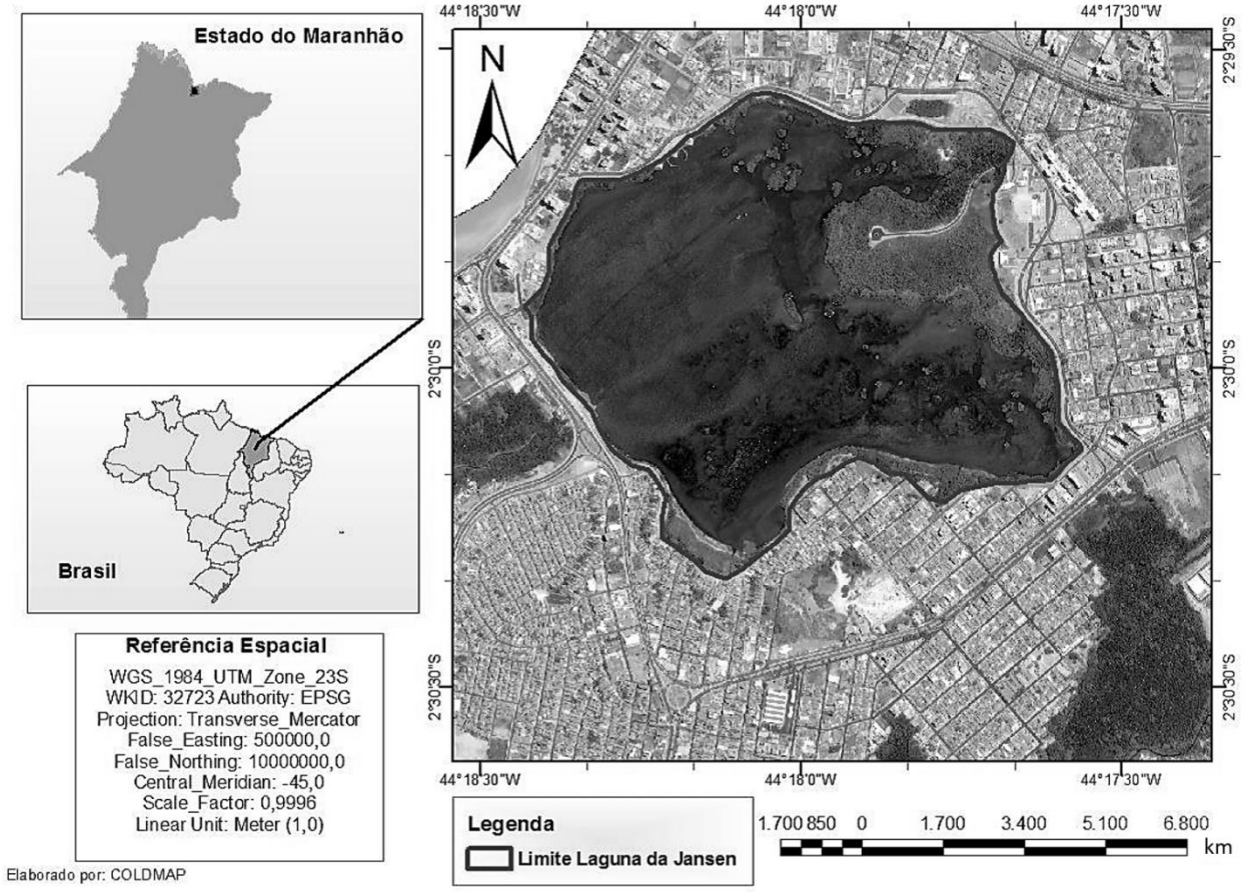

Figure 1. Jansen Lagoon Ecological Park, São Luís, Maranhão, Brazil.

Table 1. Categories of Histological Alteration Index (HAI) values.

HAI	Lesion
0-10	Without alterations
11-20	Mild alterations
21-50	Moderate alterations
51-100	Severe alterations
>100	Irreversible alterations

molecular weight fatty acids (Noga, 1996; Roberts, 2001). The increased mucus production, combined with cephalic gland secretions, causes a reduction in the respiratory capacity of the host (Thatcher & Brites-Neto, 1994).

These parasites can be found in all the fish organs, including the gills, causing serious structural damage (Takashima & Hibiya, 1995). Campos *et al*. (2011) found alterations caused by parasites in the gills of *Piaractus mesopotamicus* and *Prochilodus lineatus* from the Aquidauana River in the municipal region of Aquidauana, Mato Grosso do Sul. According to Moraes & Martins (2004), infections in fish caused by parasites

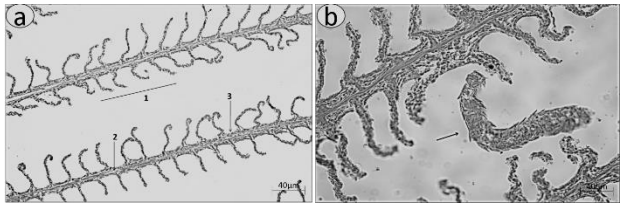

Figure 2. a) Normal gill filaments of *C. undecimalis*: 1. Secondary lamella, 2. Venous sinus, 3. Filament epithelium, b) monogenean parasite-arrow.

occur due to the parasite-host-environment imbalance generally associated with a poor quality environment. Carvalho (2010) also observed infection in the gills of *Geophagus brasiliensis* acquired from the Guandu River and *Trichiurus lepturus* from Guanabara Bay, both in Rio de Janeiro, by parasites from the Dactylogyridae and Ancyrocephalinae families.

Given that the gill respiratory epithelium is in direct contact with the external environment and physiological stress-inducing factors, and has a large surface area, poor quality of water (Mallatt, 1985; Hinton & Lauren, 1990), irritating toxic agents, infectious agents

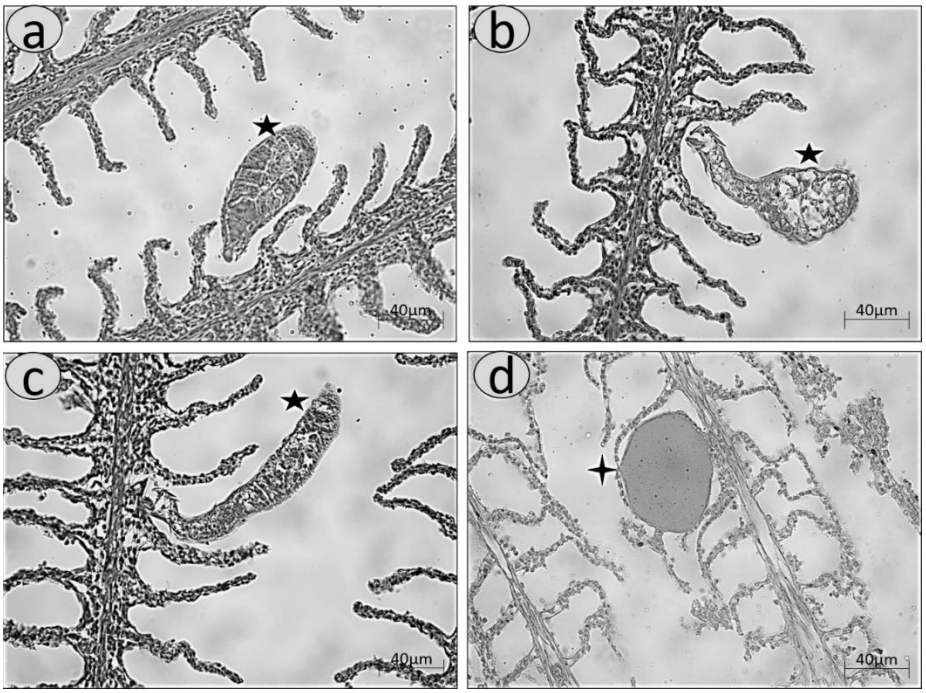

Figure 3. Gill filaments of *C. undecimalis* parasitized by monogeneans (a, b, c-stars), d) myxosporean cyst.

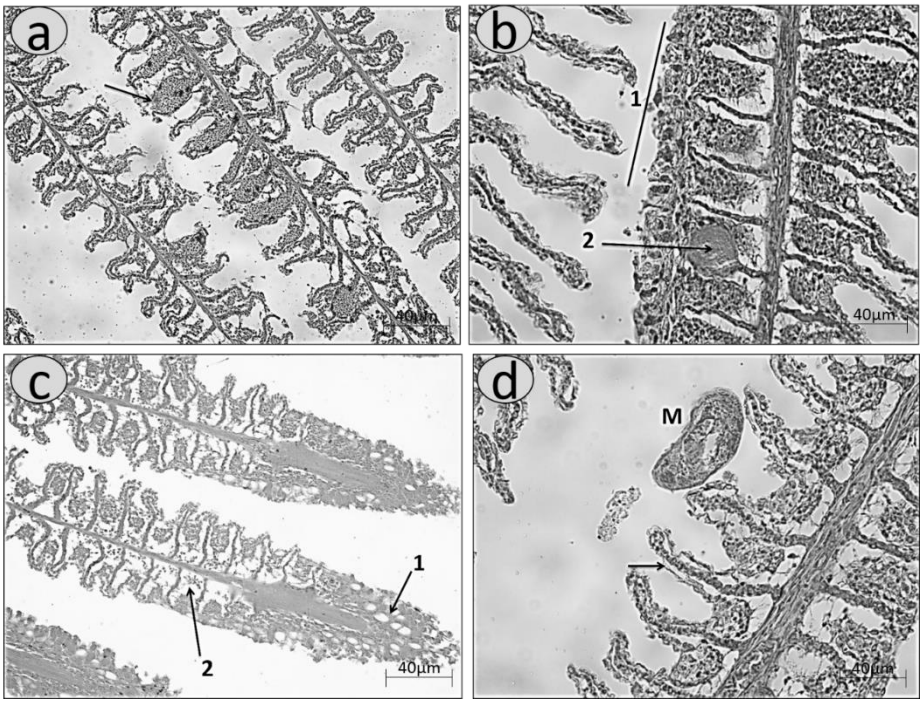

Figure 4. Histological changes in *C. undecimalis* gills. a) Arrow-aneurysm, b) 1-total fusion of secondary lamellae caused by hyperplasia of the epithelial cells, 2-aneurysm, c) 1-mucus cells; 2-uncontrolled thickening of the filament and lamellae, d) arrow-lifting of the lamellar epithelium; M: Myxosporidia.

and the presence of parasites (Pavanelli *et al*., 2004) lead to functional impairment of the filaments and gill lamellae.

The most frequent lesions found in the gills of *C. undecimalis* were the lifting of the lamellar epithelium, lamellar epithelium hyperplasia, and lamellar fusion. Gills fusion resulted from epithelial cell hyperplasia.

Table 2. Classification of histological alterations of *Centropomus undecimalis* gills collected from the Jansen Lagoon Ecological Park, São Luís, Maranhão, Brazil. *Progressive stages of tissue damages: I: mild to moderate alteration, II: moderate to severe alteration, III: irreparable alteration.

Gill histological changes	Stage*
Hyperplasia/hypertrophy of the lamellar epithelium	
Lifting of the lamellar epithelium	
Disorganization of lamellae	
Incomplete fusion of several lamellae	I
Complete fusion of several lamellae	
Vascular congestion	
Dilation	
Presence of parasites	
Rupture of the lamellar epithelium	
Hyperplasia and hypertrophy of mucous cells	II
Uncontrolled thickening of the filament	
Lamellar proliferative tissue	
Lamellar aneurysm	III

The degree of fusion depends on the intensity and location of the hyperplasia. The fusion will be partial if the hyperplasia is restricted to the filament base or only the lamellae portion. However, if the hyperplasia is present throughout the filaments, the fusion will be total (Meletti *et al*., 2003). The lifting of the lamellar epithelium, the proliferation of filament epithelium and the fusion of the lamellae observed in large quantities in the gills of *C. undecimalis* function as a defense mechanism as they reduce the surface area of the gills and/or increase the diffusion barrier to pollutants (Erkmen & Kolankaya, 2000), impairing oxygen absorption.

