Assessment of heavy metal concentration and blood profiles of African catfish (*Clarias gariepinus*) in Sabiyal Lake, Aliero Local Government, Kebbi State, Nigeria

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Abstract

Although sabiyal wetland is supposed to sieve pollutants carried by the lake, growing human activity has caused it to deteriorate and shrink in size, reducing its ability to do so and allowing waste to flow into the lake. This makes evaluating the effects of wastewater in that lake urgently necessary. Since little is known about the use of serum and haematological assays to assess fish health in relation to heavy metals, this study is essential. This study aims to evaluate African catfish (*Clarias gariepinus*) blood profiles and heavy metal concentrations in Sabiyal Lake, Aliero, Nigeria. The study was conducted in Kebbi State's Aliero Local Government Area, specifically at Sabiyal Lake. For this investigation, five percent (5%) of the *C. gariepinus* that were caught were used. Samples of blood, gills, and bone were obtained using conventional techniques. Heavy metal analysis was done on all samples, while haematological, plasma, and serum analyses were done on the blood samples. The findings indicated that there was a huge variation in the haematological variables during the course of the research periods ($p<0.05$). Decreased plasma biochemistry ($p<0.05$) and different enzymes ($p<0.05$). The blood, gills, and bone of fish collected from Sabiyal Lake showed varying levels of heavy metal contamination. Fish collected in October had the highest concentration of lead, copper, zinc, cadmium, and iron, while fish recorded in December had the lowest concentration. The investigation found that the high concentration of heavy metals had an impact on the fish from Sabiyal Lake's haematology, biochemistry, and blood enzymes. Therefore, more study should be done to monitor and control the fish and water in Sabiyal Lake.

Keywords: Haematology, Serum, Plasma biochemistry, Heavy metals, Sabiyal Lake

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Introduction

Due to contamination from home waste, industrial effluent, agricultural runoff, oil spills, mining effluents, and unsavoury fishing methods, Nigeria's inland water bodies have seen a variety of degradations (Ndimele, 2008). Consuming fish from such contaminated water bodies puts consumers at grave risk since it destroys the ecology, the biota, and the related fishery that provide a living for fishermen (Numbere *et al*., 2023). Among these contaminants are heavy metals. Heavy metals are set of metals and metalloids that have an atomic density of more than 4 g/cm3 or five times that of water (Garbarino *et al*., 1995; Hawkes, 1997; Lenntech, 2004). Because the majority of heavy metal contaminants are not biodegradable, they remain in the biota system and bioaccumulate up the food chain, which has caused concern for aquatic ecologists (Ndimele *et al*., 2009; Sharma *et al*., 2024). Natural processes or naturally occurring deposits and human activity are the two main sources of contamination that lead to the build-up of heavy metals in aquatic environments (Jagaba *et al*., 2024).

Particularly in the past ten years, studies on heavy metal levels in fish, lakes, and rivers have drawn significant attention from the environmental community (Fernandes *et al*., 2008; Pote *et al*., 2008). Heavy metals depict clear understanding of the level of pollution of water body, which is why water is frequently used as a pollution indicator. Nonetheless, the concentration of processes occurring within the catchments determines the variance in parameter concentration. The colour, odour, density, taste, and temperature characteristics of the water can all alter due to variations in physico-chemical parameters and heavy metal concentration, which can negatively impact fish health (Dippong *et al*., 2024).

Almost all household trash is dumped into Sabiyal Lake without being properly treated, leading to a build-up of harmful substances in the biota and the entire system, resulting into a loss of dissolved oxygen in the water, and other water thresholds. Although the lake's water is a valuable resource for the region, the water below is extremely muddy and discoloured, and the surrounding flora looks burnt. It is utilized for construction, cleaning, irrigation, animal and bird drinking, leisure, human consumption, and transportation. It is well recognized that wetlands serve as organic filters for pollutants and nutrients coming from the catchment region, maintaining the quality of the water. Regrettably, Sabiyal wetland is expected to bio-filtrate contaminants from the lake but human anthropogenic activities have reduced its filtration capacity and cleaning potential thus allowing waste to seep into the lake. This creates an urgent need to assess the impact of wastewater in that lake.

