

THE ACUTE TOXICITY OF IRON ON THE BEHAVIOUR OF *Oreochromis niloticus* FINGERLINGS IN KADUNA NIGERIA

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Abstract

Industrial discharges containing toxic and hazardous substances, including heavy metals contribute hugely to aquatic ecosystem. The study was conducted in the Department of Biological Sciences laboratory of Kaduna State University, Nigeria using described methods for the Examination of Water and Waste water APHA. The acute toxicity of iron to *O. niloticus* and the LC_{50} value of 1.05mgL^{-1} were determined by using the regression analysis and probit analysis curve. Fish were exposed to selected concentration of Iron (0.016, 0.024 and 0.032mg/L). Fish displayed random and haphazard swimming behaviour, air gulping then a period of quiescence and finally death. Mortality data were determined after 24, 48, 72 and 96 hours validity period. The study showed that heavy metals can be hazardous to fish and indirectly to humans through food chain.

KEYWORD: Acute toxicity, Behaviour, Iron, *Oreochromis niloticus* (Trewavas)

1.0 INTRODUCTION

Heavy metals are natural trace components of the aquatic environment, but their levels have been increased due to domestic, industrial and agricultural movements. It poses greatest threat to the health of Indian ecosystem (Rani *et al.*, 2001; Desai *et al.*, 2002). Discharge of heavy metals into the aquatic environment can change both aquatic species diversity and ecosystems, due to their toxicity and incremental behavior. Aquatic organisms including fish collect metals many

times higher than present in water thus causing an adverse effect on the fish metabolism (Madhusudan *et al.*, 2003). Studies proved that, fish subjected to metal toxicity shows that biochemical and histopathological alterations in different target tissues like gills, liver and kidney. Histopathological lesions and increase in size of gills were reported in fishes exposed to heavy metals (Devlin, 2006; Gupta and Ashwini, 2006). Necrosis and rupture of gills of *Labeorohita* on exposure to Copper was reported by Kalele and Dhande (2006) Bioaccumulation of zinc in kidney of fish *Channapunctatus* (Bloch) have been studied by Gupta and Srivastava, (2006). Impact of cadmium on the biochemical constituents of *Oreochromis mossambicus* was studied by Hameed *et al.*, (2006).

Iron is the fourth most abundant element by weight in the earth's crust, and is often a major constituent of soils (especially clays). It has an atomic number of 26 and atomic weight of 55.85 and exists primarily (91.7%) as ^{56}Fe but has one radioactive isotope (^{54}Fe , 5.8% of total mass) and two stable isotopes ^{57}Fe (2.2%) and ^{58}Fe (0.3%) Moore, (1991) reviews iron as an inorganic contaminant in water and summarize relevant production, chemistry and toxicity.

Iron is an absolute requirement for all forms of life. Importance of iron is especially notable in biogeochemical processes because of its unique ability to serve as both an electron donor and acceptor and thus can play an important role in metabolic processes of many organisms. Iron can also be potentially toxic at high concentrations. Iron's ability to donate and accept electrons means that if iron is free within the cell, it can catalyze the conversion of hydrogen peroxide into free radicals. Free radicals can cause damage to a wide variety of cellular structures, and ultimately kill the cell (Crichton *et al.* 2002). To prevent that kind of damage, life forms have evolved a biochemical protection mechanism by binding the iron atoms to proteins.

This allows the cells to use the benefits of iron, but also limit its ability to do harm (Andrews, 1999).

The problems of environmental pollution and its harmful effect on aquatic biota, including fish is receiving focus during the last few decades (Jagadeesan *et al.*, 2001; Zikic and Stajn, 2001). Industrial discharges containing toxic and hazardous substances, including heavy metals contribute hugely to aquatic ecosystem (Ghemet *et al.*, 2001; Woodling *et al.*, 2001).

Oreochromis niloticus is of economic importance, hardy and nutritious. It is also demanded in Nigeria hence it was used for this study.

2.0 MATERIALS AND METHODS

The method employed in this experiment is based on recommended method for the test of acute toxicity of pollutant to fish described by Standard Method of Examination of Water and Waste Water (APHA, 1985 and APHA, 1998).

