Research Journal of Animal, Veterinary and Fishery Sciences ____________________________ISSN 2320 – 6535
Vol. **11(1),** 1-13, May **(2023)** *Res. J. Animal, Veterinary and Fishery Sci.*

Changes in blood tissue of estuarine tilapia *Sarotherodon melanotheron* (Rüppell, 1852) captured in polluted Lake Nokoué (Benin, West Africa)

AGBOHESSI Prudencio[1*], KINSICLOUNON Gilles[2], OUEDRAOGO Alfred[3], GUEDEGBA Léa[1], ADOUWEKONOU Vanessa[1], HOUNDJI Alexis[1] and IMOROU TOKO Ibrahim[1]
[1]Research Laboratory in Aquaculture and Aquatic Ecotoxicology (LaRAEAq), Faculty of Agronomy, University of Parakou, Benin
[2]Laboratory of Applied Research in Biology (LARBA), University of Abomey-Calavi, Cotonou, Benin
[3]Laboratory of Histology, Embryology, Cytogenetics and Biology of reproduction of the University Hospital of Bogodogo; University of Ouaga 1 Prof. Joseph Ki Zerbo, Burkina-Faso
agbohessip@yahoo.fr

Available online at: www.isca.in, www.isca.me
Received 26th August 2022, revised 2nd March 2023, accepted 11th April 2023

Abstract

Lake Nokoué is the most productive Lake in West Africa, but unfortunately its resources are declining due to several factors including organic and chemical pollution. The present study aims to investigate the impact of this advanced pollution onestuarine tilapia Sarotherodon melanotheron. Thus, 60 sexually mature individuals (30 males and 30 females) of this specie were captured from the Lakes Nokoué and Toho with the Lake Toho considered as a reference. Blood samples were taken from fish of both sites to compare the hematological parameters (hemoglobin level (Hb), total leukocyte count (TLC), differential leukocyte count) and biochemical parameters (glycemia level, creatinine and transaminases). In addition, the physicochemical parameters of water from three sites (S1, S2 and S3) of Lake Nokoué and two sites (S4 and S5) of Lake Toho were recorded and the organic pollution indices (OPI) calculated. The findings of this investigation revealed an OPI = 1.33 ± 0.14 for Lake Nokoué, confirming its very high organic pollution compared to Lake Toho which has compatible OPI for living fish (OPI=3.875 ± 0.18).Regarding the hematological parameters, the study revealed in S. melanotheron from Lake Nokoué a low Hb and a high TLC emphasizing leukocytosis due to the compensation and defensive response versus the toxic effect of pollutants on fish. As for the biochemical parameters, high levels of plasma glucose, creatinine and transaminases reveal an environmental stress and hepato-nephrotoxic effects of the cocktail of pollutants on S. melanotheron in Lake Nokoué. All together, on one hand the study demonstrated the poor state and threat to the survival of S. melanotheron in Lake Nokoué and on the other hand, raises a public health problem related to the consumption of such fish.

Keywords: Hematological parameters; Biochemical parameters; Lake Nokoué; Cocktail of pollutants; *Sarotherodon melanotheron*.

Introduction

Lake Nokoué (Figure-1) is the most productive watercourse in West Africa with an annual fish production of more than 1 to 2 tons per hectare, against an average annual of 290 kg per hectare for all West African lagoons[1-3]. In the year 2000, the halieutic production of the lagoon complex Lake Nokoué - Porto-Novo lagoon represented 65 to 70% of the production of the continental rivers of Benin and the Lake Nokoué contributed for nearly 90%[2]. Unfortunately, these resources have now experienced a sharp decline due to the advanced pollution of this river. For example, in their inventory intended to identify species threatened with extinction, Lalèye *et al.*[4] listed 67 species while Gras[5] had listed 87. A difference of 20 species in 36 years which clearly shows an erosion of biodiversity for which one of the essential causes is pollution. Indeed, several authors have shown the pressure on Lake Nokoué which alone receives solid and liquid wastes from the surrounding populations and users of Dantokpa market, urban effluents from the city of Cotonou, Abomey-Calavi and Sô-Ava, agricultural effluents containing pesticides drained from the cotton-growing area of northern Benin by the Ouémé river or from agricultural areas surrounding the watercourse, industrial and hospital effluents from the city of Cotonou, fuel transit on this water course, etc[2,6-9]. All of these effluents release various types of pollutants into this ecosystem. The solid and liquid wastes of the surrounding populations and users of Dantokpa market contain nutrient salts including nitrogen, phosphate and trace minerals[9]. There are also dyes, lotions, solvents, shampoos and detergents, particularly alkyl phenol polyethoxylates (octylphenol and nonylphenol), plastics and industrial micropollutants such as polycyclic aromatic hydrocarbons (naphthalene, acenaphthene, fluoranthene, etc.) and dioxins and derived products[10,11] . We mainly find toxic metals such as mercury, lead, arsenic, cadmium, zinc, copper, etc., whose effects are multiple and varied[7]. The urban effluents coming from the city of Cotonou, Abomey-Calavi and Sô-Ava are themselves made up of effluents from hairdressing, dyeing, laundries and mechanics workshops, effluents from market gardening sites, water from paved roads, animal and sometimes human wastes, etc. These various effluents release not only the pollutants mentioned above but also the pesticides contained in market gardening effluents (organochlorines, organophosphates, pyrethrinoides, etc)[12-14]. Agricultural effluents drained from the cotton-growing area of northern Benin by the Ouémé river or from agricultural areas surrounding the river are also sources of organochlorine pesticides, organophosphates, etc[2]. Yehouénou A. Pazou[15] has also revealed levels in the water of DDT metabolites in this Lake, namely DDE and DDD in Ganvié (11.5 and 23µg/kg), in Abomey- Calavi (7.5 and 15µg/kg), Ahouansori (74 and 56 µg/kg) and Ladji (280 and 128µg/kg). These agricultural effluents also generate organic pollution which leads to an increase in ammonium, sulphates, phosphates with a continuous presence of nitrates and nitrites[11]. These types of environments are deficient in oxygen with very low primary production[16]. Industrial and hospital effluents from the city of Cotonou are also sources of toxic metals such as mercury from paper pulp bleaching activities, exhaust pipes and broken thermometers, cadmium from foundry and electroplating activities, lead and copper from used batteries, etc., and other chemical pollutants such as dioxins and alkylphenol polyethoxylates contained in biomedical wastes and in effluents from pulp and paper industries[17]. Fuel transportation and business on this Lake generate, among other things, petrogenic polycyclic aromatic hydrocarbons (PAHs), the most dangerous of which are benzo(a)pyrene, benzo(k)fluoranthene and benzo(ghi)perylene, organic lead known for its toxicity especially for fish, mercury, sulphur, radionuclides, etc[7,18,19]. Dovonou *et al.*[18] also showed the bacteriological pollution of this biotope due, among other things, to the putrefaction of various organisms thrown into this ecosystem. Fish and other aquatic organisms are therefore bathed in this cocktail of pollutants. Several studies have already revealed the high level of pollution and the multiplicity of toxicants in this ecosystem. Unfortunately, few studies have addressed to our knowledge the impact of this mixture of pollutants on the hematological and biochemical parameters, the physiological functions of interest in maintaining species in the environment. The present study therefore aims to assess the physiological status of estuarine tilapia *Sarotherodon melanotheron* captured in this Lake. We have assessed the ecological quality of the Lake in which the fish bathe and determined the hematological and biochemical parameters of *S. melanotheron*, one of the species in this ecosystem that is highly consumed by the population.

Materials and methods

Study area and site selection: Lake Nokoué is an aquatic ecosystem in south-eastern Benin (6°25′N, 2°26′E), with an area of 150 km^2 (Figure-1). It measures 20 km in the east-west direction and 11 km from north to south. This Lake is connected to the east with the lagoon of Porto-Novo with which it forms a freshwater Lake with an area of approximately 180km^2. Lake Nokoué is also attached to the Atlantic Ocean by the Channel of Cotonou, named Cotonou Lagoon[2], forming an expanse with a total length of 4.5km. To record the physicochemical parameters of the water, three sites (S1, S2 and S3) were chosen on this Lake. The first near the Dantokpa market (Cotonou), the second at the Abomey-Calavi pier and the third in Sô-Ava. Two reference sites (S4 and S5) were chosen on Lake Toho at Pahou (Ouidah) for data comparison. These sites are easily accessible at any season, but the study was carried out during the flood period in September-October 2021. Lake Toho is chosen as the reference watercourse in this study because it is located in the same geographical area as Lake Nokoué. Lake Toho is considered less polluted than Lake Nokoué. Located between the Agamé plateau and the northwest of the Bopa plateau, Lake Toho (6°37′09″N, 1°46′36″E) covers an area of 9.6 km² at low water and 15km² during flooding, and is in average 7x2.5km (length x width) in the south and around 500m (width) in the north.

