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Trace Metals in *Tilapia zilli* and *Clarias batrachus* Fishes Associated with Water and Soil Sediment from River Rukubi in Doma, Nasarawa State, Nigeria

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Abstract

This study was conducted to investigate the pollution level of heavy metals such as Pb, Cr, Ni, Mn, Zn and Fe in the water, sediment and body organs (head, intestine & tile) of *Tilapia zilli* and *Clarias batrachus* from River Rukubi Doma, Nasarawa State. This was carried out using Atomic Absorption Spectrophotometer. Magnesium was the most highly concentrated in the various matrices. Metal levels in the various body organs of the two fishes studied were found to be highly concentrated in the head. Comparison of metal levels in the body parts of the two fishes, sediment and water showed that Cu (85.9%) had the widest variability while Mg (13.1%) was the least. Also comparison of metal levels in the body parts of the *Tilapia zilli* and *Clarias batrachus* showed that Mg has the highest mean metal concentration in *Clarias batrachus* (655.85 µgg^{-1}) than in *Tilapia zilli* (608.40 µgg^{-1}). Iron showed the highest mean metal concentration in *Tilapia zilli* (166.51 µgg^{-1}) than in *Clarias batrachus* (57.20 µgg^{-1}). The heavy metals determined were mostly within the ranged of acceptable limit with exception of Pb, Cr and Mn. In view of this study it is recommended that biological monitoring of fishes meant for consumption from this water body be carried out regularly to ensure human safety. This study contributes to understanding the evaluation of metals level in fishes and their importance in food balanced approach for essential benefits.

Keywords: *Clarias batrachus*, heavy metal, River Rukubi, sediment, *Tilapia zilli*

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Introduction

Fish is a highly perishable food, and smoking has been a preferred cheap method of preservation. This method is carried out over smoldering wood, saw dust, or other sources of energy using traditional oven constructed with locally sourced materials [1]. Fish is a valuable and cheap source of protein to man and dried cat fish has been a favoured delicacy option especially in Nigeria. But the rapid development of industrialization has resulted in heavy metal pollution in our water bodies, which is a significant environmental hazard for invertebrates, fish and humans [1]. Heavy metals tend to accumulate in advanced organisms through bio - magnification along the food chain. Through this, they enter into human tissues, posing serious chronic toxicity. Chronic assimilation of heavy metals is known to cause cancer and can damage vital organs [1]. Ahmad *et al.* found that heavy metals such as Fe, Mn, Cr and Pb pose a number of hazards to humans [1].

These metals are also potent carcinogenic and mutagenic elements. Heavy metal toxicity can result in damaged or reduced mental and central nervous system function, lower energy levels, and damage to blood composition, lungs, kidney, liver and other vital organs. Long term exposure may result in slowly progressing physical, muscular, and Alzheimer's disease, Parkinson's disease, muscular dystrophy, and multiple sclerosis (International Occupational Safety and Health Information Centre [2]. Heavy metal intakes by fish in polluted aquatic environment results in the accumulation of metals in tissues through absorption and humans are exposed through the food web. This causes acute and chronic effect on humans [2].

Fish are an important protein sources for human health because of having rich contents of essential minerals, vitamins and unsaturated fatty acids [2]. The concentrations of heavy metals in sea water, sediments and aquatic organisms such as fish, have been investigated all around the world. Heavy metals are present in the aquatic environment as results of industrial, agricultural and mining activities, etc. The heavy metals can accumulate in the living organisms and thus in the food chain. But the presence of heavy metals in sea water and aquatic products not only threatens aquatic organisms but also may affect human health via the food chain [3]. The bioaccumulation of heavy metals occurs in the various tissues in fish, therefore, may become toxic for public health. Cr and Pb are biologically non-essential metals for living organisms. Fish may contain several chemical contaminants, including heavy metals, and cause a threat to the safety of consumers [3]. The presence of heavy metals in food products is the consequence of environmental contamination of soil and air (plant products), feeding animals with contaminated feed (animal products), acquisition of polluted waters (fish), and other sources (technological water, utensils, pots, etc.) [3].

Many studies have included the determination of heavy metals in fish but they were carried out on fresh fish. Meanwhile, due to the fact that freshwater fish are not customarily eaten fresh, there is a need for determination of heavy metal levels in fish products. During thermal processing, as well as causing loss of weight and water, the application of heat accelerates protein degradation and therefore chemical contaminants may also be affected by the heat applied [4]. These contaminants can cause serious human health problems [5]. They affect the organism in two ways; first is the disruption of normal cell processes that leads to toxicity, and the second one is bioaccumulation [5]. Among heavy metals, cadmium, lead, mercury, arsenic, nickel and chromium are of particular concern [5].

Fish is consumed all over the world for its nutritional value, along with its high quality proteins, great omega-3 fatty acid content, and low saturated fat content and due to having a good level of vitamin-B [5]. It is anticipated that fish contributes about 17% of animal protein and almost 6% of all protein consumed by human beings [5]. However fish become threat to human health because of heavy metals accumulation in marine and fresh water via various urbanization, agricultural and industrial activities [6, 7]. Heavy metals like Cadmium (Cd), Chromium (Cr), Lead (Pb), Iron (Fe), Manganese (Mn), Copper (Cu), Nickel (Ni), Arsenic (As), Mercury (Hg) and Zinc (Zn) have high level of toxicity, and persistence capacity possessing potential for bio magnification, bioaccumulation and incorporation into the food chain after reaching a certain limit in the aquatic environment [5].

Fish also have diverse bioaccumulation characteristics, and accumulation can occur in the head, liver, bones, kidney, stomach, heart, muscle, operculum, vertebrae and flesh of fish species [8]. It happens in different parts of different fish species due to the varying soluble nature of metals in water, bioavailability and the different habitats, life cycles, nature of feeding, ecology and physiological nature of fish [8].

Fish like other food aquatic animals contain essential amino acids, fatty acids, protein, carbohydrates, vitamins and minerals [8]. Vitamins such as calcium, phosphorus and especially iron can be found in fishes.

Among all of these aquatic foods, fish is mostly consumed by humans and became a major part of human diet. It was found out that fish is a source of animal protein; it contains high level of protein around 17-20% with an amino acid profile which is similar to meat [9]. Studying fishes for heavy metal contamination would benefit majority of individuals living in a country surrounded by body of water, and Philippines would be an example for this. Researchers focus their attention in studying heavy metal contamination to fishes to be able help Filipinos in the increasing problem of renal diseases [9]. Long-term Pb consumption can cause mental retardation and even mortality of humans [10]. Non- essential heavy metals (Cr and Pb) even at trace amounts can exhibit extreme toxicity while essential metals (Cu, Zn and Fe) can also cause health problems due to overdosing. As heavy metals cannot be degraded they accumulate in aquatic organisms or animal and are deposited in sediments, it is very essential to assess the health risk assessment of heavy metals in human health through consumption of aquatic animal. The sources of heavy metals in water-bodies include the paint on ships, diverse industrial processes e.g. stabilizers for PVC and agricultural activities such as the use of fungicides, pesticides etc [9]. Where rapid industrialization and mechanized agricultural activities enhance the heavy metal pollution rate the environmental hazards to invertebrates, and vertebrates, including fish and humans are increased [11].

However there is less data available about the presence of heavy metals in fish and other aquatic organism consumed around the area and consequently about their contribution to the dietary intake of their population. Considering this situation it is important to determine the concentration of heavy metals in edible commercially important fish species available in the River Rukubi Doma of Nasarawa State, Nigeria. In this study concentration of heavy metals (Pb, Mg, Zn, Cr, Fe, Mn and Cu) in various organs of two commonly consumed fish namely *Tilapia zilli* and *Clarias batrachus* were determined.

Materials and Methods

Study area

River Rukubi in Rukubi is located in Doma Local Government Area of Nasarawa State, Nigeria. The area lies on latitude 8^{o} 23’42” N and longitude 8^{o} 21’38” E (Fig. 1). The river support domestic use, irrigation and aquatic activities such as fishery, which contribute significantly to the local economy. The river captures water from various tributaries and seasonal rainfall, providing a consistent water supply for the region's farmers especially the OLAM FARM. The Local Government Area in particular and the State as a whole experiences a tropical climate characterized by distinct wet and dry seasons. The wet season typically occurs between April and October, bringing significant rainfall that supports agricultural activities. The dry season, from November to March, is marked by lower rainfall and higher temperatures [13].

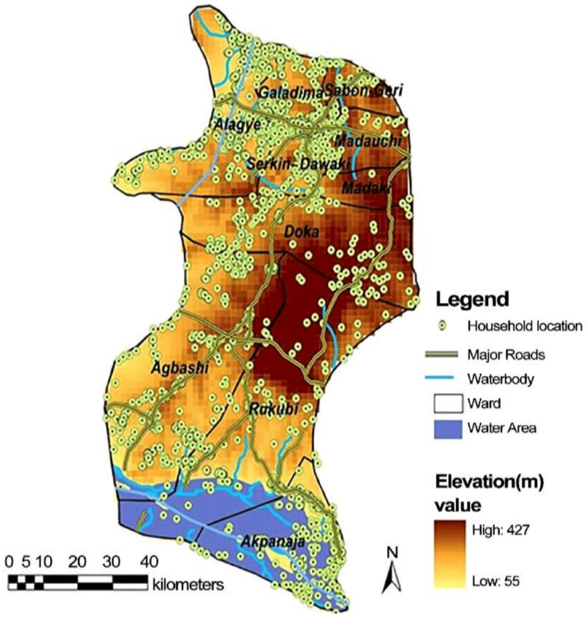

Figure 1: Map of Doma showing River Rukubi the study site

Samples collection

Sample of bottom soil of the river was collected by soil sampler [14]; air dried and kept in polythene bags prior to analysis. The water sample was collected in plastic containers, acidified with 0.01M HNO_3 for preservation [15] and stored in polyethylene bottles prior to analysis. Fish samples of two different species namely, *Tilapia zilli* and *Clarias batrachus* were purchased from fishermen at the river site, then stored at 4^oC prior to analysis.

Tilapia zilli and *Clarias batrachus* were chosen because they appear to have great economic value and are readily available in terms of populations, suggesting their ecological importance in the area. Similarly, these species were chosen with consideration for the sensitivity of aquaculture organisms in terms of heavy metals contamination and accumulation. The fish samples caught freshly were thoroughly washed and put in sterile polythene bags and then placed in an icebox to maintain the freshness and transported immediately to the laboratory for dissection. Before dissection, the mean weight and mean length of the fish samples were measured with measuring tape and weighing balance, respectively. The mean weight was 800 g, while the mean length was 25.5 cm. The taxonomical identification of the fish species was done according to the field guide to Nigerian freshwater fishes [16]. All samples were collected at 7.00 h Green Wich Mean Time (GMT) or 8.00 local time while temperature of the water was 28^o C at the time of collection.

Sample treatment and digestion

Before digestion, air-dried soil sample were ground using a mortar and passed through a 2 mm sieve. 2 g of the soil samples were weighed into acid –washed glass beaker. Soil samples were

digested by adding 20 cm^3 of aqua regia (mixture of HCL and HNO_3, ratio 3:1). The beaker was covered with a watch glass and heated over a hot plate at a temperature of 90^oC for two hours. The beaker wall and watch glass were washed with distilled water and the samples filtered out into flasks to separate the insoluble solid from the supernatant liquid. The volumes were adjusted to 100 cm^3 with distilled water [14, 17]. The water sample was digested using the method of [15].

The fish samples were separated according to species and stored inside the deep freezer at about -100°C and allowed to thaw. Scales were removed and washed with running tap water to remove dirt before being dissected with sterile scissors. The needed fish parts (head, tile and intestine) were oven dried at 70 to 73°C until constant weight was obtained. The specimens were then ground to fine powder and stored in desiccators in order to avoid moisture accumulation before digestion. The digestion procedure was carried out as described by [18]. Twenty milliliter (20 ml) of concentrated nitric acid (55%) and 10 ml of perchloric acid (70%) was added to approximately 1 g tissue (dry mass) in a 100 ml Erlenmeyer flask. The digestion was done on a hotplate (200 to 250°C) until the solutions were clear. The solutions were then filtered through an acid resistant 0.45 mm filter paper and made up to 50 ml each with distilled water.

Heavy metal analysis

The digested samples were poured into auto-analyzer cups, and the concentration of heavy metals (Pb, Mg,Zn, Cr, Mn, Fe and Cu) in each sample was determined using a Varian AA 240 Atomic Absorption Spectrophotometer (AAS) according to the method of American Public Health Association [19]. The absorption wavelengths and detection limits for the heavy metals were noted. The result of the experiment was compared with the Joint WHO/FAO standard for fish consumption.

Statistical analysis

The data collected was analyzed using analysis of variance (ANOVA), and $P < 0.05$ was considered to indicate a statistically significant difference. All with SPSS 20 applications.

Results and Discussion

Mean metal concentration in water & soil sediment

The mean metal concentrations, standard deviation and coefficient of variation percent of water and sediment are presented in Table 1. Iron had the highest concentration in both water (5636.6 μgml^{-1}) and sediment (1138.40 μgg^{-1}) while the least concentrated metal in water was lead (173.1 μgml^{-1}) and in sediment was copper (2.76 μgg^{-1}). However the value of iron (in water) in the present study falls within the World Health Organization (WHO) recommended range [20]. Excessive concentrations of iron are undesirable in domestic water because of problem of staining as a result red colour formation. It also imparts a metallic taste and smell to home water. When compared the levels of metals in the water and sediment samples, the highest variability was found in copper (140.7%) while Fe was the least varied (93.3%).

Table 1: Mean metal concentration in water (μgml^{-1}) and soil sediment (μgg^{-1})

Parameter	Pb	Mg	Zn	Cr	Fe	Mn	Cu
Water	293.1 ±0.10	830.3 ±0.10	173.1 ±0.10	4345.5 ±0.20	5636.6 ±0.20	710.3 ±0.10	1016.3 ±0.20
Sediment	3.03 ±0.10	162.63 ±0.20	4.94 ±0.30	38.11 ±0.20	1138.46 ±0.20	116.47 ±0.10	2.76 ±0.10
Grand mean	148.07	496.47	89.02	2191.81	3387.53	413.39	509.53
SD	205.11	472.11	118.91	3045.79	3180.67	419.90	716.68
CV%	138.5	95.1	133.6	139.0	93.9	101.6	140.7

Mean value ± standard deviation; SD standard deviation; CV% coefficient of variation percent

Heavy metal concentration in fish tissues

The findings of this study showed that the mean concentration of heavy metals in the fish tissues differs significantly amongst the sampled fish species in the River Rukubi (p=0.000). The variation in the level of heavy metals was further explained in Tables 2 – 3 and compared in Fig. 2. The heavy metal levels in the two species of fish *Tilapia. zilli* (Tilapia fish) and *Clarias batrachus* (catfish) include Lead (3.27 - 1.63 μgg^{-1}) magnesium (608.40 - 655.85 μgg^{-1}), zinc (20.34 - 13.55 μgg^{-1}), Chromium (40.63 - 20.81 μgg^{-1}), iron (166.51 - 57.20 μgg^{-1}), Manganese (50.12 - 12.31 μgg^{-1}), and Copper (3.56 - 2.94 μgg^{-1}). The fish tissue (head, intestine, and tile) recorded differences in the heavy metal concentrations.

The study also established that heavy metal accumulations were higher in the head, followed by tile and intestine. In the same vein, the level of heavy metals recorded in *Tilapia. zilli* was higher than in *Clarias batrachus.* This finding agrees with some studies [20, 21]. The differences in metal concentrations can also be attributed to differences in metal uptake, as observed by some researchers [22, 23]. Other probable reasons include fish size and where fish reside in water. For instance, Adebayo reported that heavy metal concentrations were higher in omnivorous/herbivore fish such as *Heterotis niloticus* and lower in carnivore fish such as *Clarias anguillaris* [23].

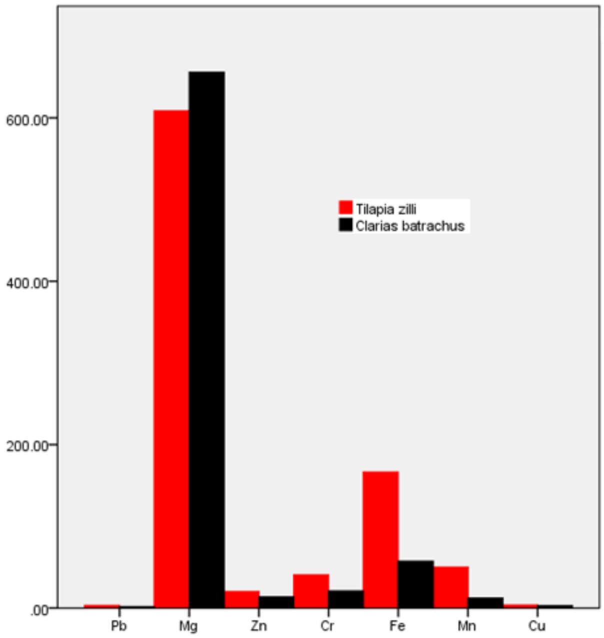

Figure 2: Comparison of metals concentrations in mean body parts of *Tilapia zilli* and *Clarias batrachus* (μgg^{-1})

However, the results are in agreement with the reports of some authors whose observations demonstrated that heavy metal bioaccumulation in various tissues of fishes living in the same water body vary according to the species of fish [24, 25]. The concentration of the metals determined in the body parts of *Tilapia Zilli* followed the order Mg > Pb > Fe > Cr > Cu > Mn > Zn. Fish head recorded the highest level of metals as compared to intestine and tile. The concentration of heavy metals recorded in the fish head, intestine and tile could also be attributed to the high influx of heavy metals in the River Rukubi from anthropogenic activity.

Table 2: Mean metal concentration in body part of *Tilapia zilli* (μgg^{-1})

Parameter	Pb	Mg	Zn	Cr	Fe	Mn	Cu
Head	3.06±0.02	624.84±0.01	24.46±0.02	38.68±0.01	161.59±0.01	53.10±0.58	5.23±0.01
Intestine	1.89±0.01	522.03±0.02	17.73±0.02	64.29±0.01	245.78±0.03	62.53±0.02	5.43±0.02
Tile	4.87±0.01	678.34±0.02	18.82±0.02	18.93±0.01	92.16±0.01	34.72±0.01	0.03±0.10
Grand mean	3.27	608.40	20.34	40.63	166.51	50.12	3.56
SD	1.50	79.44	3.61	22.74	76.93	14.14	3.06
CV%	45.9	13.1	17.8	56.0	46.2	28.2	85.9

Mean value ± standard deviation; SD standard deviation; CV% coefficient of variation percent

Table 3: Mean metal concentration in body part of *Clariasbatrachus* (μgg^{-1})

Parameter	Pb	Mg	Zn	Cr	Fe	Mn	Cu
Head	0.87±0.03	673.85±0.02	17.80±0.10	22.38±0.02	60.03±0.01	17.37±0.01	0.81±0.01
Intestine	1.64±0.02	646.67±0.01	13.56±0.02	8.05±0.02	54.50±0.01	10.19±0.01	2.35±0.01
Tile	2.37±0.02	647.04±0.03	9.28±0.01	32.00±1.00	57.06±0.01	9.36±0.02	5.66±0.01
Grand mean	1.63	655.85	13.55	20.81	57.20	12.31	2.94
SD	0.75	15.59	4.26	12.05	2.77	4.41	2.48
CV%	46.1	2.4	31.4	57.9	4.8	35.8	84.3

Mean value ± standard deviation; SD standard deviation; CV% coefficient of variation percent

Table 4: Level of metals in the body parts of *T. zilli* (μgg^{-1}), *C. batrachus* (μgg^{-1}), sediment (μgg^{-1}) and water (μgml^{-1}) compared.

Sample	Pb		Mg		Zn		Cr		Fe		Mn		Cu	
	T. zilli	*C. batrachus*	*T zilli.*	*C. Batrachus*	*T. zilli*	*C. batrachus*	*T. zilli*	*C. batrachus*	*T. zilli*	*C. Batrachus*	*T. zilli*	*C. batrachus*	*T. zilli*	*C. batrachus*
Head	3.06 ±0.02	0.87 ±0.03	624.84 ±0.01	673.85 ±0.02	24.46 ±0.02	17.80 ±0.10	38.68 ±0.01	22.38 ±0.02	161.59 ±0.01	60.03 ±0.01	53.10 ±0.58	17.37 ±0.01	5.23 ±0.01	0.81 ±0.01
Intestine	1.89 ±0.01	1.64 ±0.02	522.03 ±0.02	646.67 ±0.01	17.73 ±0.02	13.56 ±0.02	64.29 ±0.01	8.05 ±0.02	245.78 ±0.03	54.50 ±0.01	62.53 ±0.02	10.19 ±0.01	5.43 ±0.02	2.35 ±0.01
Tile	4.87 ±0.01	2.37 ±0.02	678.34 ±0.02	647.04 ±0.03	18.82 ±0.02	9.28 ±0.01	18.93 ±0.01	32.00 ±1.00	92.16 ±0.01	57.06 ±0.01	34.72 ±0.01	9.36 ±0.02	0.03 ±0.10	5.66 ±0.01
Sediment	3.03 ±0.10	3.03 ±0.10	162.63 ±0.20	162.63 ±0.20	4.94 ±0.30	4.94 ±0.30	38.11 ±0.20	38.11 ±0.20	1138.46 ±0.20	1138.46 ±0.20	116.47 ±0.10	116.47 ±0.10	2.76 ±0.10	2.76 ±0.10
Water	293.10 ±0.10	293.10 ±0.10	830.30 ±0.10	830.30 ±0.10	173.10 ±0.10	173.10 ±0.10	4345.50 ±0.20	4345.50 ±0.20	5636.60 ±0.20	5636.60 ±0.20	710.30 ±0.10	710.30 ±0.10	1016.30 ±0.20	1016.30 ±0.20
G. mean	61.19	60.20	563.63	592.10	47.81	43.74	901.10	889.21	1454.92	1389.33	195.42	172.74	205.95	205.58
SD	129.65	130.20	250.28	251.92	70.40	72.48	1925.54	1932.16	2375.84	2420.02	289.43	303.89	453.01	453.21
CV%	211.9	216.3	44.4	42.5	147.3	165.7	213.7	217.3	163.3	174.2	148.1	175.9	220.0	220.5

T.zilli Tilapia zilli; C. batrachus Clarias batrachus; Mean value ± standard deviation; SD standard deviation; CV% coefficient of variation percent

A summary of the levels of the various metals on the body parts of the *Tilapia. zilli* and *Clarias batrachus*, sediment and water are shown in Table 4. From the result recorded the various CV% gives an indication of an unequal distribution of the metals in the various matrices.

Differences between heavy metal concentration in the fish tissue and joint WHO/FAO standard limit

The findings of this study also indicate that there is a significant difference between heavy metal concentration in the fish tissue and joint WHO/FAO standard limit (p=0.027). The finding of the study is validated by the result in Table 2 – 3. The heavy metal level in the two species of fish: *Tilapia zilli* and *Clarias batrachus* ranges as follows: Lead (3.27 - 1.63) magnesium (608.40 - 655.85), zinc (20.34 - 13.55), Chromium (40.63 - 20.81), iron (166.51 - 57.20), Manganese (50.12 - 12.31), and Copper (3.56 - 2.94). These set of values differ from joint WHO/FAO standard limit (lead with a value of 0.5 μgg^{-1}, magnesium (500 μgg^{-1}), zinc (40 μgg^{-1}), chromium (0'05 μgg^{-1}), iron (186 μgg^{-1}), manganese (12.97 μgg^{-1}) and copper (30 μgg^{-1})). The concentration of heavy metals (Lead, magnesium, Chromium and Manganese) in fish samples from River Rukubi is higher than the joint FAO/WHO [27] standard for fish diet, except Zn, Fe, and Cu, which are within the standard. However the consumption of these fish species at present may not be a potential health hazard. These findings agree with many studies [20, 30, 31, 26, 28, 29, 12] that reported heavy metal concentration above the World Health Organization, Food and Agriculture Organization (FAO), the European Commission (EC), USEPA, WHO/FAO, UNEP/FAO and FEPA standards. It is important to note that some individuals may consume fish more often, and their dietary intake of these heavy metals would further accumulate, which may lead to elevated health risks. Chronic poisoning with heavy metals (Pb) may lead to anemia and brain damage. Research has shown that long-term low-level Lead exposure in children may lead to diminished intellectual capacity and health risks to pregnant women WHO [28]. It has also been reported that metals such as iron, copper, cadmium, chromium, lead, mercury, and nickel can produce reactive oxygen species. The result of this is lipid peroxidation, DNA damage, and altered calcium homeostasis.

Conclusion

The determination of heavy metals in ***Tilapia zilli*** and ***Clarias batrachus*** fishes, as well as in water and soil sediment from River Rukubi, Doma, Nasarawa State, Nigeria, revealed significant levels of heavy metal contamination. This study showed that the heavy metal concentration in various tissues of fish samples differed significantly. While the levels of Zn, Fe, and Cu are within the standard limit set by the joint FAO/WHO [27], the concentration of Pb and Cr exceeds the limit.

Heavy metal level in the fish at present is a potential hazard, