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Hematological parameters of *Pimelodus maculatus* (Osteichthyes: Pimelodidae) from polluted and non-polluted sites in the Itajaí-Açu river, Santa Catarina State, Brazil

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ABSTRACT. This study evaluated the hematological response of *Pimelodus maculatus* captured in two environments with different levels of pollution in the Itajaí-Açu river, Santa Catarina State, southern Brazil. One of them, hereby named reference site, is a water captation site that supplies the city of Blumenau, in which the riparian forest is preserved and there is no sewage discharge. The other, denominated polluted site, is characterized by discharge of domestic sewage. After water quality analysis, fish were captured, transported to the laboratory and anesthetized for the hematological exam. In the polluted site, the most probable number of fecal coliforms and water transparency were respectively higher and lower than that observed at the reference site. The main results were an increased percentage of hematocrit and reduced numbers of lymphocytes and monocytes in the differential counting in fish from the polluted site. Red blood cells, total white blood cells and total counting of thrombocytes did not vary between environments. The number of neutrophils remained at high levels in fish from both environments. This study demonstrated that water quality might affect the hematological parameters in free-living fish.

Key words: siluriform fish, hematology, sewage, water quality.

RESUMO. Parâmetros hematológicos de *Pimelodus maculatus* (Osteichthyes: Pimelodidae) em local poluído e não poluído no rio Itajaí-Açu, Estado de Santa Catarina, Brasil. Este estudo avaliou a resposta hematológica de *Pimelodus maculatus* capturado em dois ambientes com diferentes níveis de poluição no rio Itajaí-Açu, Estado de Santa Catarina, Sul do Brasil. Um deles é um local de captação de água para abastecimento da cidade de Blumenau, onde a mata ciliar é preservada e não há descarga de esgoto denominado de local referência. O outro é caracterizado por descarga de esgoto doméstico denominado local poluído. Após a análise da qualidade da água, os peixes foram capturados, transportados para o laboratório e anestesiados para o exame hematológico. No local poluído, o número mais provável de coliformes fecais foi maior e a transparência da água menor do que o local sem a descarga de esgoto. Os principais resultados foram o aumento no percentual de hematócrito e a redução no número de linfócitos e monócitos na contagem diferencial de leucócitos nos peixes do local que recebia descarga de esgoto. Os números totais de eritrócitos, leucócitos e trombócitos não foram diferentes entre os locais. O número de neutrófilos foi mantido alto nos peixes dos dois ambientes. Este estudo demonstrou que a qualidade da água influencia os parâmetros hematológicos de peixes no ambiente natural.

Palavras-chave: peixe siluriforme, hematologia, esgoto, qualidade de água.

Introduction

The teleost fish *Pimelodus maculatus* Lacépède 1803, commonly known as **yellow mandi**, is a widespread species in South American rivers. It is an important commercial fish with great acceptance by consumers (LUZ; ZANIBONI-FILHO, 2002). It has been frequently found in the Itajaí-Açu river, located in Blumenau, Santa Catarina State, Brazil. Therefore, it is considered the main nourishment source by local inhabitants. Despite the pollution found in the river, fishing is a traditional activity (BACHMANN et al., 2007).

The waters of rivers and streams are characterized by interactions among environmental factors such as soil type, size, composition of

vegetation and the type and degree of human interference (KRUPEK et al., 2008). These variables must be considered and require studies to improve the understanding of its structure and dynamics, rather than human action decreases irreversibly this interaction (SMITH; PETRERE JR., 2000), as well to investigate the impact on fish health.

