



Assessment of Trace Metal Concentrations in internal Organs of *Tilapia zilli* Caught from Orashi River, Niger Delta, Nigeria.

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Abstract: Environmental pollution associated with trace metal concentrations is an emerging issue in most developed and undeveloped countries. Orashi River is open to several inputs from industrial and other human mediated activities within the environment. This research was therefore aim to determine the concentrations of trace metal in the tissues (intestine, muscles, liver, gills) of *Tilapia zilli* for 12 months (between May 2016 and April 2017). From the results it was observed that trace metals bioaccumulated in the tissues of *T. zilli* with high concentrations in the intestine and muscles. Significant variations for upstream and downstream were observed for Cd, Cr, Cu, Pb, Mn and Zn ($p < 0.05$) exception for Iron ($p > 0.05$). The high concentration of Fe and Zn recorded during the study can be attributed to dissolution of zinc from oil pipelines fixed across the water body by oil companies and the use of iron coagulants or the corrosion of steel and cast-iron pipes during the process of water distribution. Transfer factor index used in this study as a tool to assess the level of bioaccumulation in the studied species showed evidence of bioaccumulation of heavy metals in the tissues which calls for public health concern. However, the level of trace metal were above recommended tolerable limits for most of the metals when compared to world standards for safety of consumption of aquatic organism exception for Pb and Cr. Based on findings, it is however, recommended that trace metals concentrations in edible seafood species from aquatic ecosystems should be monitored at regular interval for safety of humans consuming these seafoods.

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1. Introduction

Increased industrialization is causing serious environmental pollution through improper and indiscriminate effluent discharges and emissions arising from indiscriminate disposal of waste generated from these industries. (Inyang-Etor, and George, 2019). In general, heavy metals related studies can be very important and interesting particularly in the fields of ocean sciences and other related discipline. The alarming increase in the rate of recorded cases of diseases associated with impacts of heavy metals and the direct effects of these contaminants on the aquatic ecosystems have mandated concern on the need to monitor the heavy metal concentrations in indigenous lotic and lentic systems; particularly metals which pose an imminent health hazard to humans (George *et. al.* 2013). Metals accumulations in fish vary with species. This may be due to genetic factors, food and feeding habits, rate of deposition and excretion rates. The assessment of toxic heavy metals in fish tissues can serve as bio-indicator and may give an insight to the degree and

extent of contamination of a particular water body (Farkas *et al.*, 2000).

Bioaccumulation of heavy metals in tissues of aquatic organisms has been acknowledged as an indirect measure of the abundance and availability of metals in the aquatic environment (Kucuksegin *et al.*, 2006). It is on this note that, monitoring fish tissue contamination serves an important function for early warning indicator of sediment contamination or related water quality problems (Mansour and Sidky, 2002) which enables one to take appropriate action to protect public health and the environment.

Several authors have indicated much concern over the issue of metals contamination and pollution of both biota and the environment and this is indicated through research efforts which abound in literature. This is partly because of metals general characteristics of gross persistence, acute and chronic toxic effects as well its tendencies to permeate membranes and accumulate in edible tissues of living organisms (Has-Schon, 2006; Mbong, *et. al.* 2013). Also, the need to frequently monitor the levels of xenobiotics in our

environment and edible biota cannot be over emphasized especially in the oil rich Niger delta region of Nigeria where oil exploration is endemic over the last five decades. Furthermore, the high level of heavy metals (Zn, Cu, Cd, Pb and As) beyond WHO limits reported by Allinor, (2005) in Aba river and Farombi *et. al.* (2007) in ogun River proximal to industrial plants enforce the pivots of this research. From this, we aimed at quantifying the levels of some trace metals (Cd, Cu, Fe, Pb, Zn, Mn and Cr) in intestine, muscles, liver and gills of *Tilapia zilli* caught from the upstream and downstream of Orashi River, Nigeria in line with food safety and public health concerns.