2.1 Selection of Fish

It has been suggested by Fish Toxicity Testing Framework (2012) that economically local fish should be used in toxicity assay. *Oreochromis niloticus* of the family Cichlidae and genus Tilapia is of high commercial value and are very common in Nigerian fresh waters.

They are of high nutritive value and data of their nutritive value have been well documented by (Eyo *et al.*, 2000). The government of Nigeria is beginning to be more and more interested in fish farming and farmers are being encouraged to set up both small and large scale fish farms the most common fish which can be kept in captivity and grow to table size within a short time is Tilapia. On the basis of availability, commercial and ecological important *Oreochromis niloticus* was chosen for this study.

2.2 Site Determination

This Experiment was carried out in the zoology laboratory of the Department of Biological Sciences, Faculty of Science, Kaduna State University, Nigeria.

2.3 Collection of Fish

One hundred and fifty fingerlings of *Oreochromis niloticus* were collected from the local fish market of Gen. Hassan Usman Park, GAMJI Gates Kaduna north, Kaduna, Nigeria and were transported in an aquarium containing 25 liters of water to the Zoology laboratory of the Department of Biological Sciences Kaduna State University, Kaduna. The fish were distributed into 8 aquaria of length and breadth (40×25 cm) and 25 litres capacity

2.4 EXPERIMENTAL DESIGN

Eighty fish of 7.3-9.0g weight and size 5-8cm were acclimatized for one week in 8 aquaria of 25×20 size using underground water (borehole).

2.5 Formulated Diet and Feeding

The fish were fed with compound diet to satiation twice daily during acclimatization.

Feeding of the fish was stopped 2 days prior to the experiment to avoid change in the toxicity of the water due to excretory products.

2.6 Acute Toxicity Assay

Eighty fish of average weight 7.48 and average size 7.14 were distributed in eight small aquaria, ten fish to each aquarium.

Three aquaria contained the experimental samples, while three aquaria served as replicates with two aquariums serving as control to the experimental sample and the replicates respectively.

The fish samples and replicate were exposed to three iron solution concentrations (0.016, 0.024 and 0.032 mg/L) from the stock solution of FeCl₃.

2.7 Randomization

The fingerlings were properly distributed into the eight aquaria at random as described by Finney (1964) and Liang-ping Hu (2011).

2.8 Observation and Treatment of Data

After the introduction of iron salt, the fish were observed in the first 60 minutes, 24, 48, 72 and 96 hours validity period.

Mortality in each aquaria were carefully observed, recorded and dead fish were removed within the period of the experiment.

2.9 Statistical Analysis

The survival time in each concentration of the iron observed was recorded and the LC_{50} value were calculated from the regression line drawn according to (Finney, 1964, 1971; Hubert, 1984 and Babatunde 2014). The effects of the exposure were tested with one way ANOVA.

3.0 RESULTS

3.1 Water parameters

Temperature	-----	21.40-24°C
pH	-----	6.2-7.4
Dissolve Oxygen	-----	5.8-8.4 mgL ⁻¹
Water Hardness	-----	20-28 mgL ⁻¹ CaCO ₃
Alkalinity	-----	22-26mg CaCO ₃ L ⁻¹
BOD	-----	5.34-6.01mg/ml

3.2 Behavioral Observations

Symptoms of toxicities in the behaviour of the fish were clear indications that iron has adverse sub-acute effect on the fish. The fish showed some level of agitation when the toxicants were introduced then there was haphazard swimming behaviour rapid movement of the fin and operculum (figure 4 and 6) before they all began to show some level of weakness, loss of equilibrium, air gulping, period of quiescence and finally death.

3.2 Acute Toxicity Test

The percentage mortality trend in the toxicity of iron is well recorded in Table1 below. Highest mortality was recorded in the highest concentration (0.032mg/l) at the 24 h for the Test sample and replicate but there was no mortality recorded in the control samples and replicates (Table1).