Hematological assessment in reared and wild fish is an important tool to evaluate fish health. They can be induced by the presence of pollutants and factors such as temperature, salinity, pH, dissolved oxygen concentration, carbon dioxide and inadequate management (RANZANI-PAIVA; SILVA-SOUZA, 2004). Consequently, the assessment of physiological parameters such as cortisol, glycemia, ion concentration, red blood cells (RBC), hemoglobin and hematocrit offer responses to toxicant stress (LUSKOVÁ, 1998; OMOREGIE, 1998). In coregonid fish, *Coregonus lavaretus*, Moiseenko (1998) reported an increased response of white blood cells (WBC) and inferred that the response depends on environment pollution. Similarly, Saravanan et al. (2003) observed increased RBC and WBC contents in fish from polluted environments. Thus, the number of RBC and WBC may vary according to environmental influence. Some authors have reported a decrease in RBC numbers and hematocrit in infected fish (REHULKA, 2002; MARTINS et al., 2004b) and in fish exposed to pollutants (SILVEIRA-COFFIGNY et al., 2004; SIMONATO et al., 2007), while an increase in the number of leucocytes and neutrophils have been reported in parasitized fish (SOPINSKA, 1985; SILVA-SOUZA et al., 2000; GHIRALDELLI et al., 2006b).

When compared to other fish species, little information is known concerning hematological parameters in siluriform fish. Some investigators have focused their works on reared *Rhamdia quelen* (TAVARES-DIAS et al., 2002; BORGES et al., 2004) and *Pseudoplatystoma fasciatum* (RANZANI-PAIVA et al., 2005). In the Paraná river, Ranzani-Paiva et al. (2000) evaluated the hematology of *Pseudoplatystoma corruscans*, *Loricariichthys platymetopon*, *Pimelodela gracilis*, *Pterodoras granulosus*, *Parauchenipterus galeatus*, *Hypostomus* aff. *derbyi*, *Pinirampus pirinampu* and *Pimelodus maculatus*. Differential counting of leucocytes in circulating blood of *P. maculatus* revealed lymphocytes as the most frequent cells (SILVA-SOUZA, 2000).

From the fact that nothing is known on the hematology of fish from Itajaí-Açu river in southern Brazil, this study evaluated blood parameters in *P. maculatus*, an important fish for local population, captured at two sites.

Material and methods

In the period from August to September 2006, 34 specimens of *P. maculatus* were captured with line and hook at two sites with different organic pollution levels in the Itajaí-Açu river, Blumenau, Santa Catarina State, Brazil. Reference site is a water captation site (26º52'57.4"; W 49º5'48.5") of the Water Treatment Station II (SAMAE), which supplies the city of Blumenau. At this site, the preservation of the riparian forest can be observed, as well as the absence of organic pollution, lack of urbanization and low water flow. The other collection point is the polluted site (S 26º55'16.2"; W 49º3'43.7"), characterized by drainage of domestic sewage, industry discharge and the practice of sport fishing (Figure 1). The urbanization around this site is responsible for important changes on the river edges, such as the lack of riparian vegetation, as previous report by Bachmann et al. (2007).

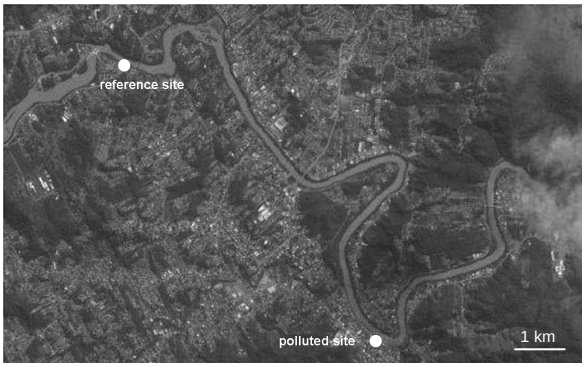

Figure 1. Geographic location of the collection sites in the Itajaí-Açu river, Blumenau, Santa Catarina State.