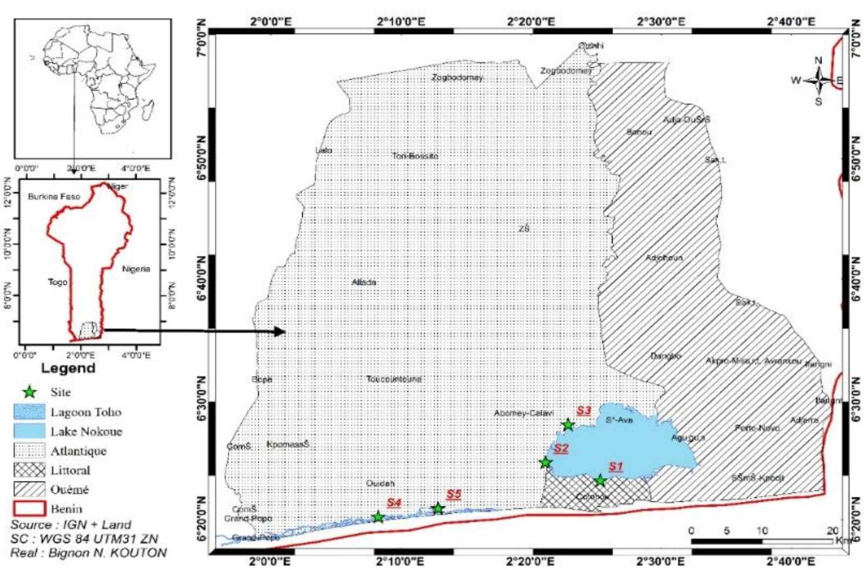

Figure-1: Card of Nokoué and Toho Lakes showing sampling sites.

Physicochemical parameters of water: The physicochemical variables of the water (dissolved oxygen, temperature, pH, transparency and electrical conductivity) were measured between 7-9 a.m. *in situ* on the different sites. Dissolved oxygen was measured using an oxythermometer (WTW oxi 197). The water temperature was measured at a depth of about 50 cm using the probe coupled to the same oxythermometer. The pH was measured using an ATC/HANNA/pHep3 pH meter with an accuracy of 0.1. Water transparency was measured using a Secchi disc fitted with a graduated string for measuring water depth. The electrical conductivity was recorded with a PCE-SM11 conductivity-meter with ± 0.5% precision and 0.01% resolution. Water samples were then taken per site and kept in a cooler at 4ºC and taken to the Research Laboratory in Aquaculture and Aquatic Ecotoxicology in Parakou for dosage of nutrients as nitrites, nitrates, phosphate and ammonium. The analyzes were carried out using a Hach Lange DR 2800 spectrophotometer. The measurement of the biological oxygen demand (BOD_5) was made by the respirometric method OxiTop in a thermostatically controlled enclosure (or a BOD-meter) at 20°C for five days. Total suspended solids (TSS) was measured by a Sension 5-HACH brand device. Each physicochemical parameter was measured 3 times at each site.

The organic pollution index (OPI) was calculated in order to assess the degree of organic pollution of the Lake[21]. Indeed, the OPI indicates the degree of alteration of water by chemical variables revealing organic pollution of the environment. It is calculated from 4 parameters divided into 5 classes (Table-1). The value of the parameter obtained is compared with the values of the table to know the number of the class to which it belongs. The OPI for a sample is equal to the mean of the parameter classes. It is given by the following formula:

$$OPI=(\sum \quad K \neq 0 \qquad C\,k, \ldots\ldots, C\,i)/n$$

with Ci: the class number of the parameter and n: the number of parameters analyzed.

Fish and blood sampling: Immediately after collecting water samples, wild *S. melanotheron* fish (Figure-2) were caught by professional fisherman using gill nets. This specie was chosen for the following reasons: i. *S. melanotheron* is the second most abundant fish species (15%) after *Ethmalosa fimbriata* (40%) in Lake Nokoué[1]; ii. *S. melanotheron* is more abundant in the catches of local fishermen and in the surrounding markets, and therefore more consumed by the populations; iii. Possibility of having sufficient numbers of this species during the study period; iv) Omnivorous species with a herbivorous tendency, columnar species with some stays in the bottom at the level of the sediments and therefore likely to be contaminated by pollutants of various kinds. Thus, 120 sexually mature individuals of *S. melanotheron* including 60 (30 males and 30 females) from Lake Nokoué and 60 (30 males and 30 females) from Lake Toho were carefully selected from fishermen's catches for the study. These fish were carefully transported to the Campus polyclinic laboratory (LAB/CPU) at Abomey-Calavi where the various samples and analyzes took place. In the laboratory, blood samples were immediately taken using 1 ml syringes through the tail veins and the blood samples taken were transferred into ETDA tubes and into previously labeled dry tubes for hematological and biochemical analyzes.

Table-1: Limit of classes for the calculation of the Organic Pollution Index[21].

Classes	NH^{4+} (mg/l)	NO^{2-} (µg/l of N)	PO_4^{3-} (µg/l)	DBO_5 (mg/l)	Class averages	Organic pollution levels
5	< 0.1	≤ 5.0	≤ 15.0	< 2.0	5.0-4.6	Null
4	0.1- 0.9	6.0-10.0	16.0-75.0	2.0-5.0	4.5-4.0	Weak
3	1.0- 2.4	11.0-50.0	76.0-250.0	5.1-10.0	3.9-3.0	Moderate
2	2.5 - 6.0	51.0-150.0	251.0-900.0	10.1-15.0	2.9-2.0	Strong
1	> 6.0	> 150.0	> 900.0	> 15.0	1.9-1.0	Very strong

Figure-2: Specimen of *Sarotherodon melanotheron* (Rüppell, 1852).

Hematological parameters: The blood smear was taken from the blood samples of each fish kept in the ETDA tubes. This smear is made by spreading a single drop of blood in a thin layer on slide, then staining for 5 min in a May-Grunwald bath diluted at 50% and Giemsa diluted at 20% in buffered water for 10 min. The various slides were observed under an OLYMPUS brand optical microscope (x 100 magnification) with a view to identifying and counting the hemoglobin content and the various leukocyte cells (neutrophils, eosinophils, basophils, lymphocytes, monocytes). The number of white blood cells was also counted using an electron microscope after diluting the blood sample with lazarus 1/50.

Biochemical parameters: Fish blood samples collected in dry tubes were centrifuged using a POWER SUPPLY 220 V model 80-2 brand electronic centrifuge to separate the serum from the pellet. For each biochemical parameter (glycemia, creatinine, transaminase) the serum was collected and mixed with its corresponding reagent. Each mixture thus produced was introduced into a peristactil pump spectrophotometer of the ERBA Manheim MODEL CHEM-7 brand. The results therefore appear on the screen of this device.

Statistical data analysis: The results are expressed as mean ± standard deviations. R software was used for statistical analyses. The homocedasticity of the data was tested using Levène's test. Thus, when the normality and homogeneity of the data are verified, the analysis of variance with two classification factors (Lakes and sites) for the physicochemical parameters (ANOVA 2) and with a single factor (Lakes) for the hematological and biochemical parameters (ANOVA 1) are performed. Duncan's multifactorial post-hoc test allowed us to make comparisons between the same sexes of fish from the two Lakes or between different sexes from the same Lake. For all tests, the significance threshold is set at 5%.

Results and discussion

Ecological quality of water: Based on Table-2, the water temperature recorded at a depth of about 50 cm between 7 and 9 a.m. varies on the two Lakes from 27.8 to 32.0°C. Dissolved oxygen is lower in Lake Nokoué (4.2 to 4.6 mg/l) than in Lake Toho (5.2 to 6.0 mg/l) even if the differences are not significant ($p>0.05$). Transparency also seems lower in Lake Nokoué (0.1 to 0.2 m) than in the reference Lake (0.71 to 0.8 m) although the differences are not significant ($p>0.05$). The pH which is neutral in Lake Toho (7.0 to 7.1) appears slightly acidic in the contaminated Lake (5.8 to 6.0). All the other parameters recorded (conductivity, nitrites, nitrates, phosphates, ammonium, COD, BOD_5 and TSS) are higher on Lake Nokoué than on Lake Toho ($p<0.05$). As for the OPI, it is low on the sites of Lake Nokoué (1.25 to 1.5) and higher on the sites of the reference Lake (3.75 to 4). Thus, the organic pollution of Lake Nokoué is remarkable and that of Lake Toho moderate.

Table-2: Physicochemical characteristics and OPI recorded at each site of Lake Nokoué and Lake Toho (n = 3). For each parameter studied, sites not bearing identical letters are statistically different at the 5% level.

	Lake Nokoué			Lake Toho	
	Cotonou	Abomey-Calavi	Sô-Ava	Pahou 1	Pahou 2
	S1	S2	S3	S4	S5
Water temperature (°C)	28.1±1.0	27.8±0.9	31.3±1.1	31.9±1.1	32.0±0.8
Dissolved oxygen (mg/l)	4.3±0.5	4.6±0.6	4.2±0.3	6.0±0.4	5.2±0.2
Transparency (m)	0.1±0.07	0.2±0.1	0.2±0.09	0.71±0.4	0.8±0.3
pH	5.8±0.6	6.0±0.5	5.8±0.5	7.1±0.9	7.0±0.5
Conductivity (µS/cm)	15.0±2.1a	12.6±1.9a	15.1±1.6a	9.8±2.7b	10.0±1.9b
NO^{2-} (mg/l)	0.9±0.7a	0.8±0.8a	0.2±0.1b	0.03±0.01b	0.01±0.01b
NO^{3-} (mg/l)	200.0±13.7a	160.0±16.8a	105.0±1.8a	20.1±3.5b	19.0±2.9b
PO_4^{3-} (mg/l)	0.6±0.2a	0.8±0.2a	0.8±0.1a	0.011±0.01b	0.012±0.01b
NH_4^+ (mg/l)	30.0±5.1a	24.0±2.3a	5.2±1.0b	0.5±0.1b	0.4±0.1b
Chemical oxygen demand (COD) (mg/l)	450.0±11.2a	255.0±8.9a	182.0±6.9a	50.0±2.9b	48.0±2.5b
Biological oxygen demand (BOD_5) (mg/l)	33.2±5.6a	30.4±5.3a	28.1±3.4a	5.1±1.1b	6.0±1.5b
Total Suspended Solid (TSS) (mg/l)	76.0±5.4a	74.0±6.3a	74.0±7.2a	53.0±8.2b	44.0±7.1b
OPI	1.25	1.25	1.5	3.75	4.0
	1.33 ± 0.14			3.875 ± 0.18	

Hematological parameters: The hemoglobin levels of individuals from Lake Nokoué are lower than those of the reference Lake (p<0.05) independently of the sex of the fish (Figure-3).