Table 1: Percentage Mortality of *O. niloticus* exposed to various concentration of Iron for 96h

Concentrations	Mortality number	Mortality rate	Mortality percentage
0.000	0	0/10	0%
0.016	2	2/10	20%
0.024	10	10/10	100%
0.032	10	10/10	100%

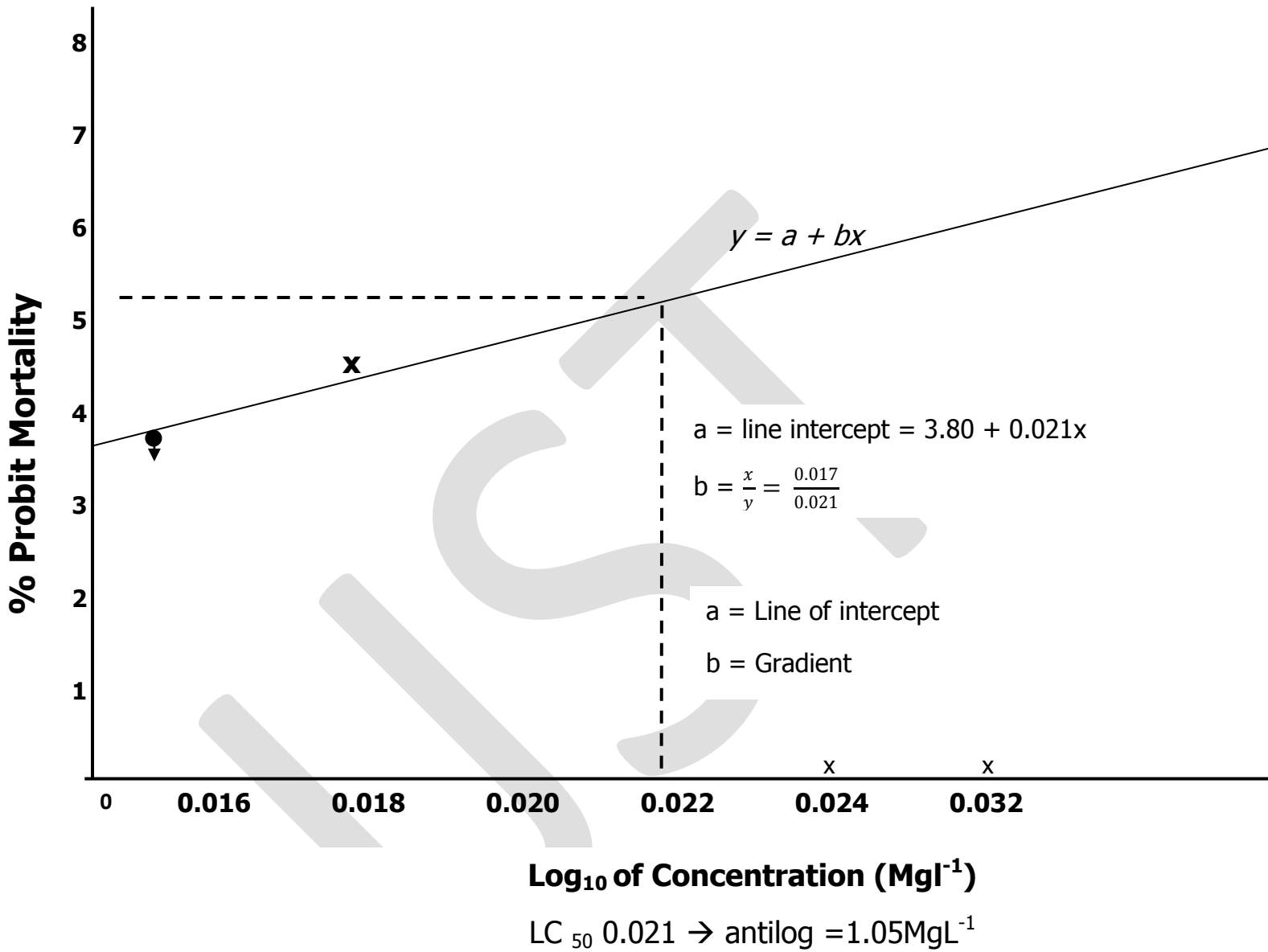


Figure 1:

