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Assessment of Heavy Metals Accumulation in Tissues of *Tilapia zilli* and *Clarias gariepinus* Found in Lake Akpoko and River Benue, Nigeria

Baturh Yarkwan* and Daniel Apeh
Biochemistry Unit, Department of Biological Sciences, University of Agriculture, P.M.B. 2373, Makurdi, Benue State, Nigeria.
* E-mail of the corresponding author: baturybatur@yahoo.com

Abstract
Water run-offs from up lands polluted by human activities constitute a potential source of heavy metal contamination of the aquatic flora in surrounding water bodies. For instance, lake Akpokpo has abattoir effluents emptying into it all through the year. To investigate this, two commonly consumed fish species, namely *Tilapia zilli* (Tilapia fish) and *Clarias gariepinus* (cat fish) were assessed for heavy metals contamination in the gills, intestines and muscles of freshly caught samples following standard procedures. Heavy metals were determined using atomic absorption spectrophotometer. The results showed that the gills of *Tilapia zilli* from river Benue has the highest concentration of all the metals, while the muscles of cat fish from the lake has the lowest metals concentration. Heavy metals accumulation in the samples ranges in the order TGR>TGL>TIL>CGL>CGR>TIR>CIL>CIR>CMR>TML>CML. In the organs, the range of accumulation is Gills > intestine > muscles. The concentration of heavy metals in the river is in the order Fe> Zn >Cr>Pb>Mn>Cd>Cu. Lead was 12.15 mg/kg in the gills of *Tilapia zilli*, but 2.08 mg/kg in the muscles of cat fish, both from the lake. The river samples showed lead concentration of 11.585 mg/kg and 2.24 mg/kg, for the Tilapia gills and cat fish muscles respectively. Fishes constitute a staple food for river bank communities. Considering daily permissive intake limits, discussions and recommendations were made.
Key words: Heavy metals pollution, *Tilapia zilli*, *Clarias gariepinus,* water pollution

1.0 Introduction

Aquatic environment is considered the main factor controlling the state of health and disease in both cultured and wild fishes (Samir *et al.*, 2008). Pollution of the aquatic environment by inorganic chemicals is a major factor posing serious threat to the survival of aquatic organisms including fish (Samir *et al.*, 2008). However, fish is the major source of protein, (Ali *et al.*, 2011, Gomna & Rana, 2007), especially among local peasants whose meals are majorly constituted of carbohydrates staples.

Bioaccumulation of heavy metals in living organisms and biomagnifications describe the processes and pathways of pollutants from one trophic level to another (Akan *et al.*, 2012). The bioaccumulation of heavy metals in aquatic organisms depends on the ability of the organisms to digest the metals, the concentration of the heavy metals in the surrounding soil sediments and the feeding habit of the organism (Eneji *et al.*, 2011). Heavy metals can be incorporated into food chains and absorbed by aquatic organisms to a level that it affects their physiological state (Akan *et al.*, 2012). The results of many field studies of metal accumulation in fish living in polluted waters show that considerable amounts of various metals may be deposited in fish tissues without causing mortality (Jezierska *et al.*, 2006). Fish accumulate metals in their tissues through absorption, and humans can be exposed to these metals via the food chain (Bashir *et al.*, 2013). This accumulation is capable of eliciting toxic effects in humans, whose extent and severity cannot be overemphasized.

Lake Akpokpo lies on the bank of river Benue. It is a source of water for irrigation, cleaning of animal hides at the abattoir stationed just on its bank and fishing activities also takes place in it. Its fishes are highly patronized by the neighbouring community harbouring thousands of people. The effluents from the abattoir flow directly into the lake. Being that it is overrun by the river water in rainy seasons, the effluents might settle in the shallow lake, but flow into the main river course and are carried along, while sediments might deposit in the lake. Moreover, rather than fetch water out into the abattoir, the workers prefer washing the internal organs directly inside the lake. This again constitutes a source of contamination. Such contaminants are likely to accumulate in the various organs of fish and may affect humans and other species that depends on such fish as food.