The Total leukocyte count (TLC) of fish from Lake Nokoué are significantly higher than those from the reference Lake (p<0.05) independently of the sex of the fish (Figure-4).

The reading of the different slides of the smear made it possible to establish the leukocyte formula of the fish. These are the means ± SD of the different proportions of leukocyte cells contained in the blood of the fish of different Lakes (Table-3). In both Lakes, the differential leukocyte count parameters of fish were the same and not related to the sex, with an upward trend in basophils in Nokoué fish.

Biochemical parameters: Based on Figure-5, the glycemia level of *S. melanotheron* from Lake Nokoué is significantly higher (p<0.05) than that of *S. melanotheron* from Lake Toho regardless of sex.

The level of creatinine (Figure-6) is statistically higher for fish from Lake Nokoué regardless of sex (p<0.05).

The transaminase level (Figure-7) is significantly higher in fish from Lake Nokoué regardless of sex (p<0.05).

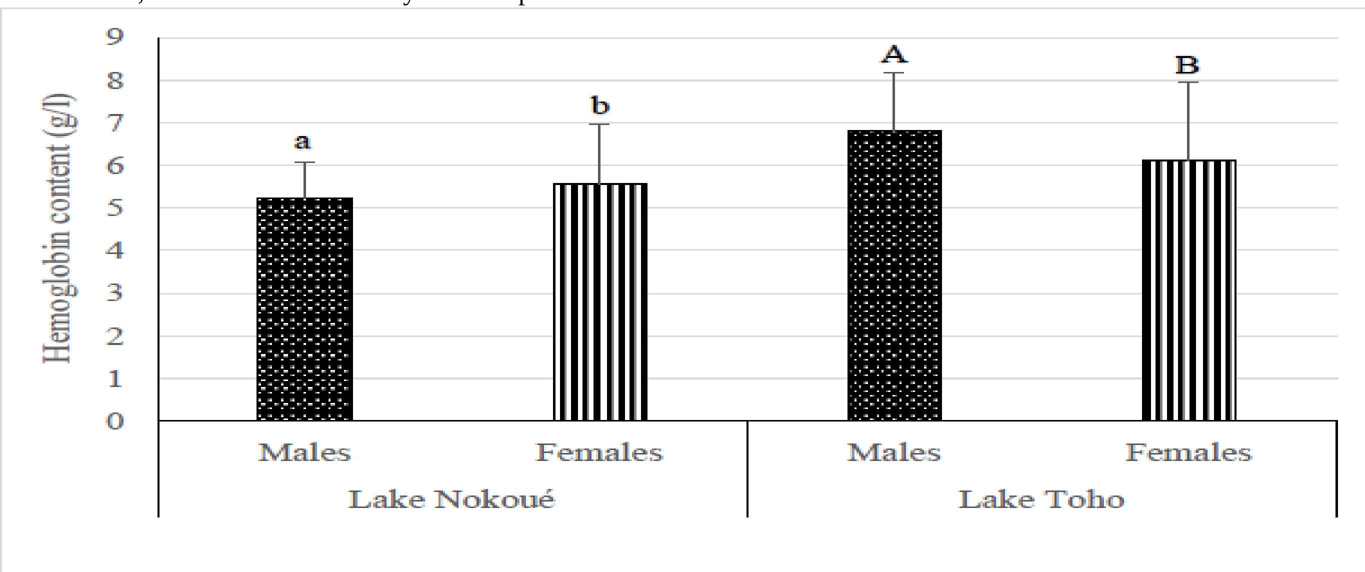

Figure-3: The hemoglobin content of *Sarotherodon melanotheron* captured in Lake Nokoué and Lake Toho. Data were expressed as mean ± SD. For each sex, the same upper and lower case letters indicate a significant difference between Lake (p< 0.05).

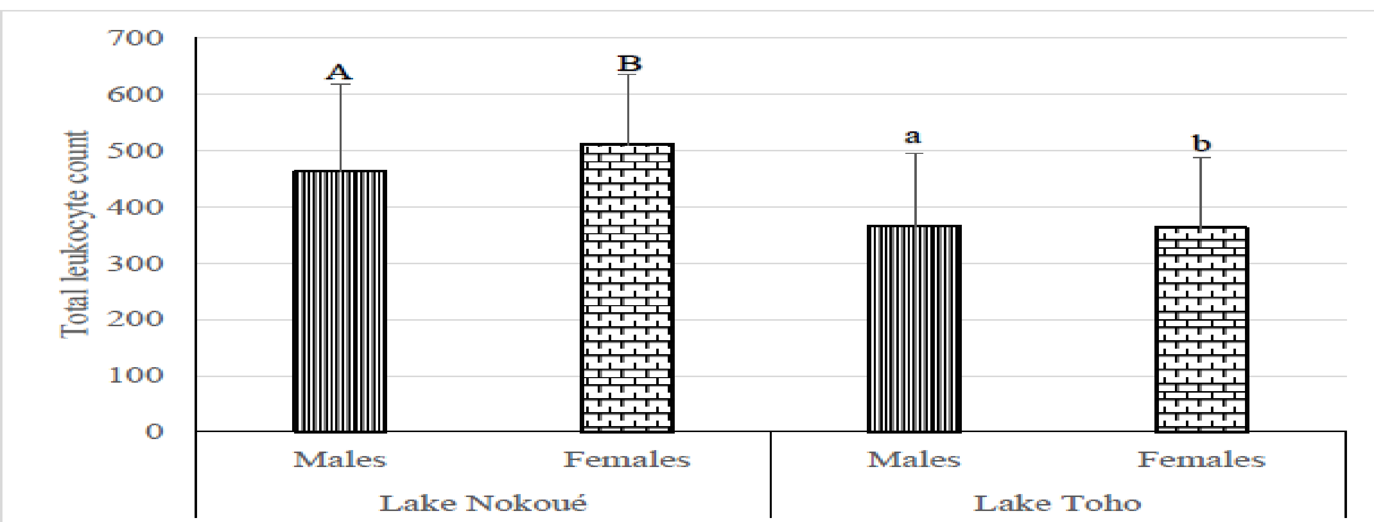

Figure-4: The Total leukocyte count of *Sarotherodon melanotheron* captured in Lake Nokoué and Lake Toho. Data were expressed as mean ± SD. For each sex, the same upper and lower case letters indicate a significant difference between Lake (p< 0.05).

Table-3: Alterations in differential leukocyte counts in *Sarotherodon melanotheron* captured in Lake Nokoué and in Lake Toho. Data were expressed as mean ± SD.

Lake	Sex	Lymphocytes	Neutrophils	Basophils	Eosinophils	Monocytes
Lake Nokoué	M	0.77 ± 0.13	0.15 ± 3.47	0.20 ± 0.10	0.06± 0.02	0.12 ± 0.07
	F	0.83 ± 0.13	0.04 ± 0.07	0.17± 0.06	0.00± 0.00	0.04 ± 0.05
Lake Toho	M	0.84 ± 0.11	0.05 ± 2.14	0.16± 0.10	0.04± 0.00	0.08 ± 0.03
	F	0.83 ± 0.09	0.07 ± 0.03	0.13 ± 0.11	0.06± 0.04	0.08 ± 0.04
P value Lake		> 0.05	> 0.05	> 0.05	> 0.05	> 0.05
P value sex		> 0.05	> 0.05	> 0.05	> 0.05	> 0.05

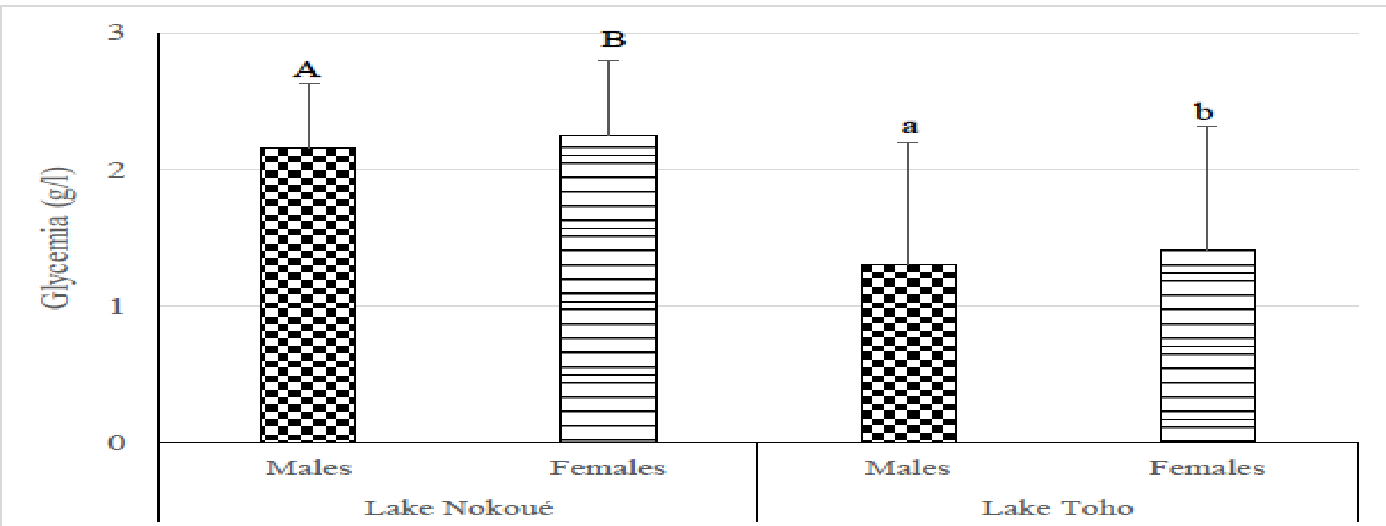

Figure-5: The glycemia level of *Sarotherodon melanotheron* captured in Lake Nokoué and Lake Toho. Data were expressed as mean ± SD. For each sex, the same upper and lower case letters indicate a significant difference between Lake (p< 0.05).