Regarding the filament and lamellar proliferative tissue thickening, stage II alterations were more frequent and were present in 48.4% of the gills analyzed. Stage II alterations are more severe and impair gill function. When water quality improves these lesions can be repairable, but if the level of pollution increases, they can progress to the stage III (Poleksic & Mitrovic-Tutundzic, 1994).

The gill lamellae of the *C. undecimalis* collected also exhibited stage II vascular changes, with an overall parasitism rate of 39.3% and 70% of lamellae with aneurysms. Aneurysms generally result from the system collapse of the pillar cells, which impairs vascular integrity by releasing large amounts of blood, pushing the lamellar epithelium outward (Hinton & Lauren, 1990). They may be associated with chemical contaminants or parasitic lesions. Stage III alterations do not allow the restoration of the gill structure, even in cases of water quality improvement.

In fish with few parasites, basal and mucous cell hyperplasia is mild to moderate, whether associated or not with the congestive process. In severe infections, there is marked basal and mucous cell hyperplasia and increased mucus production, associated with circulatory disorders such as congestion, telangiectasia and interstitial hemorrhage (Schalch *et al*., 2006), signs similar to those observed in the present study.

By impairing blood flow and increasing the distance between the water and the blood, the lesions observed in the gill tissue impair the primary function of the gills, namely the gas exchange process, in addition to negatively influencing the maintenance of the acid-base balance and fish osmosis.

The results show that parasitism causes structural damage to the gill tissue. This damage can impair the physiology of the organ, and therefore harm the health of the fish. The data obtained in the present study serve as a warning to the riverine population, as *C. undecimalis* is sold locally.

ACKNOWLEDGMENTS

The authors would like to thank the Foundation for Research and Scientific and Technological Development of Maranhão (FAPEMA), for the opportunity to carry out this study, with the support of the Scientific Initiation Scholarship Program.

REFERENCES

Adriano, E.A., S. Arana & N.S. Cordeiro. 2005. Histology, ultrastructure and prevalence of *Henneguya piaractus* (Myxosporea) infecting the gills of *Piaractus mesopotamicus* (Characidae) cultivated in Brazil. Dis. Aquat. Org., 64(3): 229-235.

Campos, C.M., J.R.E. Moraes & F.R. Moraes. 2011. Histopathology of gills of *Piaractus mesopotamicus* (Holmberg, 1887) and *Prochilodus lineatus* (Valenciennes, 1836) infested by monogenean and myxosporea, caught in the Aquidauana River, State of Mato Grosso do Sul, Brazil. Rev. Bras. Parasitol. Vet., 20(1): 67-70.

Carvalho, A.R. 2010. Taxonomia dos monogenéticos (Platyhelminthes) e variação temporal da biodiversidade, abundância numérica e biomassa dos metazoários parasitos de *Geophagus brasiliensis* (Cichlidae) e de *Trichiurus lepturus* (Trichiuridae) no Rio de Janeiro, Brasil. Tese Doutorado, Universidade Federal Rural do Rio de Janeiro, Rio de Janeiro, 146 pp.

Castro, A.C.L., N.M. Piorski & J. de R. Pinheiro-Júnior. 2002. Avaliação qualitativa da ictiofauna da Lagoa da Jansen, São Luís, MA. Bol. Laborat. Hidrobiol., 15(4): 39-50.

Eiras, J.C., R.M. Takemoto & G.C. Pavanelli. 2010. Diversidade dos parasitas de peixes de água doce do Brasil. Clichetec, Maringá, 333 pp.

Erkmen, B. & D. Kolankaya. 2000. Effects of water quality on epithelial morphology in the gill of *Capoeta tinca* living in two tributaries of Kizilirmak River, Turkey. Bull. Environ. Contam. Toxicol., 64: 418-425.

Hinton, D.E. & D.J. Lauren. 1990. Integrative histopathological approaches to detecting effects of environmental stressors on fishes. In: S.M. Adams (ed.). Biological indicators of stress in fish. American Fisheries Society, Bethesda, pp. 51-56.