There are several sources of heavy metal pollution in the aquatic environment. Basically, these are natural and anthropogenic, of which the anthropogenic are most common (Eneji, *et al.*, 2011). These arise from waste incinerations, industrial wastes, sewage disposal (Abida *et al.*, 2009; Samir *et al.*, 2008). Some of these pollutants come from polluted runoff in urban and agricultural areas and some are the result of hysterical contamination (Abida *et al.*, 2009). River Benue receives run-offs from the up-land areas. The farmers along the river bank, use fertilizers and synthetic agro-chemicals (Akan *et al.*, 2012).

The rate of bioaccumulation of heavy metals in aquatic flora depends on the ability of the organisms to digest the metal and the concentration of such metals in the river (Eneji *et al.,* 2011). Some kinds of toxic sediments kill benthic organisms, reducing the food available to larger aquatic animals, in keeping with the aquatic food web. Some contaminants in the sediments tend to bio-accumulate in benthic organisms. When larger animals feed on these contaminated organisms, the toxic substances are taken into their bodies, and thus become biomagnified (Abida *et al.*, 2009). Accumulation of heavy metals in fish depends on metals concentration, time of exposure, pH, hardness, salinity of the water, water temperature and intrinsic factors such as fish age, feeding habits, among others (Jezierska *et al.*, 2006). Before now, most of the heavy metals were taught of affecting the enzymes, thus interfering with cellular biochemical integrity. Recent developments have however, implicated some heavy metals as endocrine disruptive chemicals, whose synergistic functioning in the body results into metalbolic syndrome (Lavicoli, *et al.*, 2009)

This work was carried out to ascertain heavy metal pollution of fishes in lake Akpoko and River Benue, in two most commonly consumed species of fish, namely *Tilapia zilli* and *Claria gariepinus.*

2.0 Materials and Methods

2.1 Samples Collection

Cat fish (*Clarias gariepinus)* and Tilapia fish (*Tilapia zilli)*, were bought directly from fishermen at river Benue under the bridge at Wurukum and from Akpokpo lake at the Wurukum abattoir, in Makurdi, Benue state, Nigeria. Two supposed Cat fish and two Tilapia fish were obtained from source and transported under near frozen cooler packs to the laboratory and were identified.

2.2 Preparation of Samples

The scales of the Tilapia fish were carefully removed using a sharp knife to avoid injury on the tissue. The fishes were washed with running tap water in the laboratory. Dissection was done using a sharp stainless steel knife and the organs (intestines, gills and muscles) were removed apart each other. The organs were oven dried (GallenKamp Oven, UK) to constant weight at 105^{o}C. The tissues were removed and cooled in a dessicator and later weighed using a digital chemical balance (Adams Equipment Co. Ltd. UK). The samples were grounded using a porcelain mortar and pestle into powdered form and kept in a plastic container until digestion.

2.3 Acid Digestion of Samples

The method of Eneji *et al*., (2011) was adopted. Briefly, 10g portion of the grounded samples were carefully weighed out using digital chemical balance, 10ml of HNO_3 and 2ml of $HClO_3$ were added and heated over a hot plate for one hour. After complete digestion, the residue was diluted with 0.2% V/V HNO_3 to 20ml.

Metal concentrations were read using Atomic Absorption Spectrophotometer, VGP 215, at the respective wavelengths, following manufacturers' specifications. The standard curves were plotted from determinations made using standard metallic samples.

3.0 Results and Discussion

The result obtained indicates that the gills of *Tilapia zilli* from river Benue contains the highest concentration of the metals, while the muscles of cat fish (*Clarias gariepinus*) from the lake has the lowest concentration of heavy metals. The accumulation of heavy metals in these organs is in the order: TGR>TGL>TIL>CGL>CGR>TIR>CIL>CIR>CMR>TML>CML. The range of metal concentration in the organs of the fishes is in this order; Gill > intestine > muscles and agrees with the report of Eneji *et al.* (2011) and Bahnasawy *et al*, (2009). Khail *et al,* (2008) reported that higher metal concentrations in the gills could be due to the element complexes with the mucus that is virtually impossible to completely remove from the gill lamellae before preparation for analysis. Since the gills of *Tilapia zilli* are used for gaseous exchange and as a "sieve", heavy metals that are floating with different particles on the surface of the water easily get attached to the gills since *Tilapia zilli* often stays at the surface of shallow waters. Target organs such as gills and intestine are metabolically active parts that can accumulate heavy metals in higher concentrations as shown in various fish species, (Khail, *et al.,*2008).

