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Bioacculumation of Metals in Aquatic Food Chain in Amasiri River, Ebonyi State Nigeria

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ABSTRACT

Background and Objective: There is an unrestrained dumping of wastes and chemicals into water bodies of communities in the South East of Nigeria due to the increasing industrialization. This has led to contamination of the water bodies which serve many puroposes such as recreation, domestics and fishing. The bioaccumulation and biomagnification of some metals from water to fish and then to consumer's plasma and vitreous humour formed the crux of this study. **Materials and Methods:** Water and fish harvested from Amasiri River (Eziyi aku) in Ebonyi State Nigeria and serum and vitreous humour of experimental animals were analyzed for metals such as cadmium, chromium, mercury, lead and arsenic. The sample size of the study was made up of twelve male albino rabbits divided into control and treatment groups. Fish meal prepared from fish harvested from Amasiri River was given strictly to the treatment group. The other animals meals devoid of metals were given to both groups indiscriminately. The metals were estimated by using atomic absorption spectroscopy (AAS) and the choice statistical tool was student t-test analyzed analysed using SPSS version 18-22. **Results:** The concentrations of metals in Amasiri River water were higher than the minimum permissible limit as indicated by the World Health Organization (WHO). Similarly, there was a significant increase in concentrations of cadmium, lead and mercury, whereas chromium concentration decreased in Amasiri River water when compared to the fishes harvested from same river. Concentrations of vitreous cadmium, chromium and lead were significantly elevated in the vitreous of the treatment group when compared with that of the control. In the same vein, serum lead concentration increased significantly in the treatment group when compared to that of the control, whereas serum chromium concentration decreased. **Conclusion:** Conclusively, the findings affirm the bioaccumulation and biomagnification capacity of some metals along the aquatic-food chain cycle with the potentials of long-term systemic disruptions.

KEYWORDS

Metals, bioaccumulation, Amasiri, vitreous, serum

INTRODUCTION

Metals are the metallic chemical element that has a relatively high electric conductivity, malleability, density and is toxic at low concentrations. The metal distribution in the atmosphere, soil, water bodies or living milieu is monitored by the properties of the given metal and the various environmental factors[1].

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Metals are generally referred to as those elements which possess a specific density of more than 5 g cm^{-3} and can adversely affect the environment and living organisms[2]. These metals could be deleterious or useful depending on the type and the concentration. Metals are significant environmental pollutants with preprondrances of distorting the ecosystem and factors supportive to life[3,4]. This study is restricted to metals such as chromium, cadmium, arsenic, mercury and lead. Most of these metals are commonly found in water and could be hazardous biologically and environmentally[5].

Metals get access to the environment through human activities and natural phenomena. The sources are inclusive but not limited to soil erosion, natural weathering of the earth's crust, mining, industrial effluents, urban runoff, sewage discharge, insect or disease control agents applied to crops and many others[6,7]. Although these metals could be deleterious to biological processes, they are sometimes utilized in some metabolic processes that aid life such as homeostasis, transport, compartmentalization and binding to required cell constituents. These metals bind on to protein sites that are not made for them by displacing original metals or molecules from their natural binding sites causing malfunctioning of cells and ultimately toxicity. Previous research has found that oxidative deterioration of biological macromolecules is primarily due to the binding of metals to the DNA and nuclear proteins[8].

Fish derives its nutritional needs from the aquatic environment and in turn expel waste. This course also leads to the intake of toxicants and metals. Dieting is the major orifice of picking up undesirable substances from lower animals by higher ones. The same exposure through dieting culminates to poisoning from the bioaccumulation of toxicants picked up and transported by extracellular and intracellular fluids.

These metals can access human and animal systems through exposure to polluted water by ingestion, inhalation and/or absorption. In the body, these metals are transported in intracellular or extracellular fluids to their respective reservoirs, where they bioaccumulate, resulting in poisoning. Plasma and vitreous humour are amongst the major fluids that aid in the transportation of metals.

Vitreous humour is a transparent fluid located between the lens and the retina filling the centre of the eye[9]. Plasma is the non-cellular component of blood responsible for the transportation of nutrients and waste in the body. These fluids are the major carriers of metals from one part of the body to another and finally to a specific reservoir.

Amasiri River (Eziyi aku) is located in Amasiri. Amasiri is a village in Afikpo North Local Government Area of Ebonyi State. The water body is exposed to pollution resulting from minning, run-offs, deposition of wastes and other undesirable human activities. Regardless of the unhealthy human activities of polluting the water bodies, most residents still depends on it for livelihood such as fishing and other domestic needs. Fishes caught in polluted waters are exposed to toxicants which are transferred to humans via consumption. This activity has the preponderances of bioaccumulating in humans with a long-term devastating effect. Fish chain transfer of metal is a knotty issue in toxicology with attendant systematic disruptions as posited in the body of scientific literatures[10-12].