After capture, fish were transported to the Parasitology Laboratory of the Blumenau Regional University (FURB) for hematological assessment. Seventeen fish from the unpolluted environment showed averages of 48.3 ± 12.7 g in weight and 196.5 ± 12.9 mm in length. The fish from the polluted site showed 69.5 ± 34.2 g in weight and 206.9 ± 28.3 mm in length. During the days of each sampling, the water temperature, dissolved oxygen, pH and transparency were measured. The most probable number (MPN) of fecal coliforms was determined using the American Public Health Association (APHA, 1980) method.

Fish were anesthetized with benzocaine solution (50 mg L^{-1}) and the blood was withdrawn from the caudal vein into a syringe containing a drop of EDTA (10%) solution (Ethics Committee for Animal Experiments – FURB no. 015/2004). The blood was utilized to measure hematocrit (GOLDENFARB et al., 1971); red blood cell (RBC) count using a hemocytometer, total white blood

cells (WBC) and thrombocyte counts by indirect method (MARTINS et al., 2004a), and differential WBC counting by using the combination of May-Grünwald/Giemsa (ROSENFELD, 1947) staining, in which a hundred cells were counted for establishing of each cell's contents.

Data were submitted to one-way analysis of variance (Anova). Differences between means were tested by Tukey test ($p < 0.05$) (ZAR, 1996).

Results and discussion

The water quality, analyzed in both environments, revealed the lowest transparency in the water from the polluted environment. As a result, the most probable number of fecal coliforms was higher in the polluted site than at the reference site (Table 1), confirming the observations on the sewage discharge in this area. Dissolved oxygen and pH were similar in both environments. In the unpolluted environment, the water temperature was slightly higher when compared to the polluted one.

Table 1. Mean and standard deviation of water quality in two environments studied at Itajaí-Açu river, Santa Catarina State, Brazil.

Parameters	Reference Site	Polluted site
Fecal coliforms (MPN 100^{-1})	0	5,000
Temperature (°C)	18.0 ± 2.83	14.6 ± 2.26
pH	6.95 ± 0.06	7.14 ± 0.01
Dissolved oxygen (mg L^{-1})	6.35 ± 0.75	6.75 ± 0.28
Transparency (cm)	33.82 ± 2.32	6.32 ± 0.01

Hematocrit percentage (Table 2) was greater in fish from the polluted site. The other parameters did not vary. In fish from the reference site, hematocrit percentage was similar to that related in *R. quelen* (TAVARES-DIAS et al., 2002) and *P. fasciatum* (RANZANI-PAIVA et al., 2005). Surprisingly, the hematocrit in fish from the polluted site was similar to that related for *P. maculatus* in the Paraná river (RANZANI-PAIVA et al., 2000). It could also be observed that hematocrit may present changed behavior depending on the environment, fish management, fish health status and stress (PILARCZYK, 1986; REHULKA, 2002; MARTINS et al., 2004a). In *R. quelen,* hematocrit decreased in fish exposed to chronic stress for 10 days in relation to basal values, as observed by Barcellos et al. (2004). According to Pedro et al. (2005), red blood cells, released into blood, are common in fish after their activity. In *Ictalurus punctatus*, Kirk (1974) found an increase in hematocrit as a result of oxygen deficiency, such as in the present study. Supporting this view, the increase in hematocrit of fish from the polluted site, observed in this study, may be related to a high respiratory demand. Consequently, fish exposed to sewage water are more susceptible to environmental changes.

Table 2. Mean, standard deviation and range (in parentheses) of the hematological parameters in *Pimelodus maculatus* from Itajaí-Açu river, Santa Catarina State, Brazil. Different letters in the rows indicate significant difference between the environments ($p < 0.05$).

