

Bioaccumulation of Zn in Muscle and Brain Tissues of the African Catfish—*Clarias gariepinus*

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Abstract

Increasing rate of consumption of the common African catfish, *Clarias gariepinus*, a popular delicacy in Delta State, Nigeria has raised worries about the safety of health of consumers in the face of perceived rising input of recalcitrant pollutants such as the heavy metals in aquatic habitats. This research investigated the presence and levels of Zn in muscle and brain tissues of *C. gariepinus* sourced from selected markets in Delta State. Replicate adult fish samples were obtained from seven market locations in Oleh, Asaba, Ekpan, Ogwashi-Ukwu, Okere, Abraka and Ughelli towns, labeled and taken to the laboratory in iced coolers. The presence and levels of the heavy metal were determined spectrophotometrically. Concentrations ranged from 0.015 - 0.19 (0.09 ± 0.02) mg/kg d.w. in muscle and 0.035 - 0.36 (0.16 ± 0.03) mg/kg d.w. in brain tissues, even as accumulation levels differed significantly ($t = 0.005$) between the tissues at $p < 0.05$. There was also significant locational heterogeneity in accumulations of the metal [$F_{(100.97)} > F_{crit(4.02)}$] at the 95% confidence limit; with least accumulation of 0.03 (± 0.01) mg/kg d.w. recorded in fish samples obtained from Ughelli and maximum accumulation of 0.28 (± 0.07) mg/kg d.w. recorded in those obtained from Oleh locations. However, levels were below the Food and Agricultural Organization and World Health Organization acceptable limits for Zn in edible fish. Results revealed that lipophilic brain tissues accumulated more heavy metal than muscle tissues. Since accumulation levels were low, they do not currently constitute public health risks to consumers in Delta State.

Keywords

African Catfish, Heavy Metals, Tissue Accumulation, Delta State, Local Delicacy

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1. Introduction

In inland aquatic ecosystems, fish species have severally been utilized as biomonitors of heavy metals accumulation [1]-[3]. This is because fish absorb toxic pollutants from the surrounding aquatic environment [4], depending on a variety of factors such as the characteristics of the species under consideration, the exposure period, the concentration of the element, as well as abiotic factors such as temperature, salinity, pH and seasonal changes. Hence, harmful substances like heavy metals, released by anthropogenic activities could be accumulated in aquatic organisms through the food chain and as a result, human health can be at risk when fish contaminated with these toxic chemicals are consumed.

Delta State, one of the highly industrialized states in Nigeria, is located in the oil-rich Niger Delta region with heterogeneous habitats and characteristic rich aquatic biodiversity. Fish are widely consumed mainly because they are part of the local delicacy, and also because of their high protein, low saturated fat and omega fatty acids contents that are known to contribute to good health [5].

In this State, the main pollution sources are industrial production lines, which effluents and wastewaters are channel into nearby rivers, streams and ponds. This waste disposal option is seen by the industry operators as cost effective, even though it does not put environmental safety into consideration. Some of the discharged effluents could contain toxic chemicals such as the heavy metals from improperly treated wastewaters.

However, a variety of industrial and other commercial activities in Delta State include refineries, metallurgical productions, electricity power stations, engineering and other domestic facilities. There are also a number of agricultural establishments, markets and other domestic waste (including heavy metals) generating centres that empty into the surface waters.

Contributing to this is an ever increasing population of inhabitants of the area who are attracted by the industries. This increase has caused more fish to be consumed, even as more introductions of toxic pollutants such as the heavy metals from industrialization could threaten the suitability of the aquatic food. However, whether these heavy metals introduced in the aquatic environments are bioavailable in tissues and organs of the commonly consumed fish delicacies or not is still unconfirmed as there exist paucity of ground-truthing data on bioaccumulation status of the recalcitrant metal species in the State. This work therefore attempted to close this gap in knowledge, by investigating the presence of Zn in tissues of the commonly consumed African catfish, *Clarias gariepinus* sourced from the open waters of the area.