From the tissues analysed, it was observed that the concentration of metals in the fishes from the two sources is in the order of Fe>Zn>Cr>Pb>Mn>Cd>Cu. This order of metal concentration is similar to Fe>Zn>Pb>Cu>Cd>Hg order reported by Barbara *et al.* (2006) and the order Fe>Cr>Pb>Mn>Zn>Cu>Cd reported by Eneji *et al.* (2011). These differences in the metal concentrations arise possibly due to their concentrations in the water. Besides, it could also be because of the different complexes' pattern in which the metals exist and their resultant differential affinity for the fish tissues; differential uptake, deposition and excretion rates as well as bioavailability, intrinsic fish processes and trophic structure variations.

From the result, of all the tissues examined, the concentration of heavy metals in the gills is highest (fig.8). From interactions with the women/fishermen, usually gills are removed from fresh fishes before preparing them for meals. However, when they are being dried for preservation for subsequent consumption and marketing, in order to preserve a large biomass and keep the anatomical outlook of the fish intact (which is a cherished property in marketing fish), the gills are not removed. This is where the greatest threat is posed. These elements may diffuse to the underlying muscular tissues and remain deposited there and be consumed later. Even though the gills might be removed in the dry fish while preparing (that is if the cook is careful enough to do this, but also considering the size of tilapia fish, it is not effective doing this), thus much of these will rather be consumed in the dry fish than in the fresh fish. However, since preference is for fresh fish among fishing communities (Gomna, 2011), it is likely that much of the consumed fish is devoid of the gills, the major contaminated fish organ. Yet some of the processed fish will obviously constitute a threat to the consumers.

CG – Cat fish Gills, CM – Cat fish Muscles, CI – Cat fish Intestines, TG – Tilapia fish Gills, TI – Tilapia fish Intestines, TM – Tilapia fish Muscles. Where R and L are added to any of the above mentioned abbreviations, it represents fish sample from River Benue and Akpoko Lake respectively.

Zn had the highest concentration of 22.39 mg/kg in TGR and 15.525 mg/kg in TML. The lowest concentration of 2.45 mg/kg was observed in CML and 2.73 mg/kg in CMR (fig.1). These are higher than the 0.87 mg/kg, reported by Sambo *et al*. (2014). Eneji *et al*. (2011) reported 7.05 mg/kg of Zn in tilapia gills. However the values are less than the 71.80 mg/kg reported by Obodo (2004). Zn is an essential component of metallo-enzymes like alkaline phosphatase and lactate dehydrogenase. WHO (2011) has recommended 3-5 mg/L of Zn in drinking water. The Zn values for the gills and intestines of the fishes are however above the WHO reference value, while the 2.73 mg/kg, 3.045 mg/kg, 2.45 mg/kg, and 2.555 mg/kg, for CMR, TMR, CML and TML respectively are less than or within this range, thus posing no threat of any hazardous effects.

