

Assessment of some heavy metals and total hydrocarbons in *Clarias gariepinus* fish of Osse River, Edo State, Nigeria

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Abstract: The sequence of the heavy metals (Fe, Mn, Zn, Cu, Pb, Cd and Cr) and total hydrocarbons (THC) in the water and tissues (gills, intestine and muscle) of an abundant and economically significant fish (*Clarias gariepinus*) harvested from Osse River were investigated from April, 2014 to September, 2015. Water and fish samples were collected from 4(four) chosen stations and were analysed for the levels of heavy metals and total hydrocarbons. The ecological risk of the study area and health risk indices of the fishes were assessed. Generally, accumulation of heavy metals and total hydrocarbons in the fish was gills > intestine > muscles (anomalous sequence). The sequence may pose some threats to the health of the consumers. Iron, zinc, copper, cadmium and THC showed significant health risk indices in the fish despite their insignificant ecological risk indices. Further in-depth study on the actual levels of risks to consumers is recommended.

Keywords: Bioaccumulation factor, ecological risk indices, health risk indices, *Clarias gariepinus*

I. Introduction

Heavy metals are persistent, bio-accumulative and toxic micropollutants (PBTs). Though hydrocarbons are not referred to as PBTs; however their eco-physiological toxicity is also noteworthy. Aquatic environments receive heavy metals and hydrocarbons either through biogeochemical processes (natural sources) or anthropogenic activities (human-caused). The major points of interest are the anthropogenic sources e.g. oil spill during crude oil exploration activities. In the event of oil spill, petrogenic heavy metals and petroleum hydrocarbon are released into the aquatic environment, incorporated into the aquatic food chain and they are biomagnified through alimentation, from one trophic strata to the higher; from bottom to the top of biomasses pyramids. This causes disruption of the delicate aquatic ecological equilibrium; culminating in several eco-physiological hazards such as decreased biodiversity of aquatic organisms (Ashraj, 2005; Vosyliene and Jankaite, 2006), sub-lethal effects on finfish and shellfish, and clinical poisoning to the consumers. The fish are the most susceptible group of aquatic fauna to oil spill (Hariprasad and Dayananda, 2013); due to the vulnerability of their niche i.e. fish are at the top of the aquatic food chain; hence they have high tendency to concentrate heavy metals and hydrocarbon from organisms at lower trophic levels.

Indiscriminate consumption of unscreened food items is a major contributing factor to the relatively low average life expectancy in Nigeria which is currently 52.62 years in 2014 (CIA, 2016); particularly due to consumption of contaminated finfish and shellfish from polluted rivers mainly in the Niger Delta areas of Nigeria. Various anthropogenic activities such crude oil exploration and production, agricultural practices, laundering, dumping of domestic solid wastes etc. release pollutants into Osse River and the aquatic fauna therein are liable to accumulate these chemicals; which finally get to consumers of the fish through consumption. Fishes (mainly *Clarias gariepinus*) of Osse River are major source of their protein requirements and income generation through trade. It is therefore imperative to subject the fishes to standard screening in order to ascertain the levels of risk the allocthonous chemicals (heavy metals and total hydrocarbon) pose to the environment and the consumers of the fish. This stresses the need for risk characterization; which is the link between risk assessment and risk management. However, the risk indices are function of the octanol/ water partition coefficients or bioaccumulation factors of the chemicals in the tissues of the fishes.

Bio-accumulation of a dissolved contaminant refers to the increased contaminant concentration within an organism; which results from the passive partitioning of the contaminant between the aqueous phase and the lipid compartment of the organism. The extent of bioaccumulation is measured by the bioaccumulation factor (BAF); which is the ratio of the wet weight concentration of a contaminant in an organism and the concentration in the ambient water. Understanding the dynamic process of bio-accumulation is very essential in protecting human beings and other organisms from the adverse effects of contaminant exposure. It is also an effective tool

in regulation of contaminants such heavy metals and total hydrocarbon (OECD, 2003). The aim of the study was to evaluate the ecological risk quotients of the study area and 14 health risk indices of *Clarias gariepinus*.