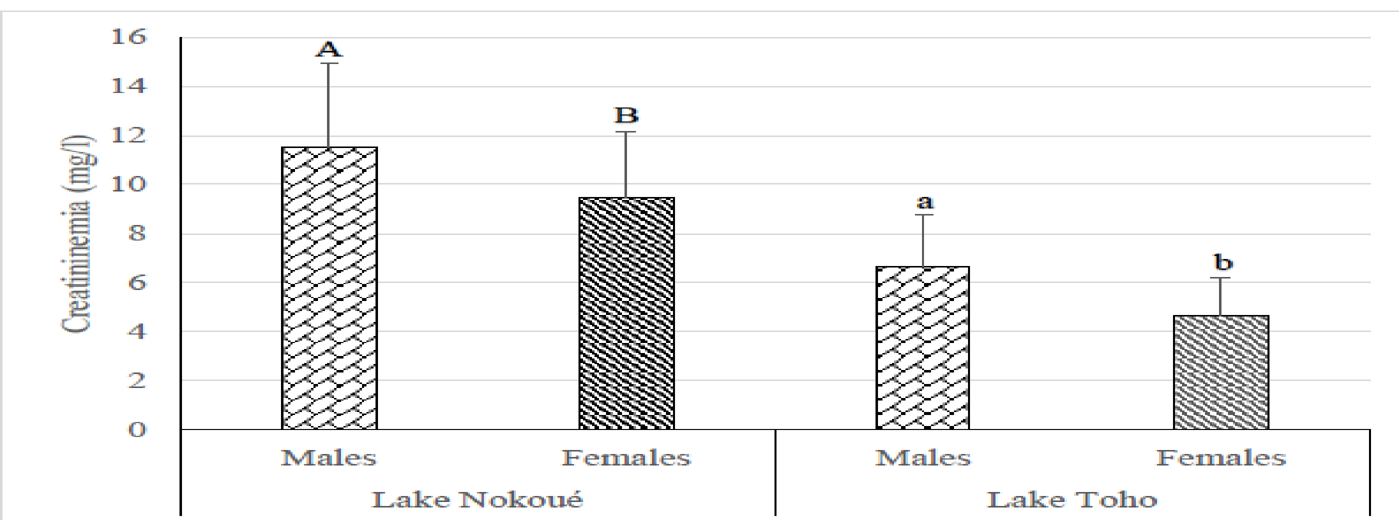

Figure-6: The Creatininemia level of *Sarotherodon melanotheron* captured in Lake Nokoué and Lake Toho. Data were expressed as mean ± SD. For each sex, the same upper and lower case letters indicate a significant difference between Lake (p< 0.05).

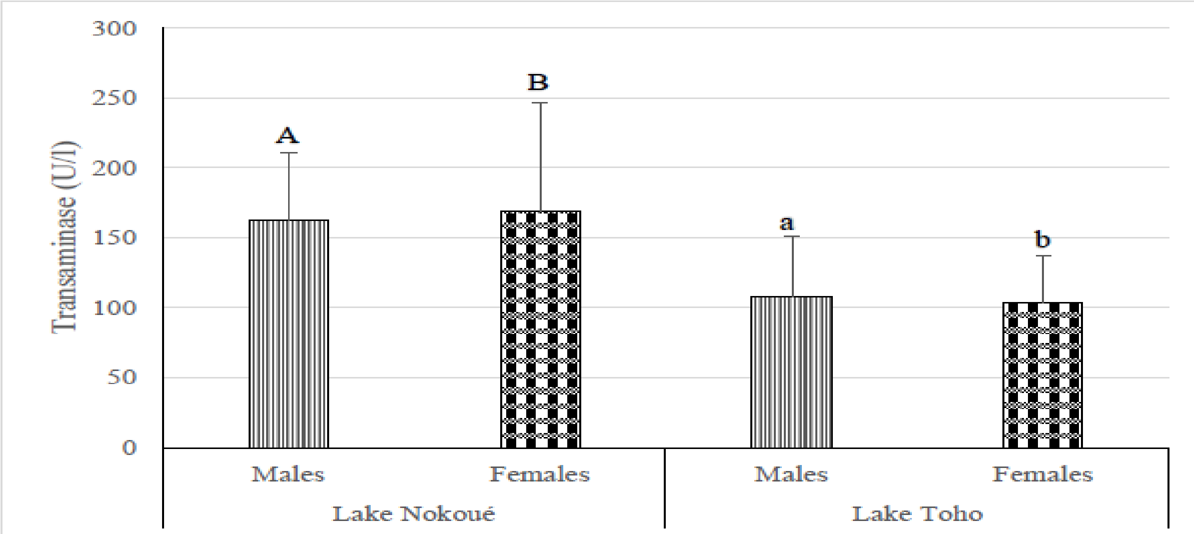

Figure-7: The Transaminase level of *Sarotherodon melanotheron* captured in Lake Nokoué and Lake Toho. Data were expressed as mean ± SD. For each sex, the same upper and lower case letters indicate a significant difference between Lake ($p < 0.05$).

Discussion

Ecological quality of water: The ecological balance of Lakes depends on their physicochemical quality depending on a certain number of intrinsic and extrinsic factors[22]. Indeed, the water quality is a key element among many parameters whose variations are decisive for the distribution of living organisms. An essential parameter in the evaluation of water quality is the temperature. Water temperature is directly subject to climatic conditions or seasons and the influence of tides. In the present study, the temperatures recorded on Lake Nokoué vary from 27.8 to 31.3°C and those of the reference Lake from 31.9 to 32.0 °C. These values are within the temperature range found by Lalèye *et al.*, Dovonou *et al.* and Capo-chichi *et al.*[1,18, 23]. Dissolved oxygen levels at Lake Nokoué sites are relatively low (4.2-4.6 mg/l) compared to Lake Toho sites (5.2-6.0 mg/l). In estuarine and lagoon environments, the decrease in oxygen can come from the consumption by benthic organisms, the decomposition of organic matter and the presence in suspension of oxygen-consuming elements as well as the arrival of waters poor in oxygen[24]. These conditions contribute, more or less, to the oxygen deficits of Lake Nokoué. Similar results were also found by Zinsou *et al*. and by Capo-chichi *et al*.[23,25]. The pH values at the Lake Nokoué sites ranged from 5.8 to 6, relatively low values compared to the Lake Toho sites (7.0-7.1). These values recorded on Lake Nokoué are not consistent with those found by Dovonou *et al*. and Capo-chichi *et al.* [18, 23]on the same Lake. These acid pH values obtained in our study for Lake Nokoué can be linked to the choice of sites that are close to dwellings and uncontrolled dumping grounds for household waste and other waste. Transparency on Lake Nokoué is low (0.1-0.2 m) compared to Lake Toho (0.71-0.8 m). These lower values of transparency on Lake Nokoué are probably related to the period of measurement (flood) of this variable. Indeed, during the flood, Lake Nokoué receives plenty of charged water[26]. These low levels of water transparency in the Nokoué also justify the high levels of suspended solids recorded on sites S1, S2 and S3. The electrical conductivity of water is a measure of its ability to conduct electric current. The values recorded on the two Lakes in this flood period are less than 100 µs/cm and show a very low mineralization of the waters of these Lakes[18]. The high values of conductivity recorded in the Nokoué, reflect a much mineralized water and too loaded with dissolved matter[27]. For all the other parameters (nitrates, nitrites, phosphates, ammonium, COD, BOD_5 and TSS), the values measured on the Nokoué are relatively higher than those of the reference Lake sites. The high presence of nitrates in water bodies is due either to the leaching of agricultural land or to the oxidative reactions of ammoniacal nitrogen and nitrites[25,28]. The most serious and oldest known effect of nitrates is methemoglobinemia[18]. Nitrites come from the decrease of nitrates under bacteria influence. The values obtained on Lake Nokoué are above the admissible limit of 0.06 mg/l corresponding to the lower threshold of the acute toxicity range [29]. The same trends had also been observed by Dovonou *et al*. and Capo-chichi *et al*.[18,23] for the same ecosystem. The concentrations of phosphorus in Nokoué are above the admissible limit of 0.5 mg/l and can be considered as an indicator of pollution[30]. Domestic effluents from the cities of Cotonou and Abomey-Calavi which are drained in the Lake, contain detergents which are according to Jen[31] sources of phosphorus. This observation was made by Dovonou *et al*. Zinsou *et al.* and Capo-chichi *et al*.[18,23,25] for the same stream. Phosphorus levels found in Lake Nokoué are below the European toxicity standard set at 6.1 mg/l. High concentrations of phosphates are not toxic. However, in the pond one should not exceed 0.5 mg/l before avoiding the proliferation of algae. In the present study, Lake Nokoué has a higher ammonium level than the reference. The distribution of ammonium in an aquatic environment varies according to the level of productivity of the ecosystem and its degree of pollution by the presence of organic matter. Its presence would result from the aerobic degradation of organic nitrogen (proteins, amino acids, urea, etc.), which comes largely from discharges of untreated water. Indeed, NH_4^+ in itself is not harmful but can be transformed under certain conditions into ammonia (NH_3), a gas soluble in water and toxic for aquatic life[32]. Biological oxygen demand (BOD_5) is a measure of water pollution by organic matter. This is the quantity of oxygen necessary for living microorganisms to ensure the oxidation and degradation of organic matter present in wastewater in 5 days at 20°C and protected from light. The high BOD_5 values of Lake Nokoué compared to Lake Toho therefore show the high organic pollution of Lake Nokoué.The main sources of organic pollution of the LakeNokoué are on one hand, the deposit of branches in the Lake (acadja) andon the other hand, the wild rubbish dumps located along the shore of Lake Nokoué in Cotonou[18]. The BOD_5 of Lake Nokoué water is between 28.1 to 33.2 mg/l. These results are slightly higher than those found by Dovonou *et al.* Mama *et al.* and Capo-chichi *et al.*[18,23,33]. This high level of BOD_5 in this study is surely linked to the choice of sites that are close to household waste dumps. These high values of BOD_5 are also explained by the decomposition of macrophytes in the Lake under the effect of high water temperature and by the absence of dilution by fresh water (Oueme River or rainwater). The decomposition of all these plants and garbage causes a significant consumption of dissolved oxygen, and will be accompanied by deoxygenation of the environment, in particular at the water/sediment interface. The high amounts of organic matter highlighted by the low values of the OPI (1.25-1.5) reflect a very high organic pollution of Lake Nokoué. These results corroborate those of Zinsou et *al*. and Capo-Chichi *et al*.[23,25]. These low values of this OPI are explained by the very high ammonium and phosphate contents[34,35]. It is urgent that the planners take measures to improve the quality of this ecosystem for the good health of the fish and the consumers of these fish.