2. Materials and Methods

2.1. Study Area

Delta State is located in the Niger Delta region of Nigeria, within the geographical coordinates 5°30'N and 6°00'E. The climate is typical of the rainforest zone of the tropics and average precipitation is about 200mm. Mean ambient temperature is 28°C, with relative humidity of about 88% [6] [7]. Wet season lasts between March-November, with a short dry season covering the rest of the year. Oil exploration and production operations have been ongoing for over 40 years in the area, even as the major activities of inhabitants include farming, hunting, petty trading/business, and artisanal labour. However, some inhabitants are civil servants.

2.2. Sampling and Sampling Locations

Fish samples were obtained from seven spatially located markets in Asaba (Oshimili South Local Government Area), Oleh (Isoko South Local Government Area), Ekpan (Uvwie Local Government Area), Ogwashi-Uku (Aniocha North Local Government Area), Okere (Warri South Local Government Area), Ughelli (Ughelli North Local Government Area) and Abraka (Ethiope East Local Government Area) of Delta State (Figure 1). The locations were chosen in such a way as to cover the industrial hubs of the state. Fish samples which were of the adult stages (approximate mean body weight = 1.2 ± 1.2 kg) were collected in replicates from each market location. Samples were stored frozen until analysis.

2.3. Laboratory Analysis

The methodologies employed for analysis were in accordance with standard methods as provided by Copat *et al.* [3]. One gram, each of muscle and brain tissues per fish was mineralized in a microwave system (Ethos TC, Milestone S. R. I. Italy) after tissue digestion (using a heated mixture of concentrated HNO₃). A digestion solution

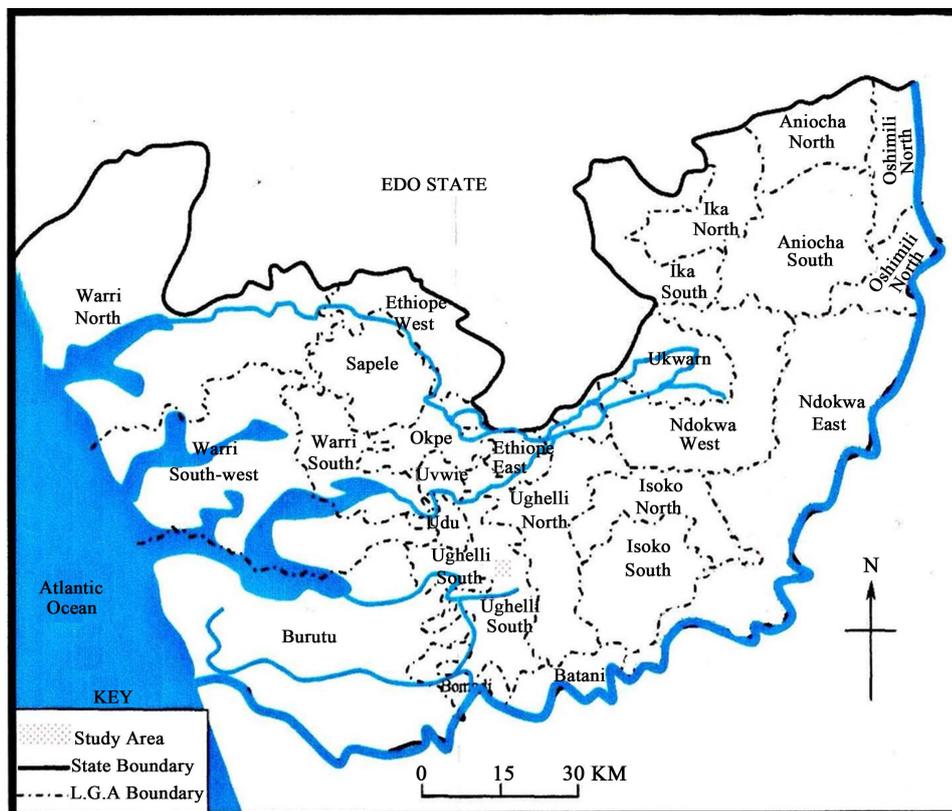


Figure 1. Map of delta state showing the local government areas of study. Source: Ministry of lands and survey and urban development, Asaba, 2002.