Serum, plasma biochemistry, and haematology are useful metrics for evaluating the health of fish. The fish's health is being determined by these analyses (Ferreira and Avenant-

Oldewage, 2013). Measurement of haematological variables has become a useful tools and indicator of fish health status and can be employed to examine the fish's stress reaction (Soivio and Oikan, 1976; Chen *et al*., 2023). This study is necessary since there is a lack of knowledge on the application of serum and haematological assays to evaluate fish health in relation to heavy metals. It is essential to assess the fish's health using blood and serum indices because it appears healthy and won't be turned down for eating. The aim of this study is to assessment heavy metal concentration and blood profiles of African catfish (*C. gariepinus*) in Sabiyal Lake, Aliero Local Government, Kebbi State, Nigeria

Materials and methods

Description of the study area

The research was carried out in Sabiyal Lake, Aliero Local Government Area, Kebbi State. Sabiyal Lake is a eutropic, perennial standing fresh water body, located between latitude 12° 44' 23" North and longitude 4° 30' 54" East. The Lake is surrounded by Sabiyal, Kashinzama, Laga, Tari, Kambaza, Bami Mairuwa, and Kyara villages, about 10km away from Aliero town, Aliero Local Government Area, Kebbi State.

Ethical statement

The protocol of the study was subjected to ethical consideration and was approved by Animal Care Use and Research Ethics Committee, University of Ibadan, Ibadan, Oyo State, Nigeria, with reference number UI-ACUREC/App/03/2017/008.

Fish sample collection

Fish were collected for three months by fisherman at Sabiyel Lake, five percent (5%) of the *C. gariepinus* were sent to the KSUSTA Fisheries Laboratory for examination. A digital Scout Pro scale (Model: M1207) was used to weigh the fish, and a metre rule calibrated in centimetres was used to measure the fish's overall length. Using a drag net, 436 (about 5% of the total) *C. gariepinus* were taken in Sabiyel Lake between October and December of 2023. The fish had a total length of 28–34 cm (mean=31 cm) and weighed between 149–154 g (mean=152 g). Using genital papillae frequently utilized in *C. gariepinus*, the sex differential of African catfish was carried out (Adeshina *et al*., 2016). The well labeled fish samples transported to the laboratory at -4^{o}C. The samples were washed under running water and stored in deep at -20^{o}C until analyzed.

Blood sample collection

Using a sterile syringe and a disposable hypodermic needle with a gauge of 21 was used to take blood samples from the caudal vein of each of the 436 fish. The blood drawn from caudal vein for haematology parameters (packed cell volume [PCV]; haemoglobin [Hb]; red blood cell [RBC]; white blood cell [WBC]; platelets; lymphocytes; heterocytes; monocytes; eosinophils; basophils; mean cell haemoglobin [MCH], the average of Hb in pictograms of a simple RBC was determined using

MCH (pg) = $\frac{Hb \times 10}{RBC}$; mean cell haemoglobin concentration [MCHC], the percentage of Hb of packed RBC was determined using MCHC (g/dL)=$\frac{Hb}{PCV} \times 100$; mean cell volume [MCV], the average volume of a single cell was determined using MCV (fL)= $\frac{PVC \times 10}{RBC}$. The plasma (total protein [TP], albumin [ALB], globulin), and blood enzyme analysis (aspartate aminotransferase [AST], alanine aminotransferase [ALT], and alkaline phosphatase [ALP] plasma biochemistry, and serum investigations after being gently transferred into a lithium heparin anticoagulant tube at room temperature (Abdel-Tawwab *et al*. 2018; Adeshina *et al*. 2019). For approximately five minutes, the slides were immersed in a staining jar filled with recently diluted May-Grunwald's stain using an equivalent volume of buffered water. The slides were moved into a second staining jar and left for 15 minutes with freshly diluted Giemsa stain diluted with nine volumes of buffered water. The slides were moved into a second stain jar filled with buffered water, quickly cleaned in three different water changes, and then allowed to stand in the buffered water for five minutes to allow for differentiation. After that, they were allowed to air dry and were mounted on a light microscope.