Parameters	Reference site	Polluted site
Hematocrit (%)	29.88 ± 4.64 a	37.35 ± 5.79 b
	(19 - 36)	(29 - 50)
Red blood cells (x10^6 μL^{-1})	2.35 ± 0.82 a	2.19 ± 0.53 a
	(1.12 - 3.73)	(1.60 - 3.40)
Thrombocytes (x10^3 μL^{-1})	44.18 ± 28.25 a	33.63 ± 24.65 a
	(4.22 - 89.52)	(3.89 - 79.73)
White blood cells (x10^3 μL^{-1})	42.65 ± 25.56 a	33.02 ± 19.48 a
	(16.3 - 113.16)	(9.15 - 51.80)

In this study, the WBC count was lower than the one presented in pimelodid *R. quelen* by Borges et al. (2004). It must be pointed out that, in the Borges et al. (2004) assay, fish were captured in ponds, and in this study they were captured in a river. On the other hand, Saravanan et al. (2003) observed a high number of WBC in fish from a polluted river, which means an opposite answer to this trial. Possibly, *P. maculatus* is well adapted and/or is more resistant to water quality changes.

In the differential WBC count, lymphocytes and neutrophils predominated, followed by monocytes and, in low number, come eosinophils, basophils and special granulocytic cells (Table 3). Predominance of lymphocytes and neutrophils in fish from the unpolluted environment was in agreement with that reported for siluriform fish (TAVARES-DIAS et al., 2002; BORGES et al., 2004) in normal environmental conditions.

Table 3. Mean, standard deviation and range (in parentheses) of the differential count of leucocytes in *Pimelodus maculatus* from the Itajaí-Açu River, Santa Catarina State, Brazil. Different letters in the rows indicate significant difference between the environments ($p < 0.05$). SGC: special granulocytic cell.

Cells	Reference site	Polluted site
Lymphocytes (x10^3 μL^{-1})	14.56 ± 11.65 a	9.40 ± 3.95 b
	(4.13 - 48.66)	(3.02 - 15.54)
Monocytes (x10^3 μL^{-1})	9.73 ± 9.60 a	2.19 ± 1.50 b
	(0.63 - 26.55)	(0.56 - 4.77)
Neutrophils (x10^3 μL^{-1})	16.47 ± 9.31 a	19.49 ± 15.73 a
	(1.63 - 35.08)	(4.58 - 56.35)
Eosinophils (x10^3 μL^{-1})	0.60 ± 1.30 a	0.24 ± 0.52 a
	(0 - 4.53)	(0 - 2.07)
Basophils (x10^3 μL^{-1})	0.28 ± 0.57 a	0.55 ± 0.72 a
	(0 - 2.26)	(0 - 2.82)
SGC (x10^3 μL^{-1})	0.90 ± 1.32 a	1.15 ± 0.98 a
	(0 - 4.72)	(0 - 3.13)

The percentage of lymphocytes and monocytes registered a significant decrease ($p < 0.05$) in fish from the polluted environment. In this study, lymphocytes

and neutrophils were the most frequent cells, and these answers corroborate the results of Ranzani-Paiva et al. (2000), Borges et al. (2004), Azevedo et al. (2006) and Ghiraldelli et al. (2006a). In the polluted site, the number of lymphocytes in *P. maculatus* blood decreased significantly. According to Silva-Souza et al. (2000), who studied the same fish in Paraná State, the number of lymphocytes was reduced in high-temperature water. Ghiraldelli et al. (2006a) observed a high number of lymphocytes in tilapia who had been fed fish entrails, cooked rice, homemade ration and restaurant scraps. However, this study registered different responses when compared to the one above, since the lymphocytes number decreased. This trend may be related to fish that live in the polluted site as it, in fact, occurred in the polluted area. This fact is supported by Iwama and Nakanishi (1996), who reported its presence in the inflammatory process and cell mediated response. Similarly, Hoeger et al. (2004) showed that rainbow trout exposed to sewage effluent had a decrease in the circulating lymphocytes, such as in the present study. Contrary to these observations, Ghiraldelli et al. (2006b) related high numbers of lymphocytes in fish fed with pig dung.