was prepared with 6 mL of the nitric acid 65% (Carlo Erba) and 2 mL of hydrogen peroxide (H₂O₂) 30% (Carlo Erba) with a 50 min operation cycle at a temperature of 200°C. After mineralization, distilled water was added to the samples to make up to 20 mL. The mixture was used for the quantification of Zn with a flame ionization atomic absorption spectrophotometer (Varian 600 Spectr AA). Analytical blanks were run in the same way as the samples and concentrations determined using standard solutions prepared in the same acid matrix. Standards for the instrument calibration were prepared on the basis of mono-element certified reference solution ICP Standard (Merck).

2.4. Statistical Analysis

The MS Excel 2007 and SPSS[®] v.22.0 softwares were used in the analyses of data. Descriptive statistics was used to compute mean, standard error, minimum, maximum as well as range of data sets. The student's t-test was used to compare mean accumulations of heavy metals in tissues of fish samples. A test of homogeneity in mean accumulations across the sampling locations was conducted with the single factor ANOVA. Post-hoc structure of means was detected with means plots.

3. Results

3.1. Accumulation of Heavy Metal in Tissues of Fish

The accumulation levels of the heavy metal in the tissues varied widely on comparative scale at the Oleh (range = 0.32 mg/kg dry weight) and Okere (range = 0.34 mg/kg dry weight) locations. At the Oleh, Asaba and Ekpan sampling locations, Zn concentrations varied in the tissues from 0.08 - 0.40 (0.28 ± 0.07), 0.03 - 0.20 (0.10 ± 0.04) and 0.09 - 0.16 (0.12 ± 0.02) mg/kg dry weight (d.w.) respectively (**Table 1**). However, at Ogwashi-Ukwu, Okere, Abraka and Ughelli, Zn concentrations in the tissues varied from 0.04 - 0.09 (0.07 ± 0.01), 0.06 - 0.40 (0.20 ± 0.08), 0.07 - 0.14 (0.11 ± 0.02) and 0.01 - 0.04 (0.03 ± 0.01) mg/kg d.w. respectively.

Table 1. Descriptive statistics of accumulations of zinc (mg/kg d.w.) in muscle and brain tissues of *C. gariepinus*.

Locations	Minimum	Maximum	Range	Mean	SE
Oleh	0.08	0.40	0.32	0.28	0.07
Asaba	0.03	0.20	0.17	0.10	0.04
Ekpan	0.09	0.16	0.07	0.12	0.02
Ogwashi-Ukwu	0.04	0.09	0.05	0.07	0.01
Okere	0.06	0.40	0.34	0.20	0.08
Abraka	0.07	0.14	0.07	0.11	0.02
Ughelli	0.01	0.04	0.03	0.03	0.01

SE = standard error of mean.

3.2. Comparison of Accumulation of Heavy Metal in Tissues

The pair-wise comparison in accumulation of Zn in the muscle and brain of *C. gariepinus* using the Student's t-test of significance revealed that mean concentrations were 0.09 (± 0.02) and 0.16 (± 0.03) mg/kg d.w. in the respective tissues (Table 2).

The test results further shows that accumulation levels of the heavy metal correlated (Sig. $r = 0.000$) and differed significantly (Sig. $t = 0.005$) between muscle and brain tissues of *C. gariepinus*.

3.3. Locational Variation in Accumulation of Heavy Metal in Tissues

At Oleh, Asaba, Ekpan and Ogwashi-Ukwu sampling locations, mean Zn accumulations were 0.19 and 0.36, 0.045 and 0.15, 0.095 and 0.14, and 0.075 and 0.06 mg/kg d.w. in the muscle and brain tissues respectively (Figure 2). However, at the Okere, Abraka and Ughelli locations, accumulation levels in the muscle and brain were 0.155 and 0.235, 0.08 and 0.14, and 0.015 and 0.035 mg/kg d.w. respectively.