2. MATERIALS AND METHODS

2.1 Description of study area

Orashi River (Figure 1) is located between latitude $5^{\circ} 08' 22.7''$ and longitude $6^{\circ} 29' 57.6''$ (up-

stream) and latitude $5^{\circ} 01' 11.6''$ and longitude $6^{\circ} 26' 04.3''$ (down-stream), with an elevation of 270 meters above sea level. The study area is inhabited by two (2) Ethnic groups (The Engenni's and The Ekpeye's) both on either side of the river. Orashi River is a crucial resource for all the surrounding communities. The river system is strongly seasonal, and the main feeder river on the east flank of the lower Niger Delta when viewed through the map. In the dry season, water entering the system comes from the flood plains and drains up-stream through Onosi, Omoku, Ndoni and Oguta Lake while during the wet season, the Orashi River is swollen by the overflow of River Niger flood which enters Orashi River mainly through Ndoni creek. Human activities along the study area are mainly dredging, illegal crude oil refining, transportation, fishing, markets, disposal of domestic waste, sewage disposal, sand dredging, jetties and agricultural run-off.

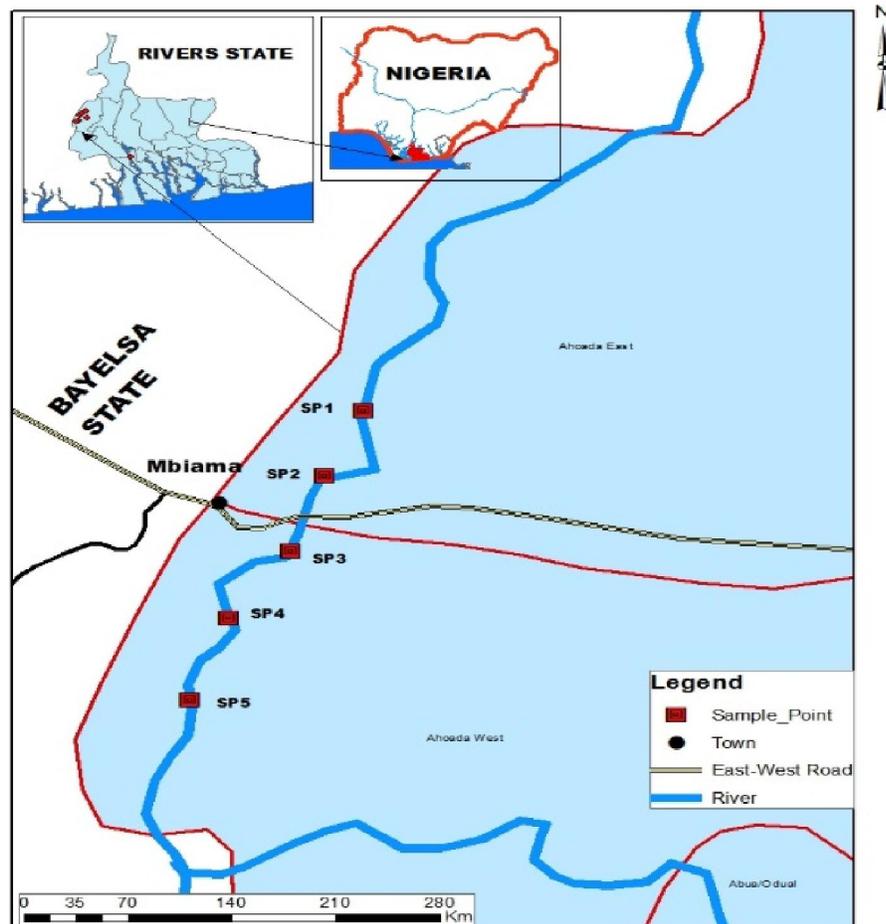


Fig. 1: Map of Study Area Showing Sampling Locations

2.2 Sampling Stations

Five (5) sampling stations from the upstream and downstream of the River were established. These

stations were chosen based on the level of ecological activities that took place along the shore of the river. station one was established in Oshiobele Community