Based on the argument on the bioaccumulation and biomagnification of metals in the fish-food chain, this study was designed to interrogate the narrative using Amasiri as a case study and rabbit as an animal model.

MATERIALS AND METHODS

Study location: The experimental aspect of the study was carried out at the Department of Biochemistry of the Federal University Otuoke, Bayelsa State from September, 2020 to June, 2021. Similarly, the laboratory analysis phase of the study was conducted at the Eni-Yimini Laboratory (eL) LTD, Yenagoa

Bayelsa State. Amasiri River (Eziyi aku) was the location where the water and fish samples were harvested. Amasiri is a town in Afikpo North Local Government Area under Amasiri Development Centre of Ebonyi State, Nigeria.

Research design/sample population: The quantitative experimental design was the choice for this study. This design uses a scientific method to establish a cause-effect relationship among a group of variables (control and test). Mead's resource equation was utilized for the calculation of the sample size[13]. A total of twelve rabbits constituted the sample size divided equally into control and treatment groups. Both groups were treated similarly except for the exclusion of fish sourced from River Amasiri from the diet of the control. Approximately 0.5 kg dried fish feed was introduced into the treatment cage daily. However, individual quantitfication was not possible due to the group feeding. The treatment regime lasted for three months excluding the 2 weeks of acclimatization.

Rabbit was the choice animal model for the research work. The suitability of rabbit as a choice animal for this study is attributed to its anatomical and physiological similarities to human[4-15]. Similarly, the choice postmortem sample used for the study was vitreous humour. Vitreous humour is seen in the field of forensic science as better and more reliable as compared to other samples. Its composition is rarely altered over the course of aging[16]. The effect of sex or gender on vitreous humour is negligible[16] and also resists putrefaction longer than other body fluids[17].

Selection criteria: Rabbits for the study were obtained from the Biochemistry Department of the University of Port Harcourt, Rivers State. Rabbits used for the study were confirmed to be healthy and active by the university veterinarian. Exclusion criteria were limited to rabbits exhibiting signs and symptoms of illness. The age ranges of the rabbits were between six to eight months and weight brackets were 1.5-2 kg.

Ethical approval: The ethical approval was sort and obtained from the Directorate of Research and Quality Assurance of the Federal University Otuoke, Bayelsa State. To ensure adherence to best practices, the Animal Welfare Act of 1985 of the United States of America for research and Institutional Animal Care and Use Committee (IACUC) protocols were followed[18].

Collection of samples: Prior to the samples collection, the animals were humanely induced using chloroform.

Vitreous humour and blood samples were extracted using the method of Agoro *et al.*[19] and Agoro[20], respectively. The samples were collected within 30 min postmortem. Averagly, the middle of the river was used for the study spanning about 1.5 km. The choice locations were mapped and samples collected for every 100 m apart in a cross sectional pattern. One litre of water were collected from ten locations along River Amasiri into sterile containers. The water samples were accordingly transported to the laboratory for metal analysis. The catfish harvested for the study was *Clarias gariepinus* as speciated by the Department of Biology of the Federal University Otuoke, Nigeria. Thirty catfish of averagly 1 kg each measuring 20-24 inches long were used for the study. The fishes were washed, heat-dried and grinded before being used. Aseptic methods were strictly followed to avoid contamination during the collection and processing stages. A total of 0.5 g portion of the fish was used for metal estimation, whereas the remaining 20 kg included in the diet of the treatment group through out the experimental stage.

Laboratory procedure: Vitreous humour and serum samples with a total volume of 1.2 mL each were thawed at room temperature and each sample was treated separately. A volume of 300 µL of samples was added to 300 µL of HNO_3 and 100 µL of H_2O_2. Sample decomposition was then carried out in a water bath at 80°C for 30 min. After digestion/decomposition, samples were diluted to 10 µL with deionized water.

Similarly, water and fish samples were weighed and digested with Aqua-regia for 1 hr. The samples were allowed to cool and filtered into a 50 mL volumetric flask after digestion was completed. The digest from all the prepared matrices was then analyzed using Varian Spectra A100 Atomic Absorption Spectroscopy (AAS) for Pb, Cd, Cr, Hg and As.

Statistics: Data were analyzed with Statistical Package for Social Science (SPSS) program version 22 (SPSS inc., Chicago, Illionois, USA, Version 18-12) and Microsoft excel. Student t-test was used in comparing the means of the two groups. Data were presented as Mean±Standard Deviation and the level of significance was pegged at less than or equal to 0.05.