Determination of heavy metals in C. gariepinus

For 48 hours, the fish samples were dried in an oven set to 75°C. Using a sterile stainless steel knife, the organs (gills and bones) were removed, and a porcelain mortar was used to grind them into a fine powder. Each homogenised fish sample weighed two grammes and was placed in a different beaker. The beakers were filled with a 3:1 concentration of HNO_3 and H_2O_2, which was then allowed to digest for 20 minutes at 120°C on a hot plate in a fume chamber. The samples were diluted up to 25 mL mark with deionized water before being examined for trace metal concentration (Singh *et al*., 2023; Muhammad *et al*., 2024).

Statistical analysis

Using the IBM Statistical Package for the Social Sciences (SPSS) version 20, the acquired data were analysed using descriptive statistics (percentage), one-way analysis of variance (ANOVA), and means were separated using the Duncan Multiple Range Test.

Results

The haematological characteristics of *C. gariepinus* in Sabiyal Lake, Aliero, Kebbi State, Nigeria are displayed in Table 1. Throughout the study periods, there was a noteworthy variation in the haematological parameters (p<0.05). Significant decreases were observed in PCV, haemoglobin, lymphocytes, eosinophils, MCH, and MCV as December approached. In contrast, December saw a rise in RBC, WBC, neutrophils, basophils, and MCHC.

Throughout the investigation, *C. gariepinus* plasma biochemistry significantly decreased (p<0.05; Table 2). October had the highest levels of total protein, albumin, globulin, and A:G,

whereas December had the lowest levels of these substances. The blood enzymes of *C. gariepinus* in Sabiyal Lake are shown in Table 3. The fish's blood enzyme levels differed significantly (p<0.05; Table 3). Fish sampled in December had significantly greater levels of ALT, AST, and ALP than fish sampled in October, which had the lowest values of these markers.

Table 1: Haematology profiles of *C. gariepinus* from Sabiyal Lake, Aliero, Kebbi State, Nigeria.

Parameters	Periods			Pooled SEM	P values	
	October	November	December		Liner	Quadratic
Packed cell volume (PCV, %)	41.3 [a]	20.6 [c]	28.8 [b]	4.182	<0.001	<0.001
Haemoglobin (Hb, g/dL)	12.53 [a]	6.85 [b]	12.42 [a]	3.109	0.001	<0.001
Red blood cells (RBC, x 10^6/µL)	3.31 [b]	2.11 [c]	3.62 [a]	0.127	0.001	0.002
White blood cells (WBC, x 10^6/µL)	12.65 [b]	13.89 [b]	18.11 [a]	9.378	<0.001	<0.001
Platelets (x 10^6/µL)	126.04 [b]	96.25 [c]	284.16 [a]	12.759	<0.001	<0.001
Lymphocytes (%)	70.3 [a]	53.9 [c]	57.3 [b]	4.218	0.001	<0.001
Neutrophils (%)	20.6 [c]	37.3 [a]	33.2b[a]	3.061	<0.001	<0.001
Monocytes (%)	3.9 [b]	3.8 [b]	4.8 [a]	0.941	0.001	0.004
Eosinophils (%)	3.9 [a]	3.6 [b]	3.2 [c]	0.022	0.001	0.003
Basophils (%)	1.3 [a]	1.4 [a]	1.5 [a]	0.008	0.205	0.311
MCH (pg)	37.85 [a]	32.46 [c]	34.31 [b]	6.286	<0.001	<0.001
MCHC (d/dL)	30.32 [c]	33.20 [b]	43.18 [a]	9.452	<0.001	<0.001
MCV (fL)	124.86 [a]	97.77 [b]	79.45 [b]	8.895	<0.001	<0.001

Means ± standard errors within a row with different superscript are significantly different (p<0.05).

Table 2: Plasma biochemistry parameters of *C. gariepinus* from Sabiyal Lake, Aliero, Kebbi State, Nigeria.

Parameters	Periods			Pooled SEM	P values	
	October	November	December		Liner	Quadratic
Total protein (d/dL)	3.42 [a]	3.38 [b]	3.13 [c]	0.231	<0.001	<0.001
Albumin (g/dL)	2.67 [a]	2.15 [b]	1.37 [c]	0.021	<0.001	<0.001
Globulin (g/dL)	4.14 [a]	3.86 [a]	2.29 [b]	0.593	<0.001	<0.001
Albumin:Globulin (A:G)	0.64 [a]	0.56 [b]	0.60 [ab]	0.001	<0.001	<0.001

Means ± standard errors within a row with different superscript are significantly different (p<0.05).