within the upstream of the River. The station was located within latitude 5° 5'10.52"N and 6°28'19.98"E. Human activities in this station are characterized by high level of agricultural activities, disposal of domestic waste and sewage run-off. Other human activities sited include; fishing, loading and offloading of farm products etc. The station received input of agricultural and human waste from adjoining creeks and stream. Station two (2) is located in a community called Akinima which lies in the upstream of the River, on latitude 5° 4'32.11"N and 6°27'55.53"E. Human perturbations in this station include, fishing, dredging of sand, loading and offloading of petroleum products and farm products. This station received inputs from agricultural run-off, industrial discharge, domestic runoff and waste from local dwellers. Station three (3) is located in Mbiama community within the downstream of the River. The distance between this station and station two was about three kilometers. The station is located within latitude 5°28'19.98"E and longitude 6°26'50.22"E. High human activities were sited in this station. These include, illegal refining of petroleum products, dredging of sand for road construction, loading and offloading of goods and farm products. The station receives effluents discharge from Shell Petroleum Development Company microwave station located directly opposite the community, agricultural runoff, and domestic and sewage waste discharge flows into this station. Station four (4) is located in a community called Odhiogbor in the downstream of the River. It lies within latitude 5° 1'36.52"N and longitude 6°26'23.04"E. The distance between this station and station one was about five kilometers. The station is equally characterized by high anthropogenic activities, such as illegal refining of petroleum products, loading and offloading of goods and farm products, fecal discharge, agricultural runoff, domestic waste and sewage discharge from the local dwellers. Station five (5) was sited in a community called Odereke in the downstream of the River. It lies within latitude of 5° 0'42.11"N and longitude of 6°25'50.47"E. Human activities that were sited in this station include, illegal refining of petroleum products, fishing, loading and offloading of petroleum products and dredging of sand.

2.3 Collection of fish specimen's

Ten (10) Samples of *Tilapia zilli* were collected on a monthly basis for 12 months (between May 2016 and April 2017) at Oshiobebe, Akinima, Mbiama, Odhiogbor and Odereke along Orashi River using the services of artisanal fishermen fishing with the aid of basket trap fishing gear.

2.4 Analysis of Samples

In the laboratory the soft tissues (intestine, muscles, liver, gills) of *T. zilli* was air - dried at room temperature for two weeks. The air-dried soft tissues were grounded to powder form, sieved, weighed and ashed at 77 °C for two hours in a furnace. Ten grams (10 g) of ashed tissues were digested with 20 ml of concentrated HNO₃ to bring the metal into solution and then transfer to 100 ml plastic can for Atomic Absorption Spectrophotometer (AAS) analysis. Heavy metals were determined using Atomic Absorption Spectrophotometer (model GBC scientific AASGF 3000) according to APHA, (1998).

2.5 Transfer Factor Index

Health risk assessment of humans consuming the studied fish species was evaluated using transfer factor index (T_F). The transfer factor index is an approach based on the water - fish transfer factor that provides a straightforward, constructive method for assessing heavy metal accumulation for the purpose of health risk assessment of humans consuming the fish. The water - fish transfer factor (T_F) of the biological accumulation coefficient (BAC), which expresses the ratio of contaminants concentration in fish to the concentration in water, was used to characterize quantitatively the transfer of an element from water to fish (Rodriguez, *et. al.* 2002; Tome *et.al.* 2003) using the formular;

$$T_F = M_{\text{tissue}} / M_{\text{water}}$$

Where,

M_{tissue} is the metal concentration in fish tissue

M_{water} is the metal concentration in water

2.6 Statistical Analysis

Mean values (±SE) of triplicate experiment were taken for each analysis. One-way analysis of variance (ANOVA) and Least Significant Difference (LSD) test were employed to separate significant differences in mean values computed for upstream and downstream.

3. Results

Table 1 and Table 2 shows the mean values of trace metal concentrations observed in the tissues of *T. zilli* for upstream and downstream along Orashi River during the study period. However, it was observed that metals were above the WHO (2011) permissible limit for seafood in the intestine exception of chromium which was below detectable limit (BDL). In the muscles most of the trace metals were within the tolerable range for seafood consumption exception for Cd, Fe and Zn which were above the threshold level as recommended by WHO, 2011. The levels of elemental concentrations of trace metals in the liver and gills recorded a similar pattern of results were all the metals were within the acceptable range

exception of Fe and Cd that were above the permissible limit. Chromium was below detectable level in the intestine and muscles, while similar observation was recorded for lead which was also found to be below detectable limit in the liver and gills of *T. Zilli* during the study duration.

The elemental concentrations of trace metals in the liver and gills was however low when compared

to the values obtained in the intestine and muscles of the same species. Significant variations for upstream and downstream were observed for Cd, Cr, Cu, Pb, Mn and Zn ($p < 0.05$) exception for Iron ($p > 0.05$). The transfer factor of trace metals for upstream and downstream in the tissues of *T. zilli* during the study duration is presented in Fig 2.

Table 1: Mean (\pm) Standard Error of Trace Metal Concentration (mg/kg) in the Organs of *Tilapia zilli* obtained from Orashi River for upstream (May, 2016 – April, 2017).