II. Materials And Methods

2.1. Study Area

The study area is a stretch of Osse River; between Benin River and Ughoton streams. The river flows in south westerly direction through Nikorowa, Ekehuan, Gelegele, and Izedema communities. It is a fresh oligotrophic (Imoobe and Adeyinka, 2009) lotic freshwater, with a thick vegetation canopy along most part of its bank. It is located in the Ovia North- East local of area of Edo State; in the tropical rainforest belt of southern Nigeria. The river lies between 5°16'40" E and 5°23'20" E; 6°2'0" N and 6°14'0" N. A detailed description of the hydrological and drainage, climatic and catchment edaphic features of Osse River has been documented (Ogbeibu and Omoigberale, 2005; Omoigberale and Ogbeibu, 2007).

Four (4) stations were designated at strategic locations along the course of the river; for the purpose of this research. Station 1 (Control station) at Nikorowa, Station 2 (Ekehuan), Station 3 (Gelegele) and Station 4 (Izedema) were chosen (Fig. 1). Distinguished anthropogenic activities take place at strategic sections of the river; which were chosen as stations. There was minimal or negligible activity at Nikorowa (Station 1; control 8 station). Innumerable vessels of locally refined diesel were transported to and stored at 9 the bank of Ekehuan section (Station 2); 4,135 metres downstream from Station 1. Dubril Oil Company carries out oil exploration and production activities at the Gelegele section (Station 3); 4, 441 metres downstream from Station 2. While immense lumbering activities take place at the bank of the Izedema section (Station 4) of the river; 1, 400 metres downstream from Station 3.

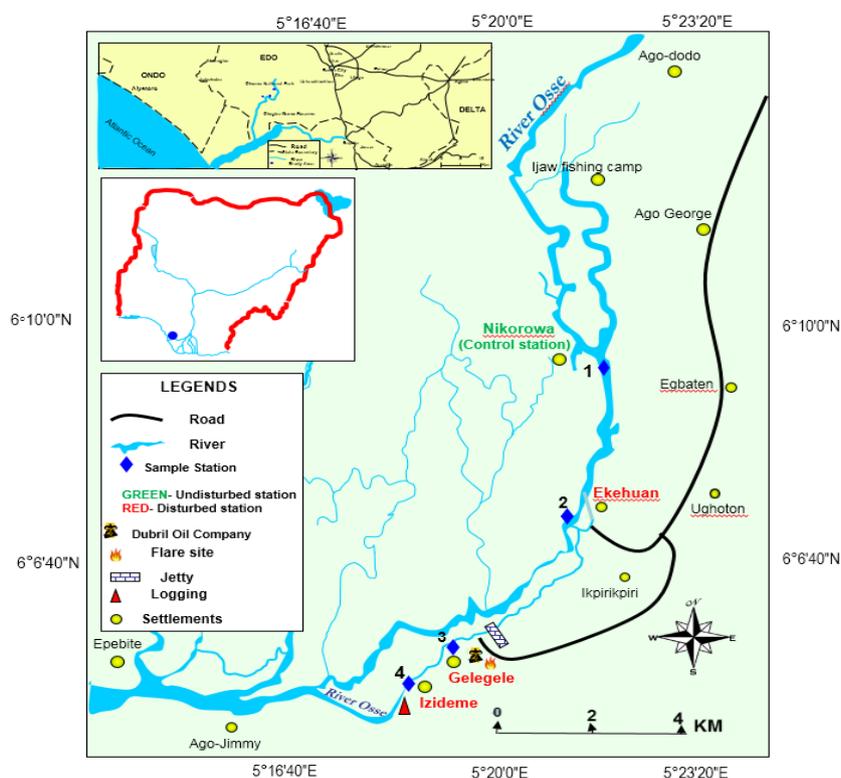


Figure 1: The Study Area

2.2. Collection and analysis of samples

For a period of 18 months (April, 2013 to September, 2014) water samples were collected at all stations using standard methods. The pH of water was taken and recorded *in-situ* using a WTW water sampler probe. All samples were transported immediately to the laboratory for analysis of the physico-chemical variables which were recorded in mg/L; using standard methods (APHA, 1998). Total petroleum hydrocarbon in water samples were analyzed using an infrared spectrophotometer (IR-Spec) at a wavelength of about 3.425 μ m.