Luna, L.G. 1968. Manual of histologic staining methods of the Armed Forces Institute of Pathology. McGraw-Hill Book Company, New York, 258 pp.

Mallatt, J. 1985. Fish gill structural changes induced by toxicants and other irritants: a statistical review. Can. J. Fish. Aquat. Sci., 42(4): 630-648.

Martins, M.L., F.R. Moraes, R.Y. Fujimoto, E.M. Onaka, S.H.C. Schalch & E.D. Silva. 2000. Parasitic infections in cultivated Brazilian freshwater fishes. A survey of diagnosticated cases. Rev. Bras. Parasitol. Vet., 9(1): 23-28.

Martins, M.L., E.M. Onaka, F.R. Moraes, F.R. Bozzo, A.M.F. Paiva & G. Adriano. 2002. Recent studies on parasitic infections of freshwater cultivated fish in the State of São Paulo, Brazil. Acta Scient., 24(4): 981-985.

Meletti, P.C., O. Rocha & C.B.R. Martinez. 2003. Avaliação da degradação ambiental na bacia do Rio Mogi-Guaçu por meio de testes de toxidade com sedimento e de análises histopatológicas em peixes. In: J. Brigante & E.L.G. Espíndola (eds.). Limnologia fluvial: um estudo no Rio Mogi-Guaçú. RiMA Artes e Texos, São Carlos, São Paulo, pp. 149-180.

Meyers, T.R. & J.D. Hendricks. 1985. Histopathology. In: G.M. Rang & S.R. Petrocelli (eds.). Fundamentals of aquatic toxicology methods and applications. Hemisphere Publishing Corp., Washington, pp. 283-331.

Moraes, F.R. & M.L. Martins. 2004. Condições predisponentes e principais enfermidades de teleósteos em piscicultura intensiva. In: J.E.P. Cyrino (ed.). Tópicos especiais em piscicultura de água doce tropical intensiva. TecArt, São Paulo, pp. 343-386.

Noga, E.J. 1996. Fish disease: diagnosis and treatment. Mosby, St. Louis, 367 pp.

Pavanelli, G.C., M.H. Machado, R.M. Takemoto, G.M. Guidelli & M.A.P. Lizama. 2004. Helminth fauna of the fishes: diversity and ecological aspects. In: S.M. Thomaz, A.A. Agostinho & N.S. Hahn (eds.). The upper Paraná River and its floodplain: physical aspects, ecology, and conservation. Backhuys Publishers, Leiden, pp. 309-329.

Poleksic, V. & V. Mitrovic-Tutundzic. 1994. Fish gills as a monitor of sub-lethal and chronic effects of pollution. In: R. Muller & R. Lloyd (eds.). Sub-lethal and chronic effects of freshwater fish. Cambridge University, Cambridge, pp. 339-352.

Roberts, R.J. 2001. Fish pathology. Saunders, London, 472 pp.

Rohde, K. 1993. Ecology of marine parasites: an introduction to marine parasitology. CAB International, Wallingford, 298 pp.

Schalch, S.H.C., F.R. Moraes & J.R.E. Moraes. 2006. Efeitos do parasitismo sobre a estrutura branquial de *Leporinus macrocephalus* Garavello and Britsk, 1988 (Anastomidae) e *Piaractus mesopotamicus* Holmberg, 1987 (Osteichthyes: Characidae). Rev. Bras. Parasitol. Vet., 15(3): 110-115.

Takashima, F. & T. Hibiya. 1995. An atlas of fish histology: normal and pathological features. Kodansha, Tokyo, 195 pp.

Thatcher, V.E. 2006. Amazon fish parasites. Pensoft Publishers, Moscow, 508 pp.

Thatcher, V.E. & J. Brites-Neto. 1994. Diagnosis, prevention, and treatment of diseases of Neotropical freshwater fishes. Rev. Bras. Med. Vet., 16(3): 111-128.

Ueda, B.H., L.C. Karling, R.M. Takemoto & G.C. Pavanelli. 2013. Parasites of the freshwater fish trade in Brazil: science metric study. Pesq. Vet. Bras., 33(7): 851-854.

Received: 16 January 2017; Accepted: 28 November 2017