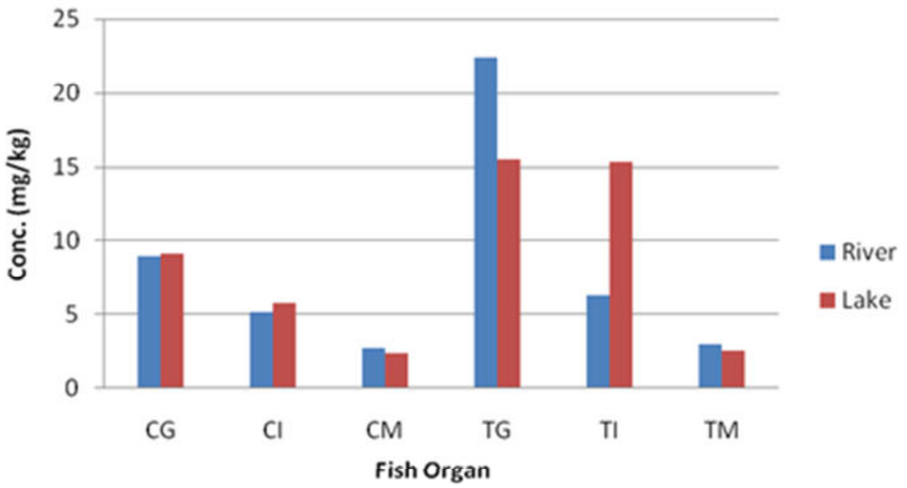

Fig. 1:Conc. of Zn in fish organs from River Benue and Akpokpo lake

The concentration of cadmium (Cd) in the gills, intestines and muscles of the two fish species is shown in Fig. 2. The gills of Tilapia had the highest concentrations of 8.63 mg/kg and 8.50 mg/kg for the river and lake samples respectively. The muscles of tilapia had the least concentration of 1.26 mg/kg and 1.23 mg/kg for the river and lake samples respectively. These values are higher than the 0.994 mg/kg and 0.927 mg/kg reported by Eneji, *et al.* (2011) for tilapia and cat fish respectively. Considering the synergistic effect of Cd, its effect on the skeletal system, and its potency in inducing kidney dysfunction, these values are high enough to attract public health concerns. These are higher than the 0.003 mg/L WHO (2011) maximum limits in drinking water. Cadmium is released when there is weathering of rocks. It is thought that the preceding flooding in the river would have contributed, with the sediments depositing along the river on the river bed close to the river bank where shallow water fishes such as tilapia dwell.

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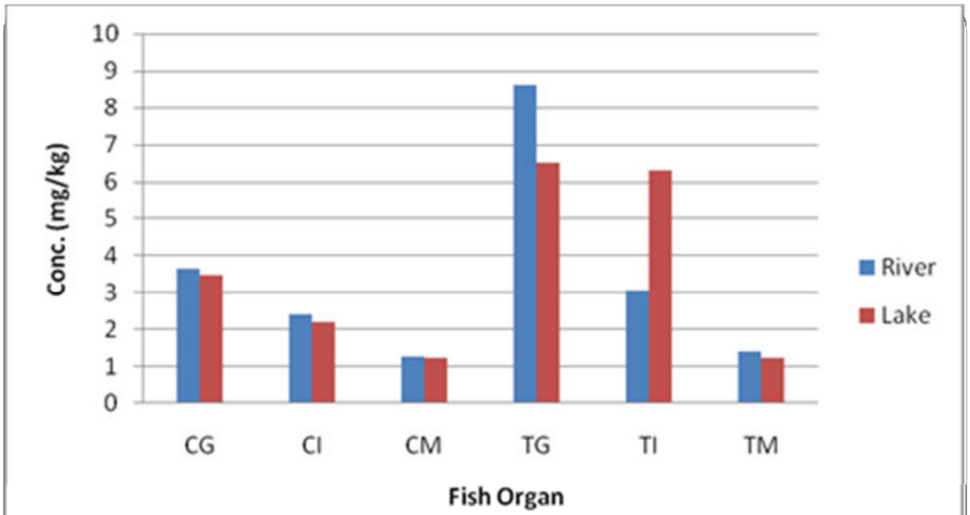