Table 3: Blood enzymes of *C. gariepinus* from Sabiyal Lake, Aliero, Kebbi State, Nigeria.

Parameters	Periods			Pooled SE	*P* values	
	October	November	December		Liner	Quadratic
ALT (IU/L)	25.23 [b]	28.21 [a]	29.07 [a]	9.236	<0.001	<0.001
AST (IU/L)	21.06 [c]	24.11 [b]	25.54 [a]	4.216	<0.001	<0.001
ALP (IU/L)	29.47 [c]	32.11 [b]	38.23 [a]	3.641	<0.001	<0.001

Means ± standard errors within a row with different superscript are significantly different (p<0.05).

The concentration of heavy metals in *C. gariepinus* from Sabiyal Lake is depicted in Figure 1. The amounts of heavy metals found in the blood, gills, and bone of fish taken from Sabiyal Lake varied considerably.

The highest concentration of lead, copper, zinc, cadmium, and iron was found in fish taken in October, while the lowest concentration was found in fish recorded in December. The times during which the fish were taken were exactly correlated with increases in the load of heavy metals.

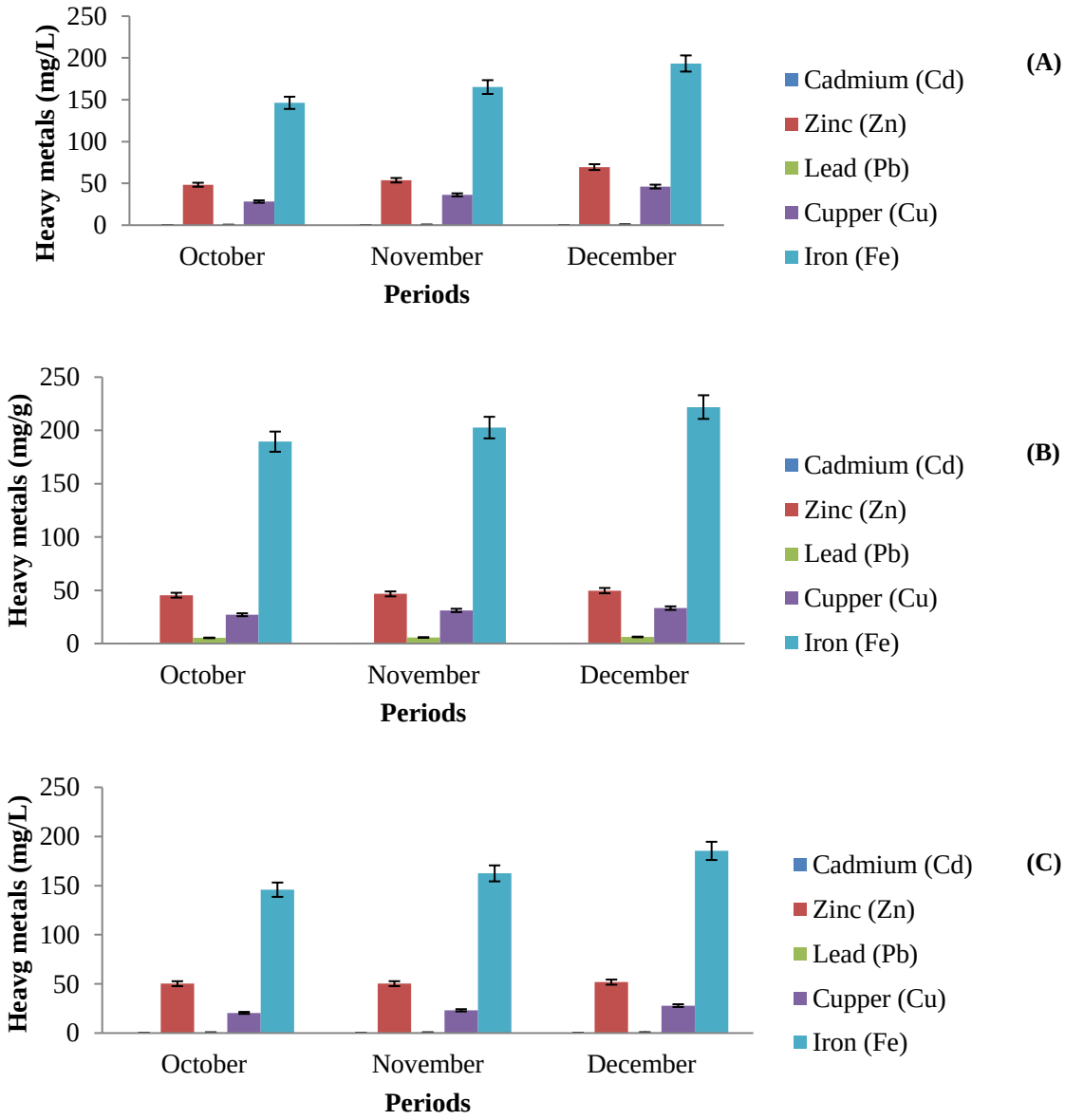

Figure 1:4 Heavy metals concentrations (mg/L) in the blood of *C. gariepinus* from Sabiyal Lake, Aliero, Kebbi State, Nigeria (A) = Blood samples, (B) = Gill samples, (C) = Bone samples.

Discussion

Variations in the fish's blood serum and haematological parameters show how healthy they are. However, utilising blood biochemistry to evaluate fish conditions has proven difficult due to reference intervals for different fish species. Fish sampled in December had lower levels of PCV, Hb, RBC, and WBC, which may indicate that they are less resilient to illness and have been exposed to heavy metals for longer.

Aquatic life forms can readily absorb heavy metals, which can accumulate in larger concentrations (Omoregie *et al*., 2002). Fish can bioaccumulate heavy metals in their muscles and organs at varying rates based on the metals they consume and the settings in which they live (Phillips and Rainbow, 1994; Jamil Emon *et al*., 2023). Rainbow *et al*. (1990) reported that there are significant differences in the fish's capacity to detoxify specific metals and their rate of buildup. This may help to explain why different samples of the *C. gariepinus* taken in December had varying