Hematological parameters of *S. melanotheron*: Hemoglobin is a blood pigment that is involved in respiration. In fish, it transport oxygen from gills to the tissues and thus contributes in cellular respiration. Hemoglobin content furnishes an indirect valuation of the number of red blood cells in the blood[36]. In the present study, the hemoglobin level is low in fish from Lake Nokoué regardless of sex compared to fish from the reference Lake. Reduce in Hb content in fish could be due to either its destruction or decreased synthesis[37]. This decline in hemoglobin density in fish from Lake Nokoué can be linked to this organic pollution noted in this ecosystem. In fact, nitrites act directly on hemoglobin by oxidizing the ferrous ion, forming methemoglobin which is incapable of transporting respiratory gases[18]. Other studies had also reported the pollution of Lake Nokoué by pesticides drained from the cotton-growing area of northern Benin by the Ouémé River or from agricultural effluents from Nokoué surroundings[2,38,39]. These are organochlorine pesticides, organophosphates, pyrethroids, etc. In fact, organophosphate pesticides would engender anemia due to significant reduce in Hb level has been reported in *Barbonymus gonionotus* exposed to quinolphos, *Cyprinus carpio* exposed to monocrotophos, *Ctenopharyngodon idella* exposed to dichlorvos, and in *Oreochromis mossambicus* exposed to chlorpyrifos[37,40-42]. Such anemic conditions have alsoin fish exposedto herbicide clomazone and fungicide difenoconazole[43,44]. Similar trend of decrease in Hb level was reported in several fish species exposed to pyrethroid pesticides cypermethrin, permethrin and deltamethrin[45-48]. A significant decrease in Hb level after exposure to pesticides and other toxicants were reported by several investigators[49-51]. The reduction in hemoglobin density found in fish from Lake Nokoué in our study may also be related to toxic metals such as mercury, cadmium, zinc and lead, found in this watercourse and shown by several studies[7,52]. In fact, for example in the blood, cadmium binds to the membrane of erythrocytes, thus causing anemia in animals according to the study by Anju[53]. At doses of 10mg/kg and 20mg/kg, a decrease in hemoglobin and red blood cells were observed[54,55.] Cadmium-induced anemia has several causes, namely that cadmium would have caused iron deficiency due to gastric absorption, hemolytic anemia due to sequestration of red blood cells in the spleen and kidney damage thus preventing the production of erythropoietin. Indeed, by accumulating in the spleen, liver and kidneys, metals inhibit erythropoietic activity by damaging the synthesis of erythropoietin which is a hormone secreted by the kidneys whose role is to stimulate red blood cells[56]. Cadmium therefore disrupts the hematopoietic system[57]. Thus, for Hounkpatin *et al.*[58], cadmium causes blood disorders. In their study on the combined effect of cadmium and mercury toxicity, they found a decrease in hemoglobin symbolizing hypochromic and macrocytic anemia. Several other pollutants have been revealed in Lake Nokoué such as polycyclic aromatic hydrocarbons and others which can also induce a drop in the level of hemoglobin in *S. melanotheron* from Nokoué[11]. This drop in hemoglobin density may also be due to the combined effect of all the pollutants present in this watercourse.

Total white blood cell count is the measurement of total number of white blood cells or leukocytes in the blood. TLC is modified due to the effect of toxics[36]. In the present study, the TLC in fish from Lake Nokoué is higher than those from Lake Toho. Pathophysiological condition of increased TLC is leukocytosis. Increased leukocyte count could be a compensation and defensive response versus the toxic effect of pesticides on fish[37]. Several studieson TLC reveal a significant increase in its value in *Channa punctatus* under the influence of deltamethrin[45], in *C. carpio* exposed to fenthion[59], in *C. carpio* due to monocrotophos[40], in *C. punctatus* exposed to endosulfan and dimethoate[60], in *Heteropneustes fossilis* due to chlorpyrifos[61], and in *C. idella* exposed to dichlorvos and its technical grade[41]. The increase in the white blood cell count may also be due to the heavy metal contamination of Lake Nokoué, such as cadmium, which induced a similar result in Wistar rats[62]. The increase in these blood cells reflects a general inflammatory state due to damage caused by cadmium toxicity[57,63]. This could be explained by the stimulation of the immune system leading to these increases[63]. This considerable increase characterizes leukemia according to a study carried out on rats contaminated by ingestion with cadmium chloride[64]. These results corroborate those of Jelena *et al.*[65] who observed a significant increase in peripheral blood leukocytes compared to the control in rats treated by intraperitoneal injection at 1mg/kg for 48 hours with cadmium. Also, Gabol *et al*. and Borane[66,67] found an increase in white blood cells and a decrease in red blood cells after administration of cadmium at respective doses of 20µg/kg to domestic chickens and 1.248 ppm to fish. Also in amphibians depending on the exposure doses (0.25; 0.50; 1.00 and 2.00 mg/l), Alex and Lawrence[63] observed an increase in leukocytes. Other pollutants including bacteria revealed in the water of this stream[18] can also promote the elevation of the white blood cell count. But the differential leukocyte count (neutrophils, basophils, eosinophils, lymphocytes and monocytes) did not vary significantly from one Lake to another and from one sex to another.

Biochemical parameters of *S. melanotheron*: Glycemia is the level of glucose in the blood, or more precisely in the blood plasma. Among the biochemical parameters, plasma glucose has been extensively used as a sensitive indicator of environmental stress in fish, because carbohydrates are the primary and immediate source of energy[68]. The blood glucose level of exposed fish was significantly elevated, which may be due to mobilization of glycogen into glucose[68]. It is well established that fish secrete large amounts of glucocorticoids and catecholamines from adrenal tissue under stress. These hormones are well known to cause hyperglycemia in animals. The hyperglycemic state recorded in fish exposed to pollutants can be attributed to increased secretion of the above mentioned hormones which induce glycolysis in the liver and muscles of fish exposed to pollutants[68]. Similar results were given by various other authors suggesting that glucose concentration increases as a general response of fish to acute pollutant effects, including organophosphate pesticides[69,70]. As observed in fish from Lake Nokoué, glucose level was increased in *Clarias gariepinus* exposed to diazinon[71], in *Cirrhinus mrigala* (Hamilton) exposed to chlorpyrifos[72], in *C. carpio* after sub-chronic exposure to copper[73], and in *O. niloticus* contamined to copper, zinc, lead and cadmium[74]. The effects on the use of glucose in fish from Lake Nokoué can be induced by other pollutants present in this Lake or by the cocktail of pollutants in this ecosystem.