Trace Metals	Units	Mean \pm S.E (Intestine)	Mean \pm S.E (Muscles)	Mean \pm S.E (Liver)	Mean \pm S.E (Gills)	WHO Permissible Limit
Cadmium	mg/kg	0.70 \pm 0.98	0.59 \pm 0.93	0.18 \pm 0.02	0.34 \pm 0.06	0.01
Copper	mg/kg	3.05 \pm 0.23	0.81 \pm 0.37	0.10 \pm 0.03	0.63 \pm 0.07	1.0
Iron	mg/kg	19.40 \pm 1.83	9.80 \pm 1.62	3.04 \pm 0.87	12.17 \pm 0.57	0.3
Lead	mg/kg	0.56 \pm 0.92	0.05 \pm 0.03	BDL	BDL	0.05
Zinc	mg/kg	31.49 \pm 2.27	9.39 \pm 3.18	2.48 \pm 0.19	2.41 \pm 0.22	3.0
Manganese	mg/kg	2.62 \pm 0.53	1.35 \pm 0.29	0.50 \pm 0.07	0.44 \pm 0.07	-
Chromium	mg/kg	BDL	BDL	0.17 \pm 0.02	0.05 \pm 0.15	0.05

Table 2: Mean (\pm) Standard Error of Trace Metal Concentration (mg/kg) in the Organs of *Tilapia zilli* obtained from Orashi River for downstream (May, 2016 – April, 2017).

Trace Metals	Units	Mean \pm S.E (Intestine)	Mean \pm S.E (Muscles)	Mean \pm S.E (Liver)	Mean \pm S.E (Gills)	WHO Permissible Limit
Cadmium	mg/kg	0.10 \pm 0.77	0.10 \pm 0.68	0.05 \pm 0.03	0.06 \pm 0.04	0.01
Copper	mg/kg	2.67 \pm 0.20	0.43 \pm 0.24	0.19 \pm 0.03	0.42 \pm 0.09	1.0
Iron	mg/kg	10.83 \pm 0.56	9.07 \pm 1.73	4.43 \pm 0.83	6.06 \pm 1.18	0.3
Lead	mg/kg	0.08 \pm 0.56	0.00 \pm 0.00	BDL	BDL	0.05
Zinc	mg/kg	13.89 \pm 3.27	3.70 \pm 0.92	1.69 \pm 0.24	0.21 \pm 0.75	3.0
Manganese	mg/kg	1.01 \pm 0.51	0.37 \pm 0.17	0.16 \pm 0.07	0.04 \pm 0.15	-
Chromium	mg/kg	BDL	BDL	0.01 \pm 0.14	0.00 \pm 0.02	0.05

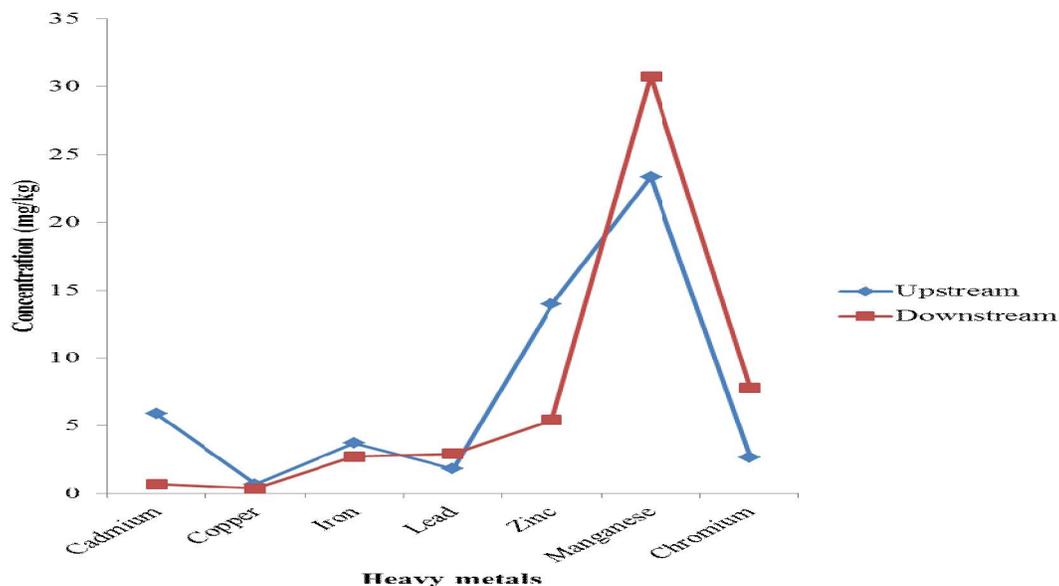


Figure 2: Transfer factor threshold for *Tilapia zilli* obtained from upstream and downstream of Orashi River