The test of homogeneity in mean variance of accumulation levels of the heavy metal across the sampling locations revealed significant heterogeneity [$F_{(100,97)} > F_{crit(4,02)}$] at $p < 0.05$. A post-hoc structure of group means that utilized the Ughelli sampling location as predictor location revealed that in Oleh (Figure 3), Asaba (Figure 4), Ekpan (Figure 5), Ogwashi-Ukwu (Figure 6), Okere (Figure 7) and Abraka sampling locations (Figure 8), accumulations in brain tissue (0.03 & 0.04 mg/kg d.w.) accounted for the observed significant heterogeneity.

4. Discussion

The current work confirms the age long knowledge that recalcitrant pollutants in the environment of organisms could get incorporated in tissues, and also varies in their accumulation levels. Authors such as Ginsberg and Taol [4], Jung and Zauke [8], European Food Safety Authority [9]-[11], United States Environmental Protection Agency [12] [13], and Canli and Atli [1], among others had earlier confirmed this, especially with fish. Delta State, located in the Niger Delta region of Nigeria has fishing as one of the major occupations of her inhabitants. Therefore, its location and heterogeneous habitats- characterized by a rich aquatic biodiversity, makes fish a widely consumed delicacy. Catfishes are widely consumed as part of the local diet and for their high protein, low saturated fat and omega fatty acids content that are known to contribute to good health [5].

Previous studies in Warri River [14], one of the major coastal rivers in the Niger Delta traversing some of the sampling locations, also revealed significant input of pollutants such as the heavy metals and deteriorations by anthropogenic wastes dumped into the surface water habitats of these aquatic organisms. Ebrahimipour *et al.* [15] had also observed leading accumulation of Zn in the freshwater fish, *Carassius gibelio* and *Esox lucius* in Anzali, Iran. However, the range recorded in this study (0.01 - 0.40 mg/kg) was below the Food and Agricultural Organization's acceptable levels for Zn in edible fish (30 mg/kg d.w.) [16]. Values were also below the 40 mg/kg permissible limits for Zn in fish by the World Health Organization [17], as well as the maximum permissible limit of 50 mg/kg by the Joint FAO and World Health Organization food standards [18].

The wide variations recorded in accumulation levels of the heavy metal at the Oleh and Okere sampling locations could represent the peculiar anthropogenic inputs such as from oil and gas and several industrial activities in the locations, which are capable of introducing heavy metals in the enmeshing aquatic ecosystem of the fish.

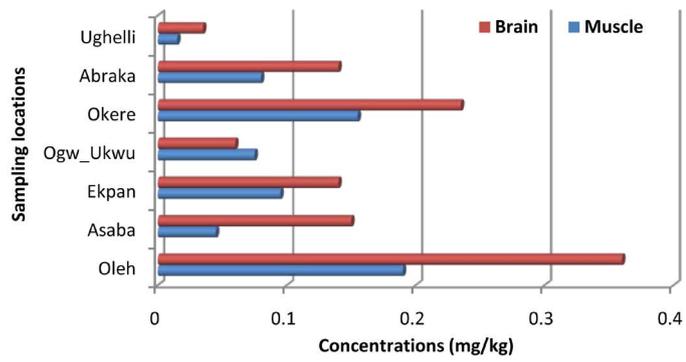


Figure 2. Mean accumulation of zinc ions in muscle and brain of the African catfish, *Clarias gariepinus* from seven markets in Delta State.