Creatinine is exclusively released through the kidneys, which is a very good marker of kidney function. The high creatinine level in *S. melanotheron* from Lake Nokoué compared to the reference Lake, indicates a very poor functioning of the kidneys of these contaminated fish. Similarly, *Clarias batrachus* exhibited elevated creatinine levels when exposed to 2.5 ppm cadmium. This elevated creatinine levels may indicate that cadmium is a nephrotoxic metal to fishes which cause kidney dysfunction[75]. Ahmad and Gautam[76] treated water catfish (*H. fossilis*) with sublethal doses of nuvan and a significant increase was noticed for creatinine. These findings were also supported by Soufy *et al.*[77] who worked on monosex tilapia by treating it with carbofuran. Reduction of serum creatinine was observed in *C. gariepinus* after paraquat dichloride toxicity[78]. Several pesticides also improved the plasma creatinine level such as malathion in *O. nilotius*[79], di-N-butyl phthalate in tilapia[80], malathion in *C. punctatus*[81], titanium dioxide nanoparticles and paraquat in *C. carpio*[82].

Transaminase or aminotransferase is an enzyme which catalyzes a type of reaction between an amino acid and an α-keto acid. Pyridoxal phosphate enzyme of the transferase class, it transfers a -NH2 group from an amino acid to another, allowing the synthesis of new amino acids, not present in the environment. There are two kinds of transaminase: Alanine-aminotransferase (ALT) and Aspartate-aminotransferase (AST). AST and ALT enzymes are used to assess the hepatotoxic effects of pollutants, and are therefore considered important diagnostic tools[83]. The results of the present investigation show that the exposure of pollutants to *S. melanotheron* significantly increase the activity of these enzymes. The activity of the enzymes AST and ALT was increased in the fish due to exposure tometals[84]. In the similar manner, Jeney *et al.*[85] has registered an increased concentration of enzymes (AST, ALT) in the serum of fish when treated with ammonia. According to them ALT is very sensitive to the changes in the environment conditions. Significantly higher values of AST and ALT activities were presented by Mekkawy *et al.*[86] in the fish blood treated with cadmium. Increased activity of AST and ALT in fish, *C. punctatus*, after the treatment of mercuric chloride[87] and monocrotophos[88] were also observed. Liver is rich in AST and ALT as suggested by Vaglio and Landriscina[89] and Palanivelu *et al.*[90] and any damage to liver could lead to increase levels of these enzymes into the blood. Changes in the activity of these enzymes in fish due to exposure to pollutants could be a sensitive indicator of cellular damage[90]. The high activity values of these enzymes found in the fish of Lake Nokoué may be attributed to damage caused to liver by the pollutants.

Conclusion

The results of this investigation indicate that Lake Nokoué is heavily polluted and these pollutants are toxic for *S. melanotheron* in which the hematological and biochemical profiles are altered. Exposure of this species to these toxics lowered hemoglobin levels and significantly increased TLC and transaminase activity. The blood glucose level was also high. This shows how toxic these pollutants are, therefore, more attention must be paid to the pollution of this Lake to curb the erosion of biodiversity and especially to preserve the health of the populations consuming these fish.

Acknowledgement

The authors sincerely thank the fishermen who helped in the capture of the fish, all the students and the laboratory technicians who took part in the transport of the fish, the blood sampling and the analyses.

References

1. Lalèye, P.A, Niyonkuru, C., Moreau, J. and Teugels, G.G. (2003). Spatial and seasonal distribution of the ichthyofauna of Lake Nokoué, Benin. West Africa. *African J. Aquatic Sci.*, 28 (2), 151-161. https://doi.org/10.2989/16085910309503779

2. Yehouenou, A., Pazou, E., Aléodjrodo, P.E., Azehoun, J.P., van Straalen, N.M., van Hattum, B., Swart, K. and van Gestel, C.A.M. (2014). Pesticide residues in sediments and aquatic species in Lake Nokoué and Cotonou lagoon in the Republic of Bénin. *Environ Monit Assess*, 186, 77–86.

3. Issola,Y., Kouassi, A.M., Dongui, B.K. and Biemi, J. (2008). Caractéristiques physico-chimiques d'une lagune côtière tropicale : lagune de Fresco (Côte d'Ivoire). *Afrique SCIENCE*, 04(3), 368 – 393.

4. Lalèye, P., Chikou, A. and Wuemenou, T. (1997). Poissons d'eaux douces et saumâtres du Bénin: Inventaire, distribution, statut et conservation. Inventaire des poissons menacés de disparition du Bénin. Rapport d'étude. Coopération bénino-néerlandaise/Ambassade Royale des Pays-Bas, Cotonou (Bénin), 80p.

5. Gras, R. (1961). Liste des espèces du bas Dahomey faisant partie de la collection du laboratoire d'hydrobiologie du service des eaux, forêts et chasses du Dahomey. *Bulletin de l'Institut Français d'Afrique Noire*, 23 (a2), 527-586.

6. Cledjo, P. (1999). La gestion locale de l'environnement dans les cités du lac Nokoué (Région urbaine du littoral du sud Bénin). Mémoire de DEA de l'Université d'Abomey-Calavi (Bénin), 60p.

7. Youssao, A., Soclo, H.H., Bonou, C., Vianou, K., Gbaguidi, M. and Dovonon, L. (2011). Évaluation de la contamination de la faune ichthyenne dans le complexe lagunaire Nokoué – chenal de Cotonou par le plomb : cas des espèces *Sarotherodon melanotheron*, *Tilapia guineensis* et *Hemichromis fasciatus* (Bénin). *Int J Biol Chem Sci*., 5(2), 595–602.

8. Yèhouénou, E.A.P., Adamou, R., Azéhoun, P.J., Edorh, P.A. and Ahoyo, T. (2013). Monitoring of heavy metals in the complex "Nokoué Lake - Cotonou and Porto-Novo lagoon" ecosystem during three years in the Republic of Benin. *Res J Chem Sci*, 3(5), 7.

9. Saizonou, J., Ouendo, E.M., Agueh, V., Tokplonou, E. and Makoutodé, M. (2014). Évaluation de la qualité de la gestion des déchets biomédicaux solides dans la zone sanitaire Klouekanme-Toviklin-Lalo au Bénin. *J Int Santé Travail*, 1, 1-11.

10. Tossou, E. S. (2000). Impact du trafic des produits pétroliers sur les écosystèmes lacustres: Cas du lac Nokoué et de la lagune de Cotonou (Bénin). Mémoire de DEA/FLASH/UAC, p132.

11. Dovonou, F. E., Ibikounle, M., Akouedegni, C. G., Aissi, V., Dossou, M. P., & Mama, D. (2019). Impacts des Hydrocarbures Aromatiques Polycycliques sur les poissons: Cas des Tilapias du lac Nokoué au Sud du Bénin (Afrique de l'Ouest). *Eur Sci J*, 15, 458-474.

12. Yehouenou, A., Pazou, E., Boko, M., Van Gestel, C.A.M., Ahissou, H., Lalèyè, P., Akpona, S., Van Hattum, B., Swart, K. and Van Straalen, N.M. (2006). Contamination of fish by organochlorine pesticide residues in the Ouémé River catchment in the Republic of Bénin. *Environ int*, 32, 594- 598.

13. Yehouenou, A., Pazou, E, Lalèye, Ph., Boko, M., Van Gestel, C.A.M., Ahissou, H., Akpona, S., Van Hattum, B., Swart, K. and Van Straalen, N.M. (2006). Organochlorine and organophosphorous pesticide residues in the Ouémé River catchment in the Republic of Bénin. *Environ int*, 32, 616- 623.

14. Agbohessi, T.P., Imorou Toko, I., Ouédraogo, A., Jauniaux, T., Mandiki, S.N.M. and Kestemont P. (2015). Assessment of the health status of wild fish inhabiting a cotton basin heavily impacted by pesticides in Benin (West Africa). *Sci Tot Environ*, 506, 567-584.

15. Yèhouénou, A.P.E. (2005). Les résidus de pesticides chimiques de synthèse dans les eaux, les sédiments et les espèces aquatiques du bassin versant du fleuve Ouémé et du lac Nokoué. Thèse de doctorat unique de l'Université d'Abomey-Calavi, Bénin, p. 217.

16. Bankolé, A.L. and Olou, A.C. (2000). Evaluation de la pollution inorganique azotée et phosphorée dans les écosystèmes du lac Nokoué et du chenal de Cotonou. Identification des sources. Mémoire pour l'obtention du DIT. / CPU. Cotonou, 82p.

17. Thompson, G., Swain, J., Kay, M. and Forstera, C.F. (2001). The treatment of pulp and paper mill effluent: a review. *Bioresour. Technol*., 77(3), 275-286.

18. Dovonou, F., Aina, M., Boukari, M. and Alassane, A. (2011). Pollution physico-chimique et bactériologique d'un écosystème aquatique et ses risques écotoxicologiques : cas du lac Nokoué au Sud Benin. *Int J Biol Chem Sci*, 5(4), 1590-1602.

19. Bawa, M.L., Djaneye-Boundjou, G. and Boukari, Y. (2005). Caractérisation de deux effluents industriels au Togo: étude d'impact sur l'environnement. *Afrique SCIENCE*, 02, 57 – 68.

20. Ahouansou-Montcho, S. (2003). Etude de l'écologie et de la production halieutique du lac Toho au Bénin. Mémoire de DESS/FSA, Université d'Abomey-Calavi, Bénin, 87p.

21. Leclercq, L. (2001). Intérêt et limites des méthodes d'estimation de la qualité de l'eau. Station scientifique des Hautes-Fagnes, Belgique, Document de travail, 44p.