4. Discussion

The present study demonstrates evidence of trace metal (Cd, Cu, Fe, Pb, Zn, Mn and Cr) accumulation in organs (intestine, muscles, liver and gills) of *Tilapia zilli* caught from Orashi River during the duration of study. The concentration of metals in the intestine increased accordingly Zn>Fe>Cu>Mn>Cd>Pb for both the upstream and downstream samples. The trend differed with the muscles exhibiting preferential uptake for Fe>Zn>Mn>Cu>Cd>Pb for upstream while downstream had a different trend which was Fe>Zn>Cu>Mn>Cd>Pb. In the liver the trend for upstream and downstream were Fe>Zn>Mn>Cd>Cr>Cu and Fe>Zn>Cu>Mn>Cd>Cr respectively. For the gills, differential uptake of trace metals was observed in the tissues with slight variations as regards the sampling stations (upstream and downstream) which followed the trend Fe>Zn>Cu>Mn>Cd>Cr and Fe>Cu>Zn>Cd>Mn>Cr respectively. The levels of trace metals in the studied tissues were in the order of intestine> muscles> gills>liver during the study duration.

The highest concentration of Cadmium was observed in the intestine for upstream and downstream during the study duration. Cadmium is a nonessential trace metal that is potentially toxic to most fish and wildlife particularly freshwater organism (Robertson *et al.*, 1991). Cadmium is a highly toxic metal present throughout the environment and accumulates in liver and kidney of mammals through the food chain. Cadmium may enter into the aquatic bodies through sewage sludge and through runoff from agricultural lands as in one of the major components of phosphate fertilizers. Cadmium forms a number of inorganic compounds such as sulphates, chlorides and acetates most of which are water soluble. Cd is a by-product of mining and smelting of Pb and Zn and is used in nickel-cadmium batteries and paint pigments. Studies have shown that Cd accumulate mainly (about 75 %) in kidney, liver and gills of freshwater fishes (Chowdhury *et al.*, 2004). However, the concentration of cadmium recorded in the tissue of the studied species exceeded the 0.05 mg/kg (EU standard) and 0.01 mg/kg (WHO standard) threshold considered injurious to fish and humans consuming the fish. Ingestion of Cd from infected species can rapidly cause feelings of nausea, vomiting, abdominal cramp and headache, as well as diarrhea.

Copper is an essential trace metals for all living organisms, and also required by fish and other aquatic organisms as an essential part of their oxygen - carrying pigment hemocyanin (Engel, *et al.*, 1981). Copper pollution is through extensive use of

fungicides, algacides, molluscicides, insecticides and indiscriminate discharge of wastes into aquatic ecosystem. WHO (1989) reported that Copper toxicity in fish is taken up directly from the water via gills and stored in the liver, the present study showed similar accumulation of Copper in the tissues of the *T. zilli*. Effects of high concentration of Copper in fish are not well established; however, there is evidence that high concentrations in fish can lead to toxicity (Woodward *et al.*, 1994). However, the concentration of Copper was observed to be higher than the maximum level of 1.0 mg/kg reported by (WHO, 2011) only in the intestine of *T. zilli*, caught from Orashi River system.

Evidence of iron accumulation was observed in all the studied tissues. However, the highest concentration of iron was recorded upstream in the intestine and gills during the study duration. This assertion is in variance with the findings of Van Rensburg (1989) and Grobler *et al.*, (1991) who reported high bio-concentration of iron in the liver and gonads of *Tilapia sparrmanii*. The high concentration of Fe in the tissues of the studied species is attributed to the abundant nature of Fe in the earth crust and the use of iron coagulants or the corrosion of steel and cast-iron pipes during the process of water distribution.