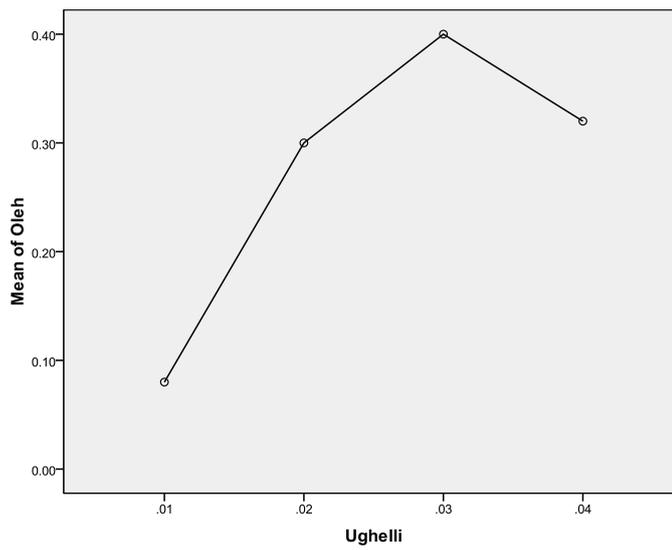


Figure 3. Means plot in accumulation levels of Zn between Ughelli and Oleh locations.

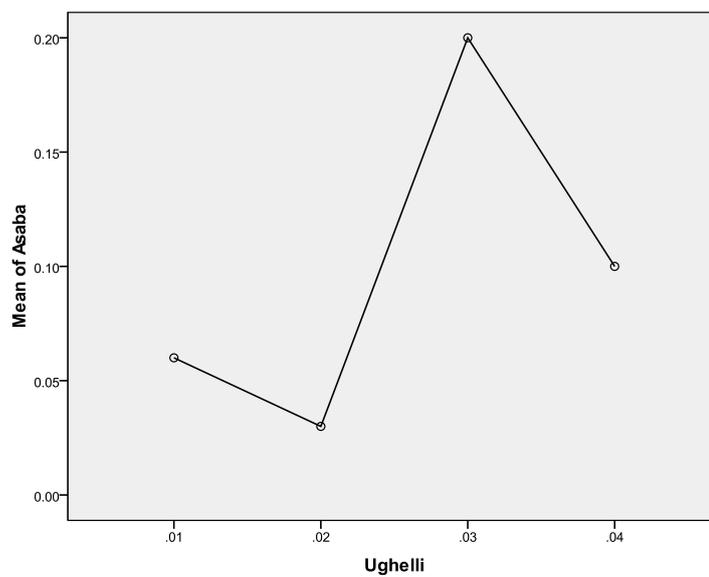


Figure 4. Means plot in accumulation levels of Zn between Ughelli and Asaba locations.

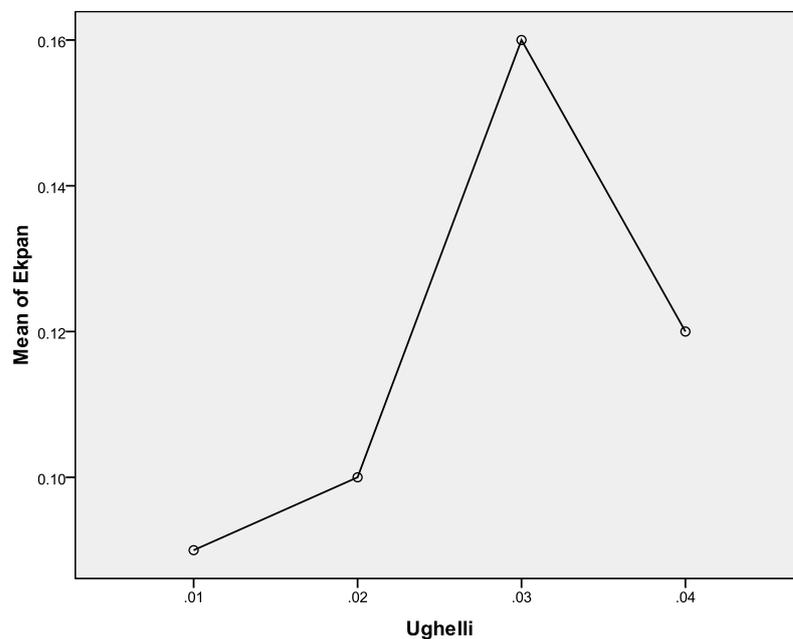


Figure 5. Means plot in accumulation levels of Zn between Ughelli and Ekpan locations.