22. Lalèye, PA. (1995). Ecologie comparée de deux espèces de Chrisisthys, poissons siluriformes (claroteidae) du complexe lagunaire Lac Nokoué-lagune de Porto-Novo au Bénin. Thèse de Doctorat en Sciences, Université de Liège (Belgique), 152p.

23. Capo-chichi, H.B.P., Adandédjan, D., Aglonon Houélomè, T.M. and Lalèye P. (2022). Physico-chimie et pollution organique du lac Nokoué au Sud du Bénin. *J Appl Biosci*, 170, 17752– 17775.

24. Villanueva, M.C.S. (2004). Biodiversité et relations trophiques dans quelques milieux estuariens et lagunaires de l'Afrique de l'Ouest: Adaptations aux pressions environnementales. *Thèse de doctorat*, 272 p.

25. Zinsou, H.L, Attingli, A.H., Gnohossou, P., Adandedjan, D. and Lalèye, P. (2016). Caractéristiques physico-chimiques et pollution de l'eau du delta de l'Ouéme au Bénin. *J Appl Biosci*, 97, 9163 – 9173.

26. Gnohossou P. (2006). La faune benthique d'une lagune ouest africaine (le Lac Nokoué au Bénin), diversité abondance, variation temporelles et spatiales, place dans la zone trophique. Thèse de Doctorat. Institut national polytechnique de toulouse. Formation doctorale: SEVAB, 169p.

27. Chouti, W., Chitou, N., Kelome, N., Kpakpo, B.H., Honvou Vlavonou, D. and Tossou, M. (2017). Caractérisation physico-chimique et étude de la toxicité de la lagune côtière, de Togbin à Grand-Popo (Sud-Ouest Bénin). *Eur Sci J*, 13, 131-151.

28. Khalaf, G., Slim, K., Saad, Z., Nakhlé, K. (2007). Evaluation de la qualité biologique des eaux du Nahr el Jaouz (Liban): application des méthodes indicielles. *Bulletins mensuels Sommaires - Société linnéenne de Lyon*, 76 (9–10), 255- 268.

29. Djibril R. (2001). Impact de l'utilisation des engrais chimiques et des pesticides sur la qualité de l'eau de surface dans la réserve de la biosphère de la Pendjabi. Mémoire de fin de formation CPU/UAC, 160 p.

30. Agblonon Houelome TM, Adandedjan D, Chikou A, Imorou Toko I, Youssao I, Laleye P. (2017). Caractérisation de la qualité physico-chimique des eaux de la rivière Alibori dans le bassin cotonnier du Bénin. *Afrique SCIENCE*; 13(4), 35 – 49.

31. Jen O. (2002). The perceived Environmental Impact of Car Washing. Ramsey-Washington Metro Watershed District, 9p.

32. Koudenoukpo, Z.C., Chikou, A., Adandedjan, D., Hazoume, R., Youssao, I., Mensah, G.A and Laleye AP. (2017). Caractérisation physico-chimique d'un système lotique en région tropicale: la rivière Sô au Sud Bénin, Afrique de l'Ouest. *J Appl Biosci*, 113, 11111-11122.

33. Mama, D., Aina, M., Alassane, A., Boukari, O.T., Chouti, W., Deluchat, V., Bowen, J., Afouda, A. and Baudu, M. (2011). Caractérisation physico-chimique et évaluation du risque d'eutrophisation du lac Nokoué (Bénin). *Int J Biol Chem Sci*, 5(5), 2076-2093.

34. Benzizoune, S., Nassali, H. and Srhiri, A. (2004). Etude de la cinétique d'adsorption du phosphore en solution sur les sédiments du lac Fouarat au Maroc. *Larhyss J*, 3, 171-184.

35. Benbouih, H., Nassali, H., Leblans, M. and Srhiri A. (2005). Contamination en métaux traces des sédiments du lac Fouarat (Maroc). *Afrique SCIENCE*, 1(1), 109-125.

36. Sinha, B.K., Gour, J.K., Singh, M.K. and Nigam, A.K. (2022). Effects of pesticides on haematological parameters of fish: Recent updates. *J Sci Res*, 66(1), 269-283.

37. Mostakim, G.Md., Zahangir, Md.M., Mishu, M.M., Rahman, Md.K. and Islam, M.S. (2015). Alteration of blood parameters and histoarchitecture of liver and kidney of silver barb after chronic exposure to quinalphos. *J Toxicol*, 415984, 1-8.

38. Okoumassoun, L.E., Brochu, C., DeBlois, C., Akponan, S., Marion, M., Averill-Bates, D. and Denizeau, F. (2002). Vitellogenin in tilapia male fishes exposed to organochlorine pesticides in Ouémé River in Republic of Benin. *Sci Tot Environ*, 299, 163–172.

39. Agbohessi, T.P., Imorou Toko, I. and Kestemont, P. (2012). État des lieux de la contamination des écosystèmes aquatiques par les pesticides organochlorés dans le bassin cotonnier béninois. *Cahiers Agricultures*, 21, 46–56.

40. Vaiyanan, V., Sridharan, G., Raveendran, S. and Chairman, K. (2015). Impact of pesticide on haematological parameters of *Cyprinus carpio*. *World j pharm pharm sci*, 4(8), 1424-1430.

41. Kumari, R.S., Kumar, M.V. and Rao, N.G. (2018). Haematological changes induced in the fish *Ctenopharyngodon idella* (Valenciennes) exposed to organophosphate Dichlorvos both technical and 76% EC (Nuvan). *J Inn Pharm Biol*, 5(2), 58-63.

42. Ghayyur, S., Tabassum, S., Ahmad, M.S., Akhtar, N. and Khan, M.F. (2019). Effect of chlorpyrifos on hematological and seral biochemical components of fish *Oreochromis mossambicus*. *Pakistan J Zool*, 51(3), 1047-1052.

43. Pereira, L., Fernandes, M.N. and Martinez, C.B.R. (2013). Hematological and biochemical alterations in the fish *Prochilodus lineatus* caused by the herbicide clomazone. *Environ Toxicol Pharm*, 36, 1-8.

44. Jasmin, J., Rahman, Md.M. and Rahman, Md. M. (2018). Haematological changes in *Labeo rohita* (H.) due to exposure of pesticides, difenoconazole and thiamethoxam. *Int J Contemporary Res Review*, 9(01), 20199-20205.

45. Jayaprakash, C. and Shettu, N. (2013). Changes in the hematology of the freshwater fish, *Channa punctatus* (Bloch) exposed to the toxicity of deltamethrin. *J Chem Pharm Res*, 5(6), 178-183.

46. Neelima, P., Sunitha, K., Gopala Rao, N., Chandra, S. and Rao, J. (2015). Haematological alterations in *Cyprinus carpio* as biomarkers of cypermethrin toxicity. *Int J Cur Res*, 7(8), 18864-18870.

47. Rao, G.N., Naik, B.K.R. and Rao, S.G. (2017). Haematological changes in the fish *Cyprinus carpio* exposed to a synthetic pyrethroid [Class I], Permethrin and its 25% EC. *Cur Trends Technol Sci*, 6 (5), 759-763.

48. Rao, G.S., Naik, B.K., Satyanarayana, S. and Rao, N.G. (2018). Haematological changes induced by the deltamethrin a synthetic pyrethroid technical grade and 11% EC (Decis) in the fish *Ctenopharyngodon idella* (Valenciennes). *J Inn Pharm Biol Sci*, 5 (2), 128-134.

49. Adewoye, S.O. (2010). Haematological and biochemical changes in *Clarias gariepinus* exposed to *Trephosia vogelii* extract. *Adv Appl Sci Res*, 1 (1), 74–79.

50. Jaya, S. and Singh, A. (2010). A comparative study on the piscicidal activity of synthetic pesticides and plant origin pesticides, to fish *Channa punctatus*. *World J Zool*, 5 (1), 20–24.

51. Akinrotimi, O.A., Gabriel, U.U. and Orokotan, O.O. (2013). Changes in enzymes activities of *Clarias gariepinus* brood fish exposed to anaesthetics metomidate. *Appl Ecol Environ Sci*, 1 (3), 37–40.

52. Yèhouénou, E.A.P., Azéhoun, P., Edorh, P., Van Hattum, B., Swart, K., Van Strallen, M.N. and Van Gestel, C.A.M. (2013). Health risks associated with pesticide residues in sediments, fish, and plants from the Ouémé valley in the Republic of Benin. *Arch Environ contam Toxicol*, 65, 260–265.

53. Anju T. (2012). Effect of cadmium acetate on immuno-hematological parameters in White Leg-horn chicks. *IAAST*, 3(4), 40-42.

54. Fawzia, Y.S., Sayed, G.H., Walid, S.E., Hassan, M.D., Amira, H.M. and Alaa, R.A. (2014). Protective effects of vitamin E, Selenium and Zinc supplementation on hematological and some biochemical parameters in pregnant rats exposed to cadmium. *Global J Pharmacol*, 8(4), 665-672.

55. Wang, H.J., Liu, Z.P., Jia, X.D., Chen, H. and Tan, Y. (2014). Endocrine disruption of cadmium in rats using the OECD Enhanced TG 407 Test System. *Biom Environ Sci*, 27(12), 950-959.