RESULTS

Figure 1 indicated an elevation of the mean concentrations of metals in Amasiri River water when compared to the minimum permissible limit as indicated by WHO. The cadmium concentrations (mg L^{-1}) were 0.5128 and 0.003 for Amasiri River water and the WHO standard, respectively. Chromium concentration was 0.07395 mg L^{-1} whereas the WHO concentration was 0.05 mg L^{-1}. Arsenic concentration in the Amasiri River was 0.6850 mg L^{-1} as against the WHO standard of 0.010 mg L^{-1}. Lead and mercury concentrations (mg L^{-1}) in Amasiri water were 0.9700 and 0.0445 when compared with the WHO standard of 0.010. Table 1 exhibited a significant increase ($p<0.05$) in cadmium (0.5128±0.0865 and 0.2291±0.0864), lead (0.9700±0.0548 and 0.6385±0.1092) and mercury (0.0445±0.0985 and >0.0001) concentrations (mg L^{-1}) in Amasiri River water when compared to the fishes harvested from the river, whereas chromium (0.07395±1.0050 and 1.0783±0.8304) was on the contrary, respectively. Table 2 showed a significant increase in concentrations (mg L^{-1}) of vitreous cadmium (0.0740±0.0017 and 0.0020±0.0008), chromium (0.0130±0.0045 and 0.0020±0.0008) and lead (0.0710±0.0094 and 0.0020±0.0082) concentrations (mg L^{-1}) when vitreous of the treatment group was compared to that of control. Serum lead (0.0850±0.0096) increased significantly ($p<0.05$) in the treatment group when compared to that of the control (0.0250±0.0075), whereas serum chromium (0.6170±0.0587 and >0.0001) took a reverse in Table 3.

Table 1: Comparison of some metal concentrations between water and fish sourced from Ebonyi State

Metals	Water	Fish	t-test	p-value
Cadmium (mg L^{-1})	0.5128±0.0865	0.2291±0.0864	4.642	0.004
Chromium (mg L^{-1})	0.07395±1.0050	1.0783±0.8304	-810	0.449
Arsenic (mg L^{-1})	0.6850±0.08448	0.5829±0.0780	1.777	0.126
Lead (mg L^{-1})	0.9700±0.0548	0.6385±0.1092	5.429	0.002
Mercury (mg L^{-1})	0.0445±0.0985	>0.0001	11.228	0.000

Table 2: Comparison of some metal concentrations between vitreous of control and treatment

Metals	Vitreous control	Vitreous treatment	t-test	p-value
Cadmium (mg L^{-1})	0.0020±0.0008	0.0740±0.0017	-8.485	0.000
Chromium (mg L^{-1})	0.0020±0.0008	0.0130±0.0045	-4.763	0.003
Arsenic (mg L^{-1})	>0.0001	>0.0001	-	-
Lead (mg L^{-1})	0.0020±0.0082	0.0710±0.0094	-14.601	0.000
Mercury (mg L^{-1})	>0.0001	>0.0001	-	-

Table 3: Comparison of some metal concentrations between serum of control and treatment

Metals	Serum control	Serum treatment	t-test	p-value
Cadmium (mg L^{-1})	0.0027±0.0006	0.0036±0.0015	-1.155	0.292
Chromium (mg L^{-1})	0.6170±0.0587	>0.0001	23.390	0.000
Arsenic (mg L^{-1})	>0.0001	>0.0001	-	-
Lead (mg L^{-1})	0.0250±0.0075	0.0850±0.0096	-9.842	0.000
Mercury (mg L^{-1})	>0.0001	>0.0001	-	-

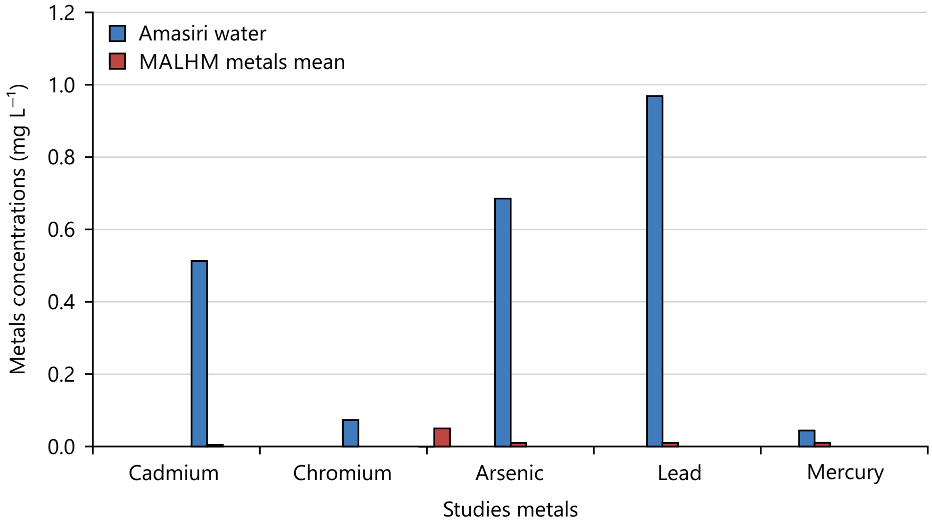

Fig. 1: Mean comparison of some metal concentrations in River Ebonyi and Minimum Acceptable Limits (MALHM)

Chromium sourced from Thomas *et al.*[21], Agoro and Ilesanmi *et al.*[35] and Santos *et al.*[36]

DISCUSSION

The mean minimum allowable concentrations of the studied metals in river water were higher in Amasiri River, Ebonyi State when compared with that of WHO-approved limits as posited by Thomas *et al.*[21] (Fig. 1). Metal concentrations in various water bodies of Ebony State have been extensively studied by researchers. The discourse on metals in water bodies of Ebonyi State has brought about two schools of thoughs. Those that discovered higher concentrations of metals as compared to the WHO minimum allowable limits and those that are of contrary opinion. The first school of thought affirmed presence of metals above the allowable concentrations in various water bodies in Ebonyi State attributing it majorly to minning, indiscriminate waste disposal and other human and geogenic activities[22-27]. The basis of the increase in metal concentrations are the same as advanced above[28,29]. This further affirms the stance of Eyankware and Ephraim[29] on the toxicological effect of lead mining in Amasiri in Ebonyi State. The presence of the metals indicated in this study is in line with that of Rajeshkumar and Li[30] on most commonly found metals in the wastewater. Metals presence above the acceptable minimum limit is deleterious to the health and ecological balance as posited by Jaishankar *et al.*[3] and Nagajyoti *et al.*[4].

In a similar vein, the study revealed a significant increase in concentrations of cadmium, lead and mercury in Amasiri water when compared to the fishes harvested, whereas chromium concentration decreased (Table 1). The significant increase in concentrations of cadmium, lead and mercury cannot be unconnected with the receptacle nature of water bodies. Metals generating materials, substances and affluence are directly dumped in to the water bodies resulting in its buildup. The observation of cadmium, chromium, lead and mercury concentrations in fishes could be as a result of bioaccumulation from the water body due to intake of water, planktons and debris containing the metals. The findings of this study are in line with the positions of Rajeshkumar and Li[30] and Nhiwatiwa *et al.*[31].

Furthermore, the comparison of metal concentrations between the control and treatment vitreous humour revealed a significant increase in concentrations of cadmium, chromium and lead in the latter (Table 2). This could not be unconnected with fish diet sourced from River Amasiri used in feeding the animals of the treatment group. The implication is an assertion of the bioaccumulation potentials of metals in the aquatic consumption chain. Furthermore, it has averred the possibilities of metals accessing the vitreous humour through the various membrane barriers of the retina and other optical tissues. A study conducted on assessing the presence of metals in the eye implicated the accumulation of lead and cadmium in human ocular tissues, particularly in the retinal pigment epithelium and choroid[32]. Pamphlett *et al.*[33] has also discovered toxic metals in the various tissues of the eye.

In same vein, serum lead increased significantly in the treatment group when compared to that of the control, whereas serum chromium decreased (Table 3). The increase in serum lead in the treatment group is a further confirmation of the transferability of metals in the aquatic- food chain to end consumers such as humans and animals. Metals such as lead could build up in the ecosystem and the body over time and cause systemic disruptions. The decrease in chromium in the treatment group could be attributed to displacement by other metals as chromium is essentially important to the body in optimal concentrations. Translocation of metals in similar matrices have also been enunciated by Perello *et al.*[34], Agoro and Ilesanmi[35]. The side effect of this phenomenon has been experienced in Minamata Bay, Japan, in the middle of the 1950s, an illness called Minamata disease[36].

This study has further advanced the course of systemic metal pollution in the environment of Southeast Nigeria. The practice of deliberate dumping of waste into water bodies and the perennial mining activities if not well regulated and monitored could be disastrous over the years with attendant biological disruptions and alterations that could be inimical to the human and the ecosystem at large.

CONCLUSION

This study has revealed the transference and bioaccumulation capacity of some heavy metals from water to fish and finally to the animals or humans which are the end consumers. The observation of some toxic metals in the water bodies, fishes and model animals is an affirmation of the already disrupted fragil environment which could be deleterious. This could be noxious if unchecked, hence strict regulatory measures and remediation should be immediately advanced by all and sundries. Further studies in this line of thought using the human model will broaden the scope of the discourse and bring to bear a robust perspective to this narrative.

SIGNIFICANCE STATEMENT

This study discovers the bioaccumulation capacity of some metals from the water body to fish and finally to the serum and vitreous humour of study animals. This study will help researchers to uncover the critical areas of pathology related to idiopathic causes and the metabolism of metals. The translocational approach of this study to metals has opened a new vista to ophthalmology and toxicity arising from abuse of the environment and the need for strict measures to forestall long-term metal epidemics in Amasiri.

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