Linear Relationship between Probit Mortality (%) and Log₁₀

Concentration (Mgl⁻¹) of *O. niloticus* exposed to various concentrations of Iron

Table 2: Acute Toxicity of Iron to *O. niloticus* Regression Analysis for 96h LC50

	Coefficients	t-value	Sig.
(Constant)	0.515	0.585	0.618
Concentration	183.357	4.491	0.046
R Square	0.910		
Adjusted R-Square	0.865		
S.E of Estimate	0.96607		
Durbin-Watson	1.962		
F-Statistics	20.173 (0.046)		

The above table shows the summary of the multiple regression analysis conducted to estimate how Concentration (LC 50) of Iron affects the mortality rate of *O. niloticus*. The Constant of the equation is 0.515 – this is the mortality rate if concentration of iron is zero, it has a t-value of 0.585 which is insignificant at the 5% level of significance.

It can be seen from the table that concentration of Iron has a coefficient value of 183.357; this means that if the value of concentration is increased by 1 unit, it will lead to a corresponding increase of 183.357 units in mortality rate, having a t-value of 4.491 which is statistically significant because the p-value is 0.046 which is less than 0.05. Based on this result, we can therefore conclude that concentration of iron has a very large and has significant impact on mortality rate.

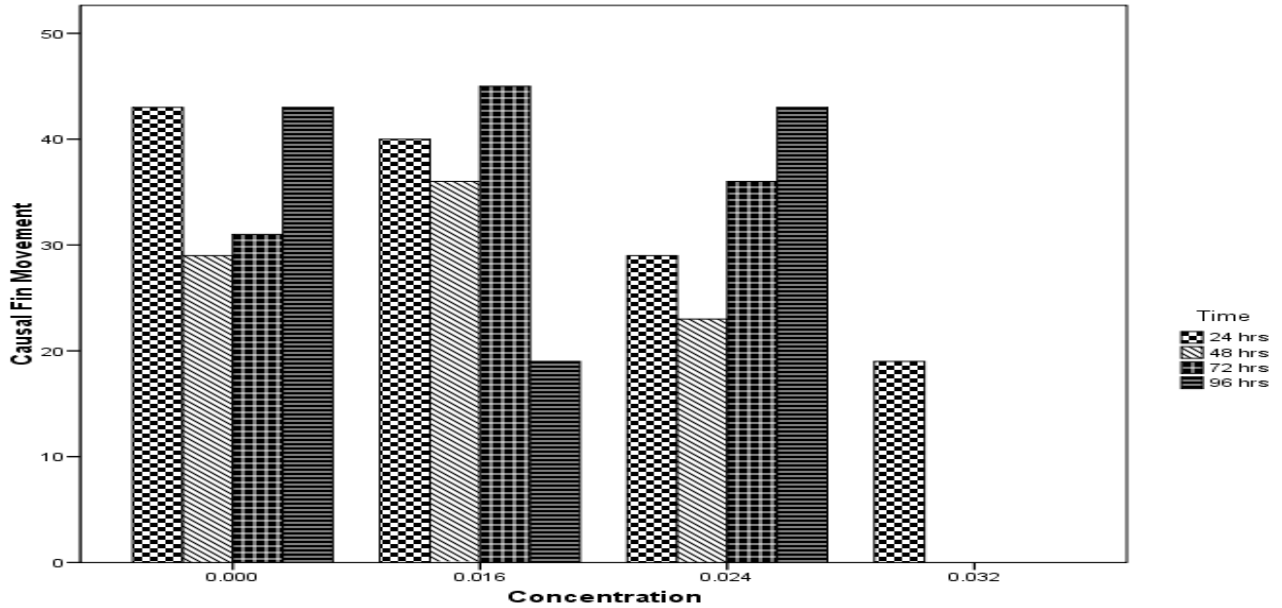


Figure 2: Caudal fin movement of *O. niloticus* at different iron concentration

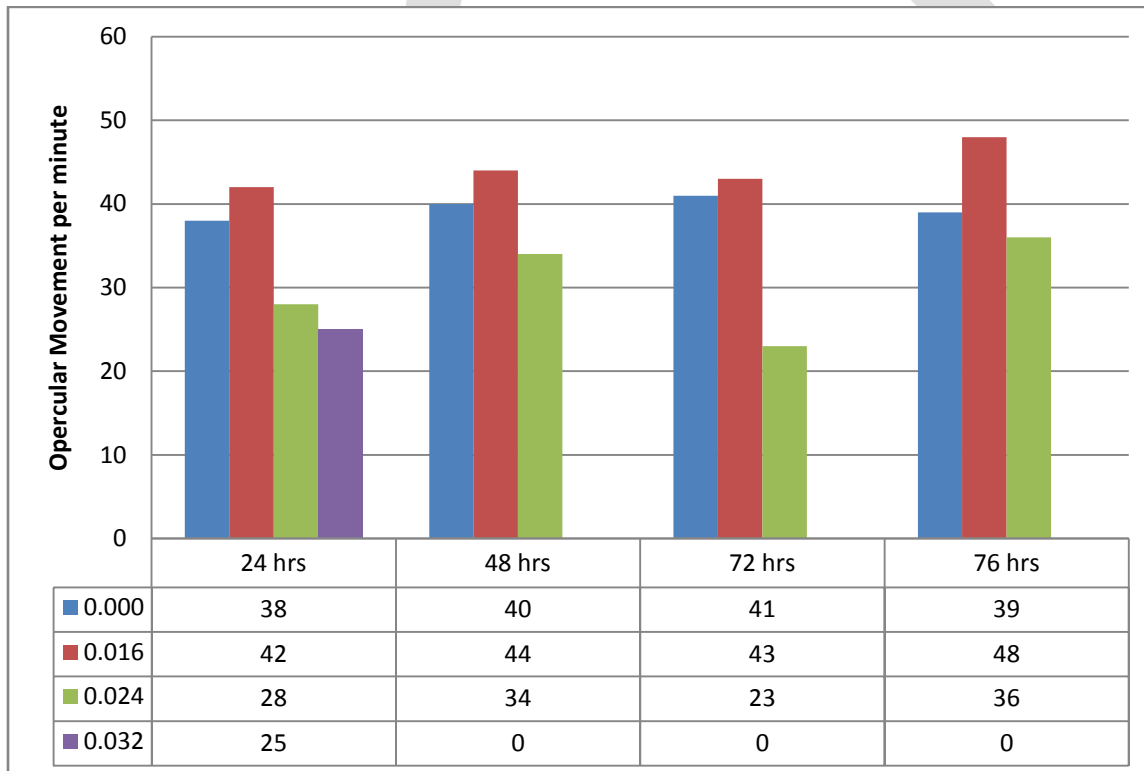


Figure 3: Opercular Movement per minute

4.0 DISCUSSION

4.1 Acute Toxicity Test

The 24, 48, 72 and 96h LC₅₀ which is 1.05mg/L was calculated using probit analysis according to Linchfield and Wilcoxon (1949). Expressing the result in Log- probit transformation were first used by Fry *et al* (1946) for testing lethal temperatures and concentrations and has since been used for assays of pollutants and toxicants. It is used in paraquat toxicity to *O. niloticus* by (Babatunde *et al.*, 2014).

In this study, the formations of precipitate by Iron on the lowest concentration was as result of aeration and alteration of pH value to about 5.0 or lower than that, making ferrous Iron to oxidize into ferric Iron due slower oxidation rate Barnes *et al* (1983). Other factors could be as a result of the reactions between hard water and the component of the plastic aquarium used for the experiment

Water hardness also affects the LC₅₀ value obtained in this study. It has been reported presently that water hardness (calcium content) affects the fish behaviour as a result of low oxygen content in the test water rather than the Iron speciation and therefore its effect may be obtained in physiological terms (Lauren and McDonald, 1985).

4.2 Behaviour

At lower concentration of iron mortality was minimum, but abnormal behaviour was observed in the test samples because concentration which do not produce immediate mortality maybe found to have a considerable effect on the behaviour, which may include feeding,

mating, courtship, migration and adaptability to environmental factors may also affect fish production.

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