In the studied areas, neutrophils were in large number without significant difference, not only in fish captured from the reference site, but also in those from polluted ones. The results of this study confirm findings by Hoeger et al. (2004) with fish exposed to sewage effluent. This cell is very suitable to environmental changes (RANZANI-PAIVA; SILVA-SOUZA, 2004; MARTINS et al., 2004b), but it is important to emphasize the fact that fish from the polluted site show a great number of neutrophils. In fact, this result proves its proliferation in blood under adverse conditions, such as infection, inflammation and stress (ELLIS, 1977; MARTINS et al., 2004a).

To conclude, increased hematocrit, low numbers of lymphocytes and monocytes, as well as the neutrophil number, kept on high levels, were the most important changes observed in fish exposed to polluted water in the Itajaí-Açu river, Santa Catarina State. Therefore, this demonstrates that these hematological parameters are good physiological indicators of the poor water quality for *P. maculatus*.

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References

APHA-American Public Health Association. **Standard methods for the examination of water and wastewater**. 15th ed. Washington, D.C.: APHA, 1980.

AZEVEDO, T. M. P.; MARTINS, M. L.; YAMASHITA, M. M.; FRANCISCO, C. J. Haematology of *Oreochromis niloticus*: comparison between fish maintained in a fish farm associated with pigs and fee fishing from Valley of Tijucas River, Santa Catarina, Brazil. **Boletim do Instituto de Pesca**, v. 32, n. 1, p. 41-49, 2006.

BACHMANN, F.; GREINERT, J. A.; BERTELLI, P. W.; SILVA-FILHO, H. H.; LARA, N. O. T.; GHIRALDELLI, L.; MARTINS, M. L. Parasitic fauna of *Pimelodus maculatus* (Osteichthyes: Pimelodidae) from the Itajaí-Açu river in Blumenau, SC, Brazil. **Acta Scientiarum. Biological Sciences**, v. 29, n. 1, p. 109-114, 2007.

BARCELLOS, L. J. G; KREUTZ, L. C.; SOUZA, C.; RODRIGUES, L. B.; FIOREZE, I.; QUEVEDO, R. M.; CERICATO, L.; SOSO, A. B.; FAGUNDES, M.; CONRAD, J.; LACERDA, L. A.; TERRA, S. Hematological changes in jundiá (*Rhamdia quelen* Quoy and Gaimard Pimelodidae) after acute and chronic stress caused by usual aquacultural management, with emphasis on immunosuppressive effects. **Aquaculture**, v. 237, n. 1-4, p. 229-236, 2004.

BORGES, A.; SCOTTI, L. V.; SIQUEIRA, D. R.; JURINITZ, D. F.; WASSERMANN, G. F. Hematologic and serum biochemical values for jundiá (*Rhamdia quelen*). **Fish Physiology and Biochemistry**, v. 30, n. 1, p. 21-25, 2004.

ELLIS, A. E. The leucocytes of fish: a review. **Journal of Fish Biology**, v. 11, n. 5, p. 453-491, 1977.

GHIRALDELLI, L.; MARTINS, M. L.; YAMASHITA, M. M.; JERÔNIMO, G. T. Haematology of *Oreochromis niloticus* (Cichlidae) and *Cyprinus carpio* (Cyprinidae) maintained in different conditions of handling and feeding from the State of Santa Catarina, Brazil. **Acta Scientiarum. Biological Sciences**, v. 28, n. 4, p. 319-325, 2006a.

GHIRALDELLI, L.; MARTINS, M. L.; YAMASHITA, M. M.; JERÔNIMO, G. T. Ectoparasites influence on the haematological parameters of Nile tilapia and carp cultured in the State of Santa Catarina, South Brazil**. Journal of Fisheries and Aquatic Sciences**, v. 1, n. 3, p. 270-276, 2006b.

GOLDENFARB, P. B.; BOWYER, F. P.; HALL, E.; BROSIOUS, E. Reproducibility in the hematology laboratory: the microhematocrit determination. **American Journal of Clinical Pathology**, v. 56, n. 1, p. 35-39, 1971.