Six (6) replicates of *Clarias gariepinus* samples were collected across the stretch of the river using fishing nets, baskets and hooks. They were preserved in ice coolers and taken to the laboratory for proper identification using standard references (Olaosebikan and Raji, 1998; and Idodo-Umeh, 2003). The gills, intestine and muscles of the fish were collected in sterile sample bottles, labeled and preserved in the freezer for

digestion and analysis of heavy metals and total hydrocarbons. 0.8 grams of each fish tissue sample (gills, intestine and muscle) was dried at 105 ° C to a constant weight. Each dried sample was ground, using porcelain mortar and pestle. The ground fish tissues were transferred to a porcelain basin and put into a Thermicon P muffle furnace at a temperature of 550 ° C for 4 hrs. Samples were digested with tri-acid 8 mixture (HNO₃:HClO₄:H₂SO₄ = 10:4:1) at a rate of 5 ml per 0.5 g of sample and was placed on a hot plate at 100 ° C temperature. Digestion continued until the liquor became 10 clear. The digested liquor was filtered through Whatmann 541 filter paper and diluted to 25 ml with distilled water of the element in the sample solution times as additional factor in µg/g dry weight. Metals (Mn, Zn, Cu, Pd, Cd, Cr; except Fe) were determined using Atomic Absorption Spectrophotometer (UNICAM 929); as illustrated by APHA (1998). Iron concentration was determined using Orthophenanthroline method; as illustrated by Oguzie and Izevbigie (2009).

2.3. Statistical analysis

The obtained water and specimen data were analysed using SPSS package (version 19.0) and the descriptive statistics were expressed as mean ± standard deviation and range; using one way analysis of variance (ANOVA) to test for the significant difference among the groups at probability level of 0.05. Furthermore, Duncan multiple range test was employed in ascertaining the actual locations of the significant differences; still 22 maintaining probability level of 0.05.

2.3.1. Risk characterization

Health Risk Index for heavy metals was calculated thus;

$$\text{Health Risk Index (HRI)} = \frac{\text{Estimated daily intake or daily intake of metal (DIM)}}{\text{Acceptable daily intake or reference oral dose}}$$

$$\text{Daily intake of metals (DIM)} = \frac{\text{M (mg/kg)} \times \text{CF} \times \text{Daily intake of fish (g/day)}}{\text{Average body weight (kg)}}$$

Where CF is Conversion factor = 0.085, M is the metal concentration in the fish tissue, 60 kg was adopted as the average body weight. The fish consumption rate in Nigeria is 48g/person/day.

The health risk index of total hydrocarbons was calculated thus;

$$\text{HRI} = \frac{\text{C} \times \text{IR} \times \text{EF} \times \text{ED}}{\text{BW} \times \text{AT}} \times \text{SF} \times \text{ADAF}$$

Where C = concentration of carcinogen in fish tissues (mg/kg)

IR = intake rate of fish; which is 48g/person/day.

BW = average body weight of exposed individuals (60 kg)

EF= exposure frequency; how many times the individuals are exposed to these carcinogens in a year. The adopted value is 365 days/ year.

ED = Exposure Duration; which is the adopted value of the average life expectancy of a Nigerian; which is 52.62 years in 2014 (CIA, 2016).

AT = length of time over which the average dose was calculated; which is 365 days X 52.62 years

SF = Slope factor; which is 2.0 (mg/kg-day)⁻¹

ADAF = Age-dependent adjustment factor. The adopted value is 1.

Concentrations of chemicals above permissible limits in the aquatic environment elicit high levels of ecological risks. These risks have to be numerically evaluated for quantification and interpretation.