Lead is toxicity is of great concern due to its ability to bio-accumulate in aquatic ecosystems, as well as persistence in the natural environment (Miller *et al.*, 2002). Lead accumulate in fish tissues such as bones, gills, liver, kidneys and scales, while gaseous exchange across the gills to the blood stream is reported to be the major uptake mechanism (Oguzie, 2003). The low concentration of Lead observed in the tissues of the studied species may be attributed to the absence of human activities related with elevating the concentrations of Pb in the study location during the duration of study. Aquatic organism's bio-accumulates Pb from water and diet, although there is evidence that Pb accumulation in fish, is most probably originated from contaminated water rather than diet (Creti *et al.*, 2010) which may account for the low concentration of lead recorded in all the tissues during the study duration.

Zinc (Zn) is the second most abundant trace element after Fe and is an essential trace element and micronutrient in living organisms, found almost in every cell and being involved in nucleic acid synthesis and occurs in many enzymes (Sfakianakis *et al.*, 2015). This assertion supports the present findings in which zinc and iron had the highest concentration in all the studied tissues. Zinc and its compounds are extensively used in commerce and in medicine. The common sources of it are galvanized

ironwork, zinc chloride used in plumbing and paints containing zinc (Clarke, *et. al.*, 1981). However, the concentrations were above WHO, 2011 threshold limit for intestine and muscles exception of the liver and gills which recorded low values below the recommended limit.

Chromium is an essential nutrient metal, necessary for metabolism of carbohydrates (Faraq *et. al.*, 2006). Chromium enter the aquatic ecosystem through effluents discharged from leather tanneries, textiles, electroplating, metal finishing, mining, dyeing, printing industries, ceramic, photographic and pharmaceutical industries etc. (Abbas and Ali, 2007). Poor treatment of these effluents can lead to the presence of Cr (VI) in the surrounding water bodies, where it is commonly found at potentially harmful levels to fish (Li *et. al.*, 2011; Pacheco *et. al.*, 2013). Generally, the concentration of chromium was low in all the studied tissues throughout the duration of study. The low concentration observed during the study could be attributed to the absence of the above mention human activities within the study area.

Manganese tends to reside in the tissues of the studied species, although in minute concentration. However, the concentration of Mn in the intestine and muscles exceeded the guideline limit of 0.7 mg/kg set by (Charbonneau & Nash, 1993) exception of the liver and gills were lower concentrations were recorded.

The result of this study shows imminent problems of contamination in Orashi River which suggest evidence of human activities. The High concentration of Fe, Cu, Cd and Zn recorded in the tissues of the studied species and also, evidence of bioaccumulation using transfer factor index calls for concern as *T. zilli* obtained from Orashi River is contaminated and calls for concern. This study emphasizes the essence of constant monitoring of trace metal levels in tissues of edible aquatic organisms to prevent sub-lethal poisoning to man as the final consumer of this seafood's along the food chain. This study confirms earlier assertion by Inyang-Etor and George, (2018) during their studies on the levels of trace metals concentration in *Tympanotonus fuscatus* Obtained from Qua Iboe River Estuary, South - South, Nigeria.

5. Conclusion

The elemental concentrations of trace metals observed during the study were higher in the intestine and muscles of *T. zilli* during the study duration. Lead and Chromium were below the threshold limit as recommended by WHO (2011) in the tissues of the studies species. This was attributed to minimal or absence of human mediated activities capable of increasing the concentrations of these trace metals in the study area. However, the concentrations of heavy

metals observed in the gills were below the WHO threshold limit exception of iron which was considerably higher than the WHO recommended threshold limit for seafood consumption. Transfer factor index for *T. Zilli* showed evidence of bioaccumulation of heavy metals in the tissues of this organism. It was observed that most of the studied trace metals had a transfer factor index > 1 which demonstrate evidence of bioaccumulation. From the result of findings, the water quality of Orashi River is severely impacted by human activities resulting from indiscriminate discharge of domestic waste, industrial waste, agricultural run-off and sewage disposal into the river. The high concentrations of heavy metal in some of the tissues of *T. zilli* calls for concern as this may result in deleterious health effects to consumers of these fishes' overtime. *T. Zilli* is a common commercial finfish consumed in Nigeria, notably the Niger Delta Region by most rural dwellers and riverine communities owing to their cheap source. However, this study emphasizes the need of constant monitoring of trace metal levels in tissues of edible aquatic species to prevent health related issues to Humans as the final consumer of this seafood's.

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