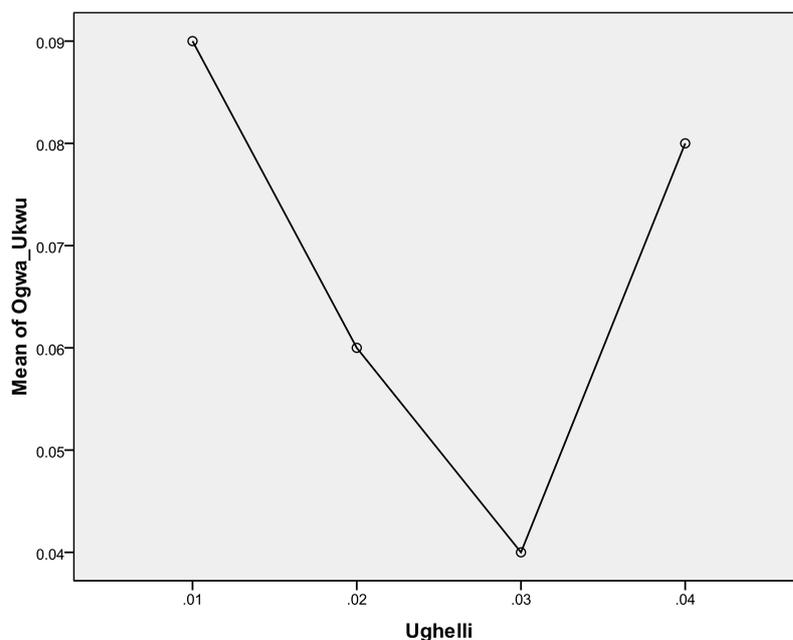


Figure 6. Means plot in accumulation levels of Zn between Ughelli and Ogwashi-Ukwu locations.

Table 2. Pair-wise comparison of accumulations of zinc in muscle and brain tissues of *Clarias gariepinus* from seven market locations in Delta State ($P < 0.05$).

Tissues	Mean	SE	r	Sig. r	t	Sig. t
Muscle	0.09	0.02	0.828	0.000	-3.332	0.005
Brain	0.16	0.03				

SE = standard error of mean.

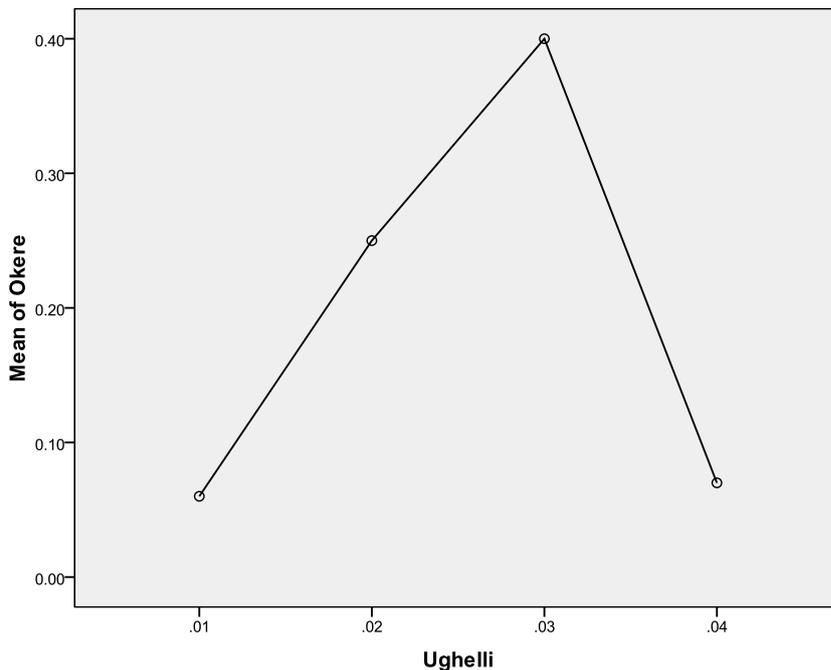


Figure 7. Means plot in accumulation levels of Zn between Ughelli and Okere locations.