Ecological risk assessment was therefore calculated thus;

$$\text{Risk Quotient (RQ)} = \frac{\text{Environmental concentration (mg/kg)}}{\text{Recommended limit (mg/kg)}}$$

III. Results And Discussion

Water quality characteristics is of high importance in the study of dynamics of heavy metals and total hydrocarbon in the aquatic ecosystem; which can be assessed through monthly sampling (Imoobe, 1997). The concentrations of the parameters analysed in the water samples were all lower than the regulatory limits (Table 1). The concentrations of iron, zinc and chromium in water of Stations 2, 3, and 4 were very much significantly than that of Station 1 (P < 0.001). This can be attributed to the various anthropogenic activities at the different stations, except Station 1; which is the control station. Significant differences across the stations in all other parameters analysed; in comparison with the control station (Station 1). These indicate a significant alteration in the background levels by the anthropogenic activities. Although the levels of the parameters were within

acceptable limits; suggesting that the water of the river supports aquatic flora and fauna. The overall result quite agrees with the earlier observations of the river by Omoigberale and Ogbeibu (2010); within the period of July, 2000 and June, 2002, and Oguzie and Ehigiator (2011) within the period of July to September, 2007. The gills was the most preferred site of accumulation due to the fact that the gills are endowed with physiological and anatomical properties that maximize absorption efficiency (Table 2). This quite agrees with the findings of Eneji *et al.*, (2011).

The sequence of heavy metals and total hydrocarbon in descending order in *Clarias gariepinus* of the river (Fig. 2) was Fe (6.38 %) > Zn (18.97%) > Mn (7.89%) > Cu (4.15%) > Cr (2.55%) > THC (0.81%) > Pb (0.2%) > Cd (0.05%) and that of the water (Fig. 3) was THC (24.7%) > Fe (23.6%) > Zn (12.5%) > Mn (11.7%) > Cr (8.5%) > Pb (8.3%) > Cu (6.2%) > Cd (4.5%). This shows that the tissues of the fish had affinity for copper; which overtook the position of chromium in the sequence observed in the fish. The tissues of the *Claris gariepinus* of the river had the highest affinity for iron; this can be attributed to the importance of the metal in the haemoglobin; required in respiration. The trend of the parameters observed in the fish can generally be attributed to the threshold of essentiality of these metals. However, this observation is at variance with the findings of Eneji (2010) in the *Clarias gariepinus* of River Benue; which was Cr > Zn > Fe > Cu > Mn > Cd > Pb. Iron, zinc, copper, cadmium and total hydrocarbon had significant health risk indices in the fish (Fig. 4); despite the fact that only manganese posed significant ecological risk (Fig. 5). This can be attributed to the high bioaccumulation factors of these chemicals in the tissue of the fish (Fig. 6). This implies that the bioaccumulation factors; also referred to as the octanol/ water partition coefficients (Log Kow) may cause chemicals of low concentrations in the aqueous phase to rise to dangerous levels in biota (Camusso *et al.*, 1995).

Distribution of heavy metals and total hydrocarbon in the tissues of *Clarias gariepinus* was in the order of: Gills (55%) > Muscle (27%) > Intestine (18%). The contaminants had the highest affinity for the gills of *Clarias gariepinus*; followed by the muscles, then the intestine (Fig.7). This is an anomalous order; as against the normal order: gills > intestine > muscle (Eneji *et al.*, 2011; Akan *et al.*, 2012). Under normal circumstances, the gill is expected to accumulate the highest concentrations amongst the tissues. This is due to the fact that the gills is endowed with physiological and anatomical properties that maximize absorption efficiency. The gill also plays outstanding physiological roles of iono-regulation, osmo-regulation and respiration; hence interacts more with chemicals in the water phase than other tissues. Asides all these, the gills is the first target of entry of these contaminants in fish. The intestine on the other hand, is expected to accumulate higher levels of these contaminants than muscle because the intestine is more metabolically active than the muscle. Contrary to these expectations, the muscles tissue of *Clarias gariepinus* of Osse River overtook the position of the intestine (second position) in the accumulation order (Fig. 7). The muscle tissue is the major edible part of fish and constitutes the major part of its body weight (Fabbri *et al.*, 1998); hence holds its nutritional and economic values. The implication of this is that the level of accumulation of heavy metals and total hydrocarbon in the muscle tissue of *Clarias gariepinus* of the river having overtaken that of the intestine, threatens to be of worse health implications to the consumers than expected (Reinfelder *et al.*, 1998). This unusual order, hereby termed critical or anomalous order of tissue contamination was however observed in silver catfish (*Chrysichthys nigrodigitatus*) and *Tilapia nilotica* of Okumeshi River, Delta State, Nigeria (Ekeanyanwu *et al.*, 2010); and also in *Chrysichthys nigrodigitatus* and *Tilapia zilli* of Badagry creek, Lagos, Nigeria by Aderinola *et al.*, (2012).