HOEGER, B.; HEUVEL, M. R. Van Den; HITZFELD, B. C.; DIETRICH, D. R. Effects of treated sewage effluent on immune function in rainbow trout (*Oncorhynchus mykiss*). **Aquatic Toxicology**, v. 70, n. 4, p. 345-355, 2004.

IWAMA, G.; NAKANISHI, T. **The fish immune system**. London: Academic Press, 1996.

KIRK, W. L. The effects of hypoxia on certain blood and tissue electrolytes of channel catfish, *Ictalurus punctatus* (Rafinesque). **Transactions of the American Fisheries Society**, v. 103, n. 3, p. 593-600, 1974.

KRUPEK, R. A.; BRANCO, C. C. Z.; PERES, C. K. Seasonal variation of some physical and chemical parameters in three rivers in one drainage basin in central-south region of Paraná State. **Acta Scientiarum. Biological Sciences**, v. 30, n. 4, p. 431-438, 2008.

LUSKOVÁ, V. Factors affecting haematological indices in free-living fish populations. **Acta Veterinaria**, v. 67, n. 4, p. 249-255, 1998.

LUZ, R. K.; ZANIBONI-FILHO, E. Larvicultura de mandi-amarelo *Pimelodus maculatus* Lacépède, 1803 (Siluriformes: Pimelodidae) em diferentes densidades de estocagem nos primeiros dias de vida. **Revista Brasileira de Zootecnia**, v. 31, n. 2, p. 560-565, 2002.

MARTINS, M. L.; PILARSKY, F.; ONAKA, E.M.; NOMURA, D. T.; FENERICK Jr., J.; RIBEIRO, K.; MIYAZAKI, D. M. Y.; CASTRO, M. P.; MALHEIROS, E. B. Haematology and acute inflammatory response of *Oreochromis niloticus* (Osteichthyes: Cichlidae) submitted to a single and consecutive stress of capture. **Boletim do Instituto de Pesca**, v. 30, n. 1, p. 71-80, 2004a.

MARTINS, M. L.; TAVARES-DIAS, M.; FUJIMOTO, R. Y.; ONAKA, E. M.; NOMURA, D. T. Haematological alterations of *Leporinus macrocephalus* (Osteichthyes: Anostomidae) naturally infected by *Goezia leporini* (Nematoda: Anisakidae) in fish pond. **Arquivo Brasileiro de Medicina Veterinária e Zootecnia**, v. 56, n. 5, p. 640-646, 2004b.

MOISEENKO, T. I. Hematological indices of fishes in the evaluation of their toxicosis with reference to *Coregonus lavaretus*. **Journal of Ichthyology**, v. 38, p. 315-324, 1998.

OMOREGIE, E. Changes in the haematology of the Nile tilapia *Oreochromis niloticus* Trewavas under the effect of crude oil. **Acta Hydrobiologica**, v. 40, p. 287-292, 1998.

PEDRO, N.; GUIJARRO, A. I.; LÓPEZ-PATIÑO, M. A.; MARTÍNEZ-ALVÁREZ, R.; DELGADO, M. J. Daily and seasonal variations in haematological and blood biochemical parameters in the tench, *Tinca tinca* Linnaeus, 1758. **Aquaculture Research**, v. 36, n. 12, p. 1185-1196, 2005.

PILARCZYK, A. Changes in selected blood parameters in carp with signs of spring viremia of carp, gill necrosis, and tapeworm invasion. **Acta Hydrobiologica**, v. 28, n. 1-2, p. 253-263, 1986.

RANZANI-PAIVA, M. J. T.; SILVA-SOUZA, A. T.; PAVANELLI, G. C.; TAKEMOTO, R. M.; EIRAS, A. C. Hematological evaluation in commercial fish species from the floodplain of the upper Paraná river, Brazil. **Acta Scientiarum. Biological Sciences**, v. 22, n. 2, p. 507-513, 2000.