56. Oluwafemi, A.O., Basiru, A.A., Babatunji, E.O. and Adebola, B.O. (2014). Hematological properties of *Irvingia gabonensis* in males adult rats. *J Pharm Sci Inn*, 3(5), 434 - 436.

57. Fahim, M.A, Nemmar, A., Dhanasekaran, S., Singh, S., Shafiullah, M., Yasin, J., Zia, S. and Hasan, M.Y. (2012). Acute cadmium exposure causes systemic and thromboembolic events in mice. *Physiol Res*, 61, 73-80.

58. Hounkpatin, A.S.Y. Edorh, P.A., Guédénon, P., Alimba, C.G., Ogunkanmi, A., Dougnon, T.V., Boni, G., Aissi, K.A., Montcho, S., Loko, F., Ouazzani, N., Mandi, L., Boko, M. and Creppy, E.E. (2013). Haematological evaluation of Wistar rats exposed to chronic doses of cadmium, mercury and combined cadmium and mercury. *African J Biotechnol*., 12(23), 3731-3737.

59. Muraidharan, L. (2012). Haemato-biochemical alterations induced by chronic exposure to fenthion in *Cyprinus carpio*. *Int J Trend Res Devel*, 1(3), 19-25.

60. Parkash, J. (2016). Effect of endosulphan and dimethoate pesticides on haematological parameters of freshwater fish *Channa punctatus*. *Sci J Zool*., 4(3), 28-33.

61. Tiwari, R.K., Singh, S., Ghosh, S. and Pandey, R.S. (2017). Studies delineating the effect of chlorpyrifos on *Heteropneustes fossilis*: histopathological and hematological aspects. *Int J Recent Sci Res*, 8(4), 16934-16938.

62. Diaby V, Yapo AF, Adon AM, Yapi HF, Djama AJ and Dosso M. (2016). Biotoxicité hématologique du sulfate de cadmium chez les rats Wistar. *International Journal of Biological and Chemical Sciences*, 10(4), 1765-1772.

63. Alex, A.E and Lawrence, I.E. (2013). The effects of sub lethal concentrations of cadmium on haematological indices of two amphibians. *Int Conf Med, Biol Pharm Sci*, 4(5), 216-219.

64. IARC. (1993). Cadmium and cadmium compound. IARC Monographs on the Evaluation of Carcinogenic Risk of Chemicals to Humans. Vol. 58. Lyon, France: *International Agency for Research on Cancer*, 119-239.

65. Jelena, D., Ivana, M., Marina, N., Aleksandra, P.A., Lidija, Z., Dragan, K. and Milena, K. (2014). Acute cadmium administration to rats exerts both immunosuppressive and proinflammatory effects in spleen. *Toxicol* , 326, 96–108.

66. Gabol, K., Zaheer, K.M., Umair, A.K.M., Peer, K., Farina, F., Saima, S., Tanveer, J., Nadeem, B., Asif, I.M., Usman, A.H.M. and Muhammad, T. (2014). Induced effects of lead, chromium and cadmium on *Gallus domesticus*. *Canadian J Pure Appl Sci*, 8(3), 3035-3042.

67. Borane, V.R. (2013). Protective role of ascorbic acid on the cadmium induced changes in hematology of the freshwater fish, *Channa orientalis* (Schneider). *Adv Appl Sci Res*, 4(2), 305-308.

68. Nemcsok, J., Benedeczky, I., Boross, B., Asztalos, B. and Orban, L. (1982). In vivo effects of deltamethrin on some biochemical parameters of carp (*Cyprinus carpio* L.). *Acta Biologica Szegediensis*, 27, 9-15.

69. Mishra, J. (1983). Malathion induced hematological and biochemical changes in the Indian catfish *Heteropneustes fossilis*. *Environ Res*, 30, 393-398.

70. Sastry, K.V., Siddiqui, A.A. and Singh, S.K. (1998). Alterations in some biochemical and enzymological parameters in the snake head fish, *Channa punctatus* exposed clinically to quinalphos. *Chemosphere,* 11, 12-21.

71. Al-Otaibia, A.M., Al-Balawia, H.F.A., Ahmada, Z. and Sulimana, E.M. (2019). Toxicity bioassay and sub-lethal effects of diazinon on blood profile and histology of liver, gills and kidney of catfish, *Clarias gariepinus. Brazilian J Biol*., 79 (2), 326-336.

72. Bhatnagar, A., Cheema, N. and Yadav, A.S. (2017). Alterations in haematological and biochemical profile of freshwater fish, *Cirrhinus mrigala* (Hamilton) exposed to sub-lethal concentrations of chlorpyrifos. *Nature Environ Poll Technol*, 16(4), 1189-1194.

73. Sevcikova, M., Modra, H., Blahova, J., Dobsikova, R., Plhalova, L., Zitka, O., Hynek, D., Kizek, R., Skoric, M. and Svobodova, Z. (2016). Biochemical, haematological and oxidative stress responses of common carp (*Cyprinus carpio* L.) after sub-chronic exposure to copper. *Veterinarni Medicina*, 61, 35-50.

74. Karayakar, F., Çiftci, N., Korkmaz, C., Ay. Ö. and Cicik, B. (2017). Effects of Copper, Zinc, Lead and Cadmium on some blood parameters of *Oreochromis niloticus*. *Aquaculture Studies*, 17, 273-282.

75. Pandharikar, S. and Joshi, P. (2011). Studies on cadmium induced some biochemical alternations in air breathing fish *Clarias batrachus* (linn.). *Rev. Res*., 1 (3), 1-4.

76. Ahmad, S.I.and Gautam, R.K. (2014). Effect of organophosphate pesticide, nuvan on serum biochemical parameters of freshwater catfish *Heteropneustes fossilis* (Bloch). *Int Res J Environ Sci*, 3(10), 1-6.

77. Soufy, H., Soliman, M., El-Manakhly, E. and Gaafa, A. (2007). Some biochemical and pathological investigations on monosex Tilapia following chronic exposure to carbofuran pesticides. *Global Veterinaria*, 1(1), 45-52.

78. Ogamba, E.N., Inyang, I.R. and Azuma, I.K. (2011). Effect of paraquat dichloride on some metabolic and enzyme parameters of *Clarias gariepinus*. *Cur Res J Biol Sci*, 3(3), 186-190.

79. Zulfiqar, A., Yasmeen, R. and Ijaz, S. (2020). Effect of Malathion on blood biochemical parameters (urea and creatinine) in Nile tilapia (*Oreochromis niloticus*). *Pakistan J Sci*, 72(1), 37-42.

80. Zeid, E.A.and Khalil, A.S.A. (2014). Toxicological consequences of Di-n-Butyl- Phthalate (DBP) on health of Nile tilapia fingerlings. *Americ J Anim Vet Sci*, 9(4), 269-276.

81. Bharti, S. and Rasool, F. (2021). Analysis of the biochemical and histopathological impact of a mild dose of commercial malathion on *Channa punctatus* (Bloch) fish. *Toxicology Reports*, 8, 443-455.

82. Banaee, M., Tahery, S., Nematdoost Haghi, B., Shahafve, Sh., and Vaziriyan, M. (2019). Blood biochemical changes in common carp (*Cyprinus carpio*) upon co-exposure to titanium dioxide nanoparticles and paraquat. *Iranian J Fish Sci*, 18(2), 242 -255.

83. Nelson, D.L., and Cox, M.M. (2000). Lehninger principles of biochemistry. New York: Worth Publishers, pp. 623-658.

84. Al-asgah, N.A., Abdel-warith, A.W.A., Younis, E.M. and Allam, H.Y. (2015). Haematological and biochemical parameters and tissue accumulatioons of cadmium in *Oreochromis niloticus* exposed to various concentrations of cadmium chloride. *Saudi J Biol Sci*, 22(5), 543-550.

85. Jeney, G., Nemcsok, J., Jeney, Z.S. and Olah, J. (1991). Acute effect of sublethal ammonia concentrations on common carp (*Cyprinus carpio* L.). II. Effect of ammonia on blood plasma transminases (GOT, GPT), GDH enzyme activity and ATP value. *Aquaculture,* 104(1-2), 149-156.

86. Mekkawy, I.A.A., Mahmoud, U.M., Wassif, E.T. and Naguib, M. (2011). Effects of cadmium on some haematological and biochemical characteristics of *Oreochromis niloticus* (Linnaeus, 1758) dietary supplemented with tomato paste and vitamin E. *Fish Physiol Biochem*, 37(1), 71-84.

87. Sastry, K.V. and Sharma, K. (1980). Diazinon effect on the activities of brain enzymes from *Ophiocephalus (Channa) punctatus*. *Bul Environ Contam Toxicol*, 24(1), 326-332.

88. Agrahari, S., Pandey, K.C., and Gopal, K. (2007). Biochemical alteration induced by monocrophos in the blood plasma of fish, *Channa punctatus* (Bloch). *Pest Biochem Physiol*, 88(3), 268-272.

89. Vaglio, A. and Landriscina, C. (1999). Changes in liver enzyme activity in the teleost, *Sparus auratus* in response to cadmium intoxication. *Ecotoxicol Environ Saf*, 43(1), 111-116.

90. Palanivelu, V., Vijayavel, K., Balasubramanian, S.E. and Balasubramamian, M.P. (2005). Influence of insecticidal derivative (cartap hydrochloride) from the marine polycheate on certain enzyme systems of the fresh water fish *Oreochromis mossambicus*. *J Environ Biol*, 26(2), 191-195.