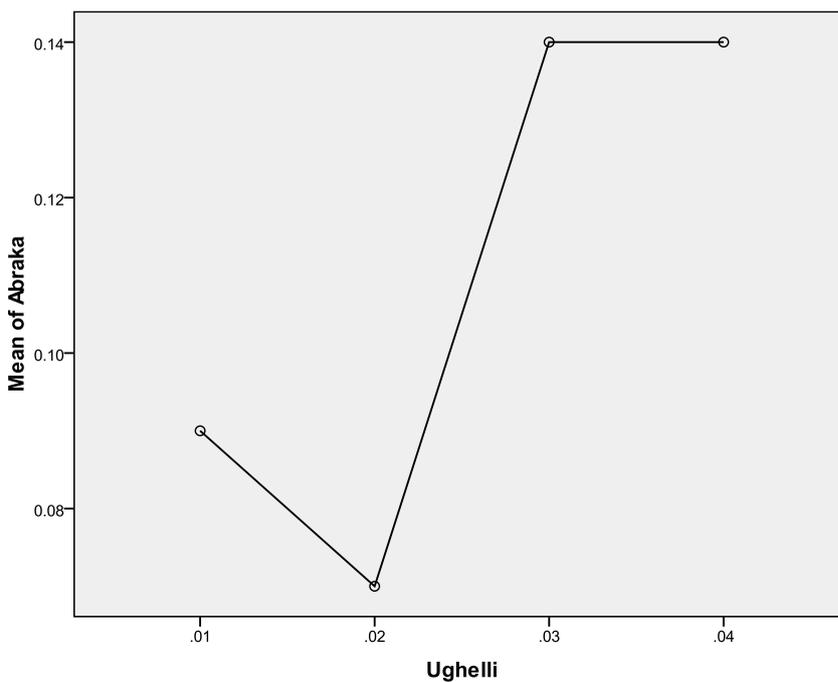


Figure 8. Means plot in accumulation levels of Zn between Ughelli and Abraka locations.

Since the aquatic system also receives some amounts of heavy metals from natural occurring deposits/natural processes, those transported as dissolved species in water or as integral part of suspended sediment originating from substrata may have also contributed to spatial variations recorded in the tissues. Copat *et al.* [3] had also observed spatial variations in heavy metals levels in fish tissues. Accordingly, the highest accumulation of the metal was recorded in fish tissue samples from Oleh, while the least accumulations were in samples from Ughelli. Additionally, spatial variations could also be contributed by the market sellers in their different methods of

handling their fish stock. Certain cutleries used by these sellers are metallic, and so could contribute Zn if it was incorporated in their fabrications.

Findings in this work also reveal that metal accumulations was higher in the brain than muscle tissues; a confirmatory observation to that of Mader [19] and Cox [20] that persistent pollutants (such as the heavy metals) are lipophilic and so could bioaccumulate more in the adipose tissues of organisms. On the other hand, the accumulation of metals in the muscle of fish has severally been observed with different species [2] [3] [21]. Though there was marked difference in accumulation levels of the metal in the muscle and brain tissues, the observed significant correlation also observed in the levels indicate that the heavy metal under study was the same- Zn, in the environment where the fish were caught.

5. Summary

Comparatively wide variations in accumulation levels were recorded in fish tissues sampled from Oleh and Okere sampling locations, with highest accumulations recorded in samples from Oleh and least accumulation recorded in samples from Ughelli location. There was significant difference in accumulation levels in the muscle and brain tissues; with higher accumulations recorded in the brain than muscle tissues.

6. Conclusion

The current work revealed that heavy metal pollution in the fish species sampled was not yet an environmental problem in the study area.

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