Fig.2: Conc. of Cd in fish organs in River Benue and Akpokpo lake

The concentration of Lead (Pb) in the gills, intestines and muscles of the two fish species is hown below in fig. 3. Highest Pb concentration of 12.15 mg/kg was observed in TGL against 11.585 mg/kg in TGR. The lowest concentrations were 2.04 mg/kg in TMR and 1.92 mg/kg in TML (fig.3). This is by far higher than the 1.40 mg/kg reported by Eneji *et al*. (2011) and the 0.27 mg/kg reported by Sambo, *et al*. (2014). These values were however less than the 61.32 mg/kg reported by Obodo (2004). This could be due to the recent over flooding of the river banks, which increased the quantity of sediment deposits in the water bodies. These results are higher than the 0.30 mg/kg set by the European Union (EU). Lead may slow cognitive development, impair intellectual performance in children and is implicated for causing increase in blood pressure and cardiovascular diseases. Moreover, its synergistic action with Hg and Cd have been reported in disruptive effect on the endocrine glands thereby causing metalbolic syndrome (Georgescu et al., 2011).

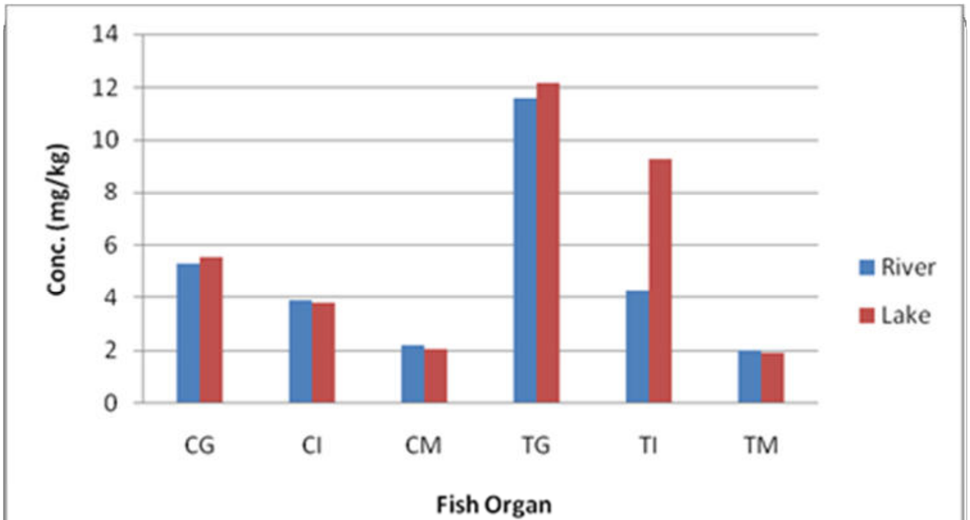

Fig.3:Conc. of Pb in Fish organs from River Benue and Akpokpo Lake

The concentration of Iron (Fe) in the gills, intestines and muscles of the two fish species is as shown in fig.4. The gills of Tilapia had the highest concentrations of 23.63 mg/kg and 17.45 mg/kg for the river and lake samples respectively. The muscles of cat fish had the least concentration of 3.145 mg/kg and 2.77 mg/kg for the river and slake samples respectively. Iron concentration in the TGR was 23.630 mg/kg against 17.425 mg/kg from TGL. The lowest concentration of 2.77 mg/kg was observed in the CML against 3.145 mg/kg in CMR (fig. 4). This may be due to higher exposure of river Benue to more pollutants (Eneji *et al.* 2011). Fe had the highest concentration from the analysis. About 70% of iron in mammals is found in haemoglobin and about 5% is found in myoglobin (Albertsen, 2006). However, free iron promotes the formation of highly reactive oxygen species such as hydroxyl radicals that can damage DNA and other macromolecules (Nelson *et al.,* 2005). Since the value is less than the WHO reference intake value per day, it may cause no physiological threat.