RANZANI-PAIVA, M. J. T.; ROMAGOSA, E.; ISHIKAWA, C. M. Hematological parameters of "Cachara", *Pseudoplatystoma fasciatum* Linnaeus, 1766 (Osteichthyes, Pimelodidae), reared in captivity. **Boletim do Instituto de Pesca**, v. 31, n. 1, p. 47-53, 2005.

RANZANI-PAIVA, M. J. T.; SILVA-SOUZA, A. T. *Hematology of Brazilian fish*. In: RANZANI-PAIVA, M. J. T.; TAKEMOTO, R. M.; LIZAMA, M. de los A. P. (Ed.). **Sanity of the aquatic organisms**. São Paulo: Varela, 2004. p. 89-120.

REHULKA, J. *Aeromonas* causes severe skin lesions in rainbow trout (*Oncorhynchus mykiss*): clinical pathology, haematology and biochemistry. **Acta Veterinaria**, v. 71, n. 3, p. 351-360, 2002.

ROSENFELD, G. Corante pancrômico para hematologia e citologia clínica. Nova combinação dos componentes de May-Grünwald e do Giemsa num só corante de emprego rápido. **Memórias do Instituto Butantan**, v. 20, p. 329-334, 1947.

SARAVANAN, T. S.; ANEEZ, M. M.; CHANDRASERAR, R.; SUNDRAMOORTHY, M. Fresh water fishes as indicators of Kaveri river pollution. **Journal of Environmental Biology**, v. 24, n. 4, p. 381-389, 2003.

SILVA-SOUZA, A. T. Haematology of fish from Tibagi river. I. Differential white blood cell counts in *Pimelodus maculatus* females. **Boletim do Instituto de Pesca**, v. 26, n. 1, p. 343-39, 2000.

SILVA-SOUZA, A. T.; ALMEIDA, S. C.; MACHADO, P. M. Effect of the infestation by *Lernaea cyprinacea* Linnaeus, 1758 (Copepoda, Lernaeidae) on the leucocytes of *Schizodon intermedius* Garavello and Britski, 1990 (Osteichthyes, Anostomidae). **Brazilian Journal of Biology**, v. 60, n. 2, p. 217-220, 2000.

SILVEIRA-COFFIGNY, R.; PRIETO-TRUJILLO, A.; ASCENCIO-VALLE, F. Effect of different stressors in haematological variables in cultured *Oreochromis aureus*. **Comparative Biochemistry and Physiology Part C**, v. 139, n. 4, p. 245-250, 2004.

SIMONATO, J. D.; GUEDES, C. L. B.; MARTÍNEZ, C. B. R. Biochemical, physiological and histological changes in the neotropical fish *Prochilodus lineatus* exposed to diesel oil. **Ecotoxicology and Environmental Safety**, v. 69, n. 1, p. 112-120, 2007.

SMITH, W. S.; PETRERE JR., M. Limnological characterization of the drainage basin of the river Sorocaba, São Paulo, Brazil. **Acta Limnologica Brasiliensis,** v. 12, n. 2, p. 15-27, 2000.

SOPINSKA, A. Effects physiological factors, stress, and disease on hematologic parameters of carp, with a particular reference to the leukocyte patterns: III. Changes in blood accompanying branchionecrosis and bothriocephalosis. **Acta Ichthyologica et Piscatoria**, v. 15, p. 141-165, 1985.

TAVARES-DIAS, M.; MELO, J. F. B.; MORAES, G.; MORAES, F. R. Haematological characteristics of Brazilian teleosts. VI. Parameters of jundiá *Rhamdia quelen* (Pimelodidae). **Ciencia Rural**, v. 32, n. 4, p. 693-698, 2002.

ZAR, J. H. **Biostatistical Analysis**. 4th ed. New Jersey: Upper Saddle River, 1996.

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