concentrations of heavy metals. The fact that the mean concentrations of heavy metals in the *C. gariepinus* obtained in December were higher than those from October lends credence to the idea of fish bioaccumulation. This outcome is in line with Murphy's (1978) research, which shown that edible fish flesh accumulated more metals. Furthermore, gill samples had the highest quantity of heavy metals, which is in line with the Zn content of the *C. gariepinus* samples that were collected from the Asa dam. This is corroborated by the findings of Titilayo and Olufemi (2014), and, who reported a significant bioaccumulation of metals in fish flesh. This finding may be related to the dung effluents that runoff water brings into rivers (Jamil Emon *et al*., 2023). Zinc is a byproduct of animal nutrition and is easily concentrated in the excretions of mature animals, which produce 7–20 mg of zinc daily on average, according to Forstner and Prosi (1979) and Targuma *et al*. (2018). There was a discernible increase in copper in the Sabiyal River sample.

The sample from the Sabiyal River had far more copper bioaccumulation in its gills than in its blood or bone. Due to copper's binding to metallothionein in the gill, an organ used for gaseous exchange, the liver has a high concentration of copper (Chen *et al*., 2024). Even though it's necessary for a healthy diet, over consumption of copper can be hazardous. The harmful toxicity is largely attributed to its cupric (Cu^{2+}) forms (Olaifa *et al*., 2004; Dahunsi *et al*., 2012; El Sherif, 2017).

Conclusion

The study concluded that haematology, biochemistry and blood enzymes of the fish from Sabiyal Lake were affected due to high heavy metal concentration. Therefore, further research should be carried control and management the fish and water from the Sabiyal Lake.

Funding

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Compliance with ethical standards

The protocol of the study was subjected to ethical consideration and was approved by Animal Care Use and Research Ethics Committee, University of Ibadan, Ibadan, Oyo State, Nigeria, with reference number UI-ACUREC/App/03/2017/008.

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