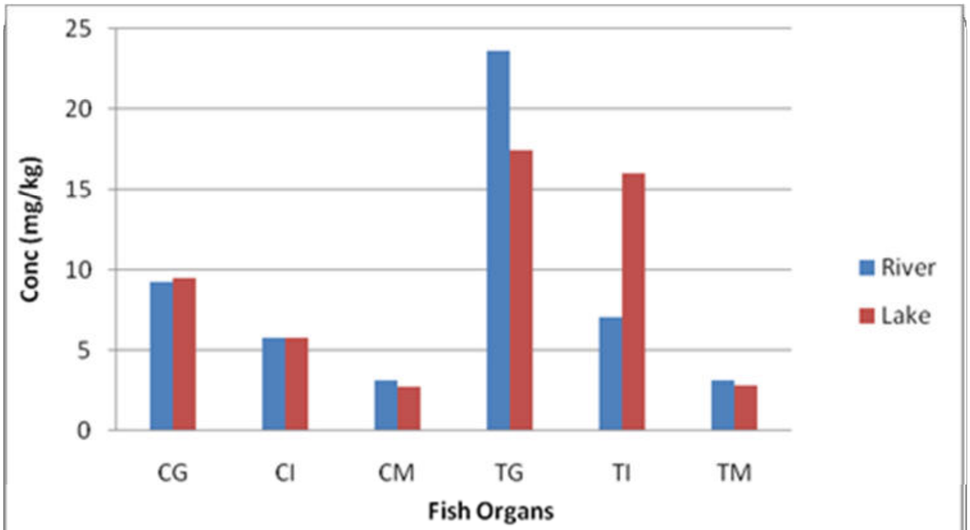

Fig.4:Conc. of Fe in Fish Organs from River Benue and Akpokpo Lake

Copper (Cu) concentration in the gills, intestines and muscles of the two fish species is shown in fig.5. Copper had the highest concentration of 5.525 mg/kg in TGL and 6.185 mg/kg in TGR. The lowest concentrations were 0.665 mg/kg and 0.835 mg/kg for TML and CMR respectively. These values are less than the 9.99 mg/k reported by Eneji *et al.* (2011*)* for tilapia fish. All the values are higher than the 0.30 mg/kg reported by Sambo *et al*. (2014). Although copper toxicity is rare, at high doses it is associated with heart diseases.

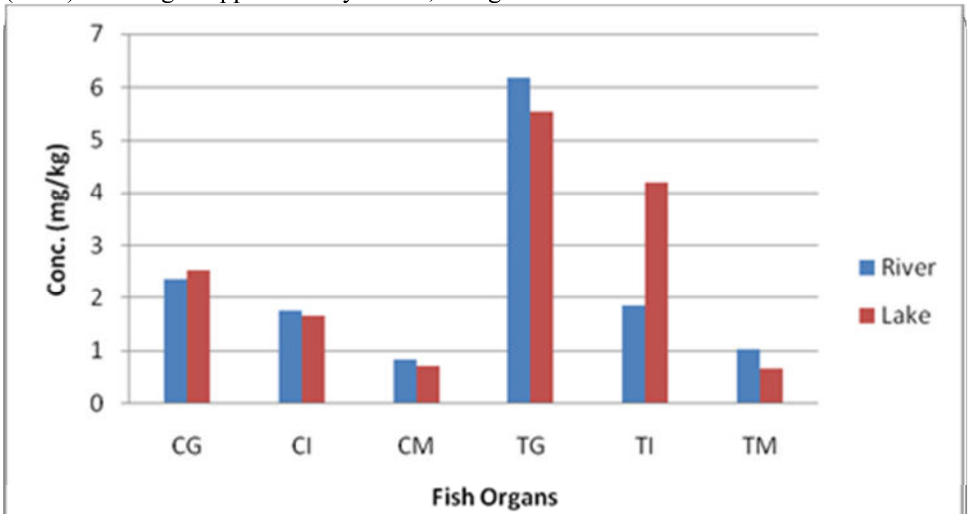

Fig.5: Conc. of Cu in fish organs in River Benue and Lake Akpokpo

The concentration of Chromium (Cr) in the gills, intestines and muscles of the two fish species is shown in fig. 6. Chromium was observed to have the highest concentration of 17.095 mg/kg in TGR and 13.800 mg/kg in TGL. The lowest concentrations were 2.345 mg/kg and 2.055 mg/kg for CMR and TML respectively. These values are all far less than the 92.9 mg/kg reported by Eneji *et al.* (2011). The muscle Cr concentration is similar to the 2.77 mg/kg reported by Sambo *et al. (2014).*WHO (2011) limits for total Cr in drinking water is 0.05 mg/L.

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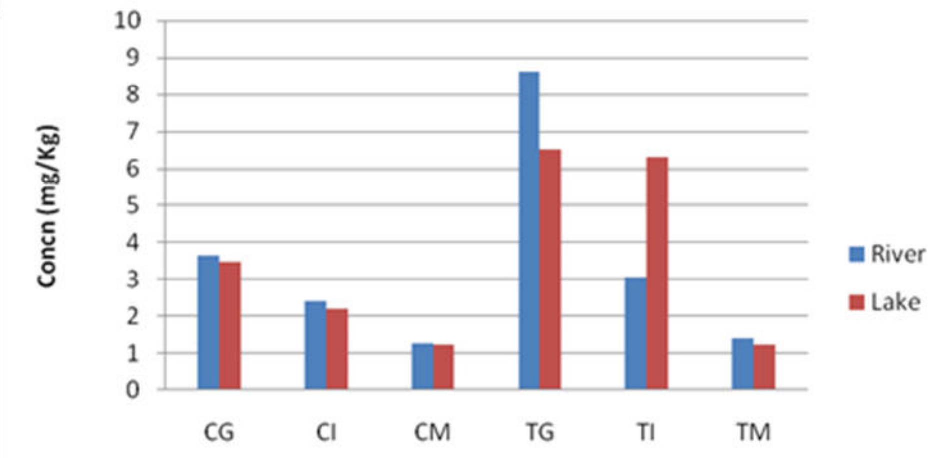

Fig.6: Conc. of Cr in fish organs in River Benue and Lake Akpokpo

The concentration of Manganese (Mn) in the gills, intestines and muscles of the two fish species is shown in fig. 7. Manganese had the highest concentration of 10.400 mg/kg in TGL and 10.930 mg/kg in TGR. The lowest concentrations were observed in TML and CMR, with 1.215 mg/kg and 1.550 mg/kg respectively. The tilapia gills concentration is more than the 7.82 mg/kg reported by Eneji, *et al*. (2011) for tilapia. They also reported 3.51 mg/kg for cat fish, which is in agreement with the 3.52 mg/kg of the present study. These values are far less than the 56.14 mg/kg reported for tilapia by Obodo (2004).

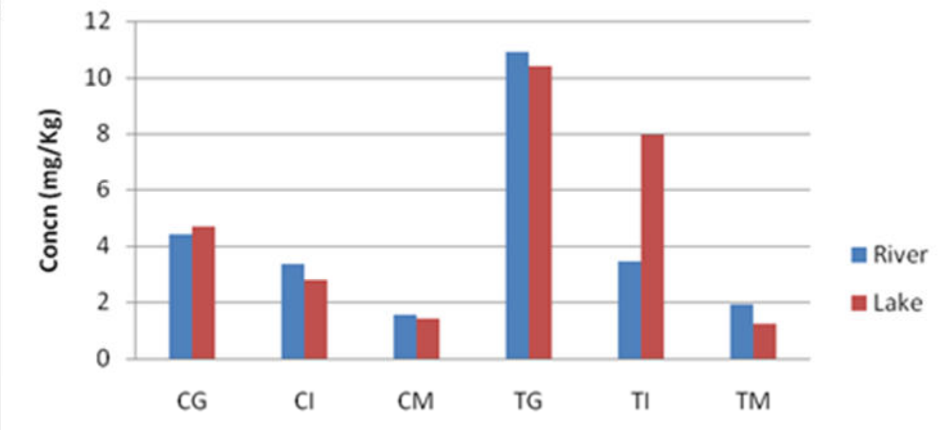

Fig.7: Conc. of Mn in fish organs in River Benue and Lake Akpokpo

Fig.8 below shows the average concentration of heavy metals in the respective fish tissues of fishes from river and lake. The gills of tilapia have the highest average heavy metal concentration of 11.62 mg/kg and 14.65 mg/kg for lake Akpoko and river Benue respectively**.** The least concentrations of 2.79 mg/kg and 2.02 mg/kg were observed in the muscles of tilapia fish from the lake and cat fish from the river respectively.

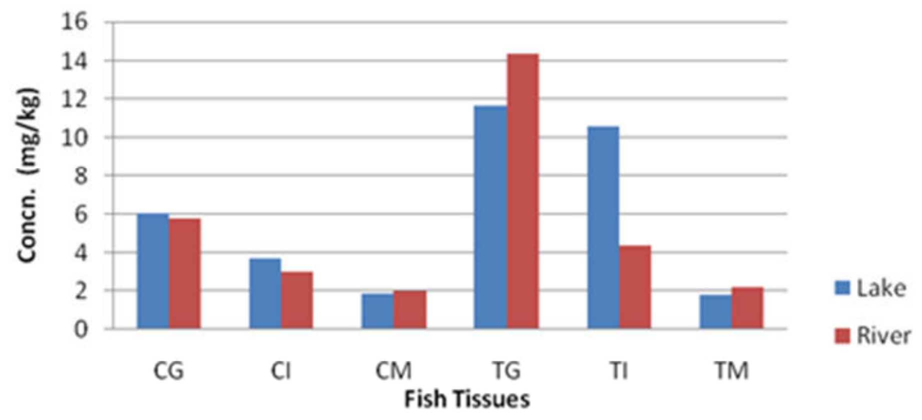

Fig. 8: Average heavy metals concentration in tissues of fishes from different water sources

In all the cases, it is observed that the heavy metals accumulate in different tissues at varying concentrations. Also, considering the different species, there were differences in accumulation of the heavy metals. Generally, all the gills had the highest concentrations of heavy metals. This is similar to the report of Bahnasawy, (2009). Higher metals concentrations were observed in the muscles of cat fish than those of tilapia fish, while the reverse was the case for the gills (fig. 8). This could be because the Tilapia fish is a shallow water dwelling fish, which survives in close proximity to the entry point of water inflows. This water from polluted uplands is taught to contain higher quantities of heavy metals carried along by the sediments. Thus, in a bid to facilitate gaseous exchange and uptake of food particles, it interacts with the highly contaminated water thereby taking up some of its pollutants on the gills. The intestine is the metalbolic transit hub for all ingested food particles. Thus it measures not just the metals content in the tissues but also of the transit food ingested which is undergoing digestion as at the time of sampling. The concentration in the muscles is thus the most authentic measure of the presence of the metals in the fish tissues. This could be traced to the regulatory ability of the fishes and their respective feeding habits. This view is shared by Eneji *et al.* (2011*).* Moreover, it is thought that since cat fish has a longer life span than tilapia, the former is capable of accumulating larger quantities of these metals within its tissues than tilapia fish.

4.0 Conclusions.

It is evident from the afore going that the two most commonly consumed fishes, namely *Tilapia zilli* and *Clarias gariepinus* inhabiting River Benue and Akpokpo Lake in Makurdi Benue State are heavily contaminated with the heavy metals investigated. The continual consumption of these fishes will lead to bioaccumulation of these heavy metals in the bodies of those who consume same, with the attendant deleterious health effects, arising from the biochemical interactions of these chemicals with the systems. The abattoir effluents did not constitute a source of heavy metals pollution on the fishes caught from Akpoko lake, since those from the river had higher heavy metals concentration.

Fish gills and intestines should always be removed from fishes before cooking. Furthermore, fish intestines and gills should be removed from fishes before preserving them to avoid diffusion of the metals into surrounding tissues. Proper sanitation should be enforced in the town, indiscriminate dumping of refuse, waste material, sewage disposal should be avoided. There is need for aggressive public enlightment campaigns to educate the river bank dwelling fishing communities on the need to carefully prepare their fishes before consumption. Subsequent studies need to be carried out to ascertain the blood level concentration of these heavy metals among the community. Moreover, dried fish should be studied to establish the metal concentration levels on the fish species.

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