TABLE 1. Summary of physico-chemical parameters of the water of Osse River between April, 2013 and September, 2014

PARAMETERS	STATION 1	STATION 2	STATION 3	STATION 4	P- VALUE	FME _{Env} Limit
	MEAN±S.E(RANGE) N= 18	MEAN±S.E(RANGE) N= 18	MEAN±S.E(RANGE) N= 18	MEAN±S.E(RANGE) N= 18		
Fe (mg/L)	0.45±0.16 ^B (0 – 2.4)	1.71±0.25 ^A (0 – 3.5)	1.44±0.19 ^A (0 – 2.9)	1.38±0.27 ^A (0.2 – 5.4)	P<0.001	20
Mn (mg/L)	0.02±0.01 ^B (0 – 0.1)	0.24±0.06 ^B (0 – 0.7)	0.97±0.22 ^A (0 – 2.3)	1.26±0.34 ^A (0 – 3.7)	P<0.001	0.5
Zn (mg/L)	0.14±0.03 ^B (0 – 0.4)	0.59±0.15 ^A (0.1 – 2.6)	0.92±0.3 ^A (0.2 – 5.2)	0.985±0.25 ^A (0 – 3.3)	P<0.05	1
Cu (mg/L)	0.02±0.01 ^B (0 – 0.1)	0.14±0.03 ^B (0 – 0.5)	0.13±0.02 ^B (0 – 0.4)	1.05±0.26 ^A (0 – 2.76)	P<0.001	<1
Pb (mg/L)	0.01±0.003 ^B (0 – 0.1)	0.08±0.01 ^B (0 – 0.2)	0.83±0.24 ^A (0 – 2.7)	0.83±0.26 ^A (0 – 2.7)	P<0.001	<1
Cd (mg/L)	0.0±0.0 ^B (0 – 0)	0.08±0.02 ^B (0 – 0.3)	0.23±0.12 ^B (0 – 1.6)	0.4±0.16 ^A (0 – 2.3)	P<0.05	<1
Cr (mg/L)	0.02±0.01 ^B (0 – 0.2)	0.35±0.1 ^A (0 – 1.1)	0.71±0.18 ^A (0 – 2.1)	0.73±0.2 ^A (0 – 2.3)	P<0.01	<1
THC (mg/L)	0.02±0.01 ^B (0 – 0.1)	3.19±0.6 ^A (0 – 10.5)	0.77±0.2 ^B (0- 1.89)	1.26±0.28 ^C (0 – 3.2)	P<0.001	10

Note: Values with similar superscript letters have no significant difference
 N= number of sample replicates. P>0.05 means there is no significant difference, P<0.05 means there is significant difference, P<0.01 means there is much significant difference, and P<0.001 means there is very much significant difference.

TABLE 2. Summary table of heavy metals and the total hydrocarbons in *Clarias gariepinus*

PARAMETERS	GILLS	INTESTINE	MUSCLE	P- VALUE
	MEAN±S.E (RANGE),	MEAN±S.E (RANGE),	MEAN±S.E (RANGE),	

	N= 18	N= 18	N= 18	
Fe (mg/L)	281.8±42.3 ^A (21-658)	66.1±7.8 ^B (15.5-153)	116.8±18.7 ^B (15.8-242.4)	P<0.001
Mn (mg/L)	24.1±5.1 ^A (1.7-62.3)	10.5±2.7 ^B (1.1-37.4)	21.5±2.5 ^A (1.3-35.1)	P<0.05
Zn (mg/L)	57.8±6.8 (10-117)	38.9±7.5 (7-118)	38.1±5.5 (7-83)	P>0.05
Cu (mg/L)	14.9±2.6 ^A (2.2-45.6)	8±1.4 ^B (0.6-20.5)	6.6±1.2 ^B (1-14)	P<0.01
Pb (mg/L)	1.1±0.2 ^A (0-3)	0.2±0.03 ^B (0-0.4)	0.1±0.03 ^B (0-0.4)	P<0.001
Cd (mg/L)	0.1±0.02 (0-0.1)	0.1±0.02 (0-0.6)	0.2±0.09 (0-1.6)	P>0.05
Cr (mg/L)	6.4±1.3 (0.01-18.5)	4.4±0.8 (0-11.4)	7.331±1.756 (0-23.03)	P>0.05
THC (mg/L)	3.5±0.5 ^A (0.6-8.2)	1.6±0.2 ^B (0.3-3.5)	0.6±0.2 ^C (0.2-2.3)	P<0.001

Note: Values of same superscript letters have no significant difference

N= number of sample replicates, P>0.05 means there is no significant difference, P<0.05 means there is significant difference, P<0.01 means there is much significant difference, P<0.001 means there is very much significant difference

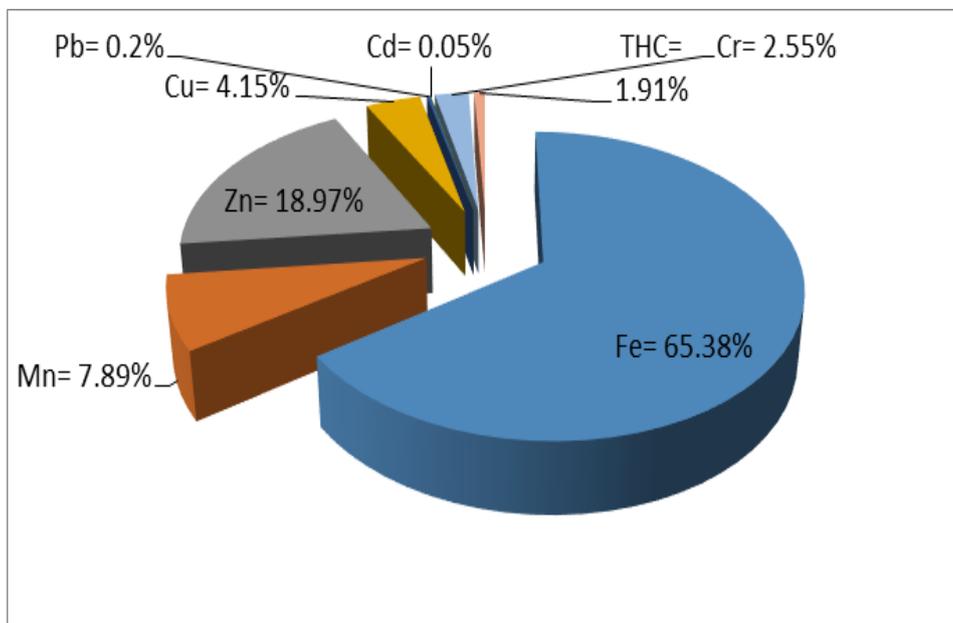


Figure 2: Percentage composition of heavy metals and total hydrocarbon in fish

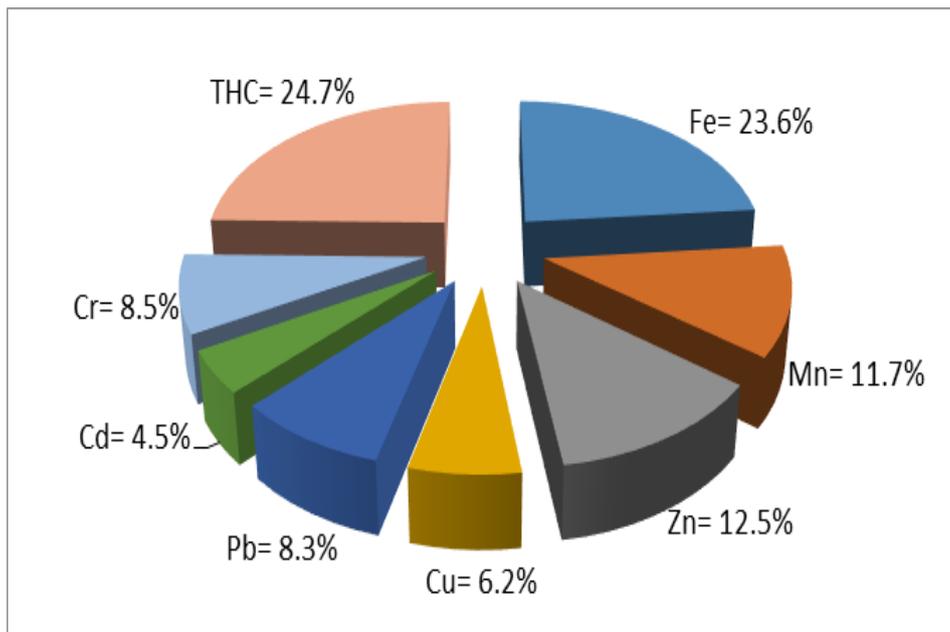


Figure 3: Percentage composition of heavy metals and total hydrocarbon in water

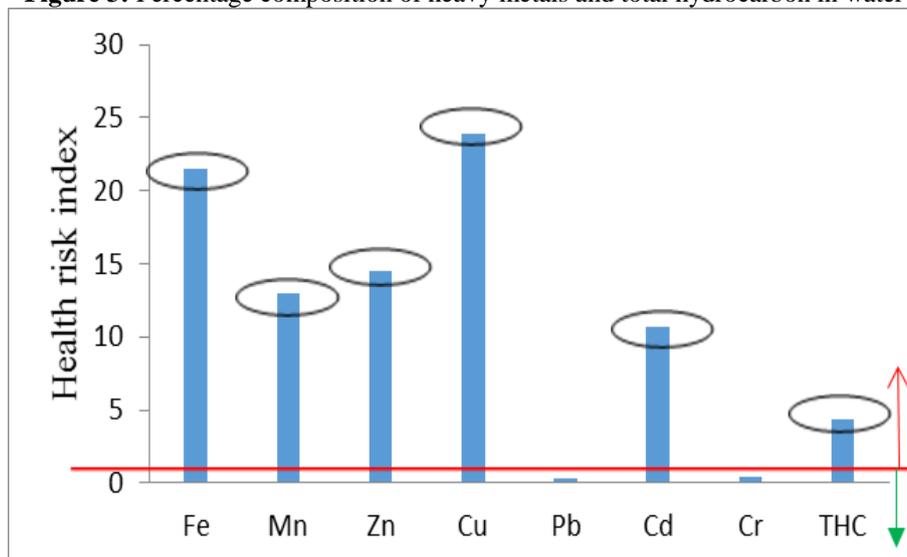


Figure 4: Health risk indices in *Clarias gariepinus*

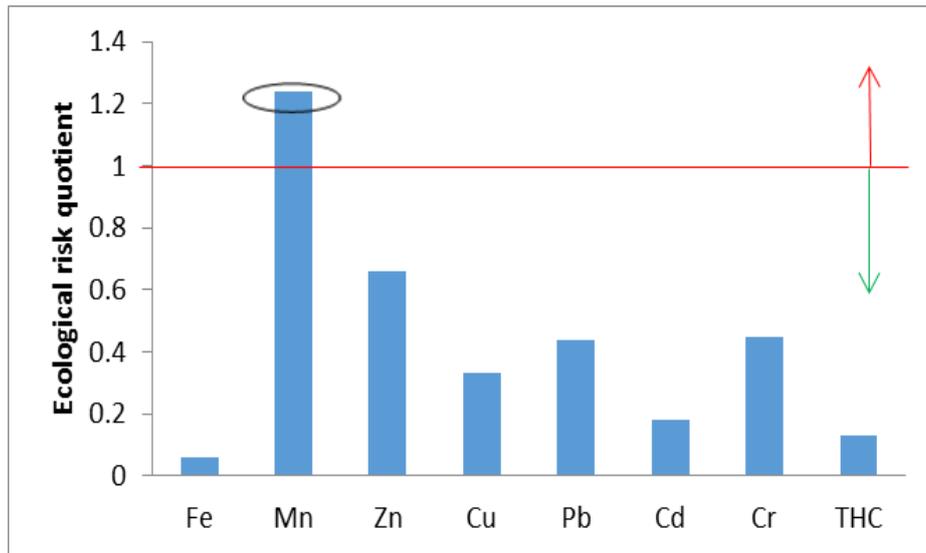


Figure 5: Ecological risk quotient

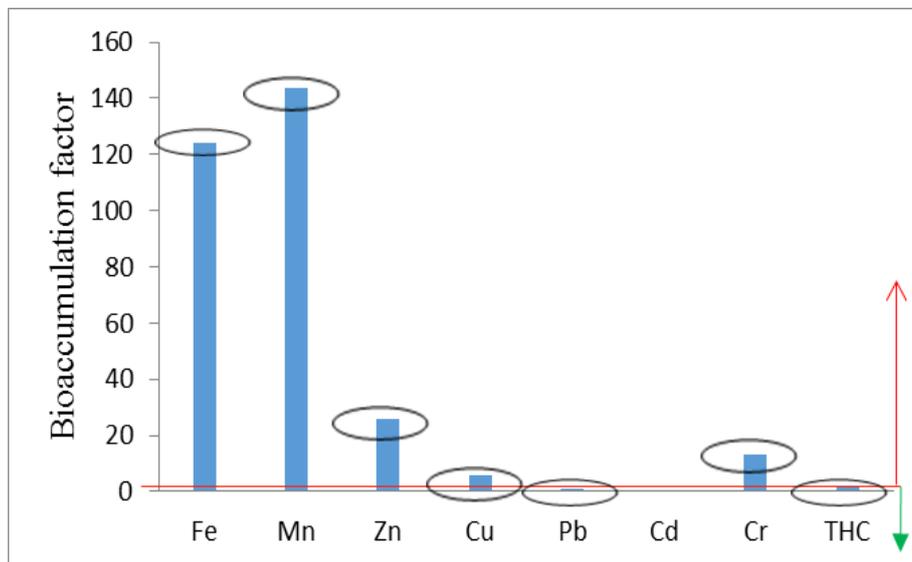


Figure 6: Bioaccumulation factor (BAF) in *Clarias gariepinus*

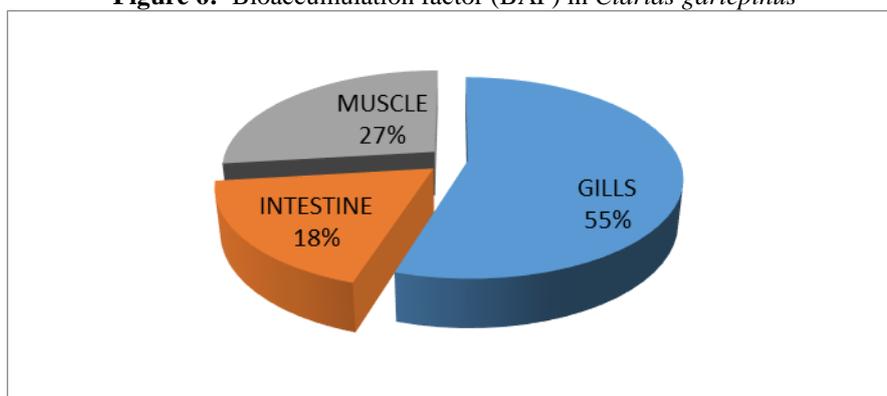


Figure 7: Percentage distribution of heavy metals and total hydrocarbon in tissues of fish

IV. Conclusion

The water of Osse River is of no ecological risk to the aquatic life and it is suitable for domestic use. However the *Clarias gariepinus* of the river may pose some levels of chronic threats to the consumers. Worse still is the outstanding contamination of the muscle which is the edible part. We hereby recommend further in-depth study in order to ascertain actual risks consumption of the fishes may pose to the health of the consumers;

as well as factors responsible for anomalous order of tissue contamination. Regulation of anthropogenic activities, immediate standard remediation processes and constant bio-monitoring studies of the aquatic environment are highly recommended. Further deeper studies to ascertain the actual levels of risks to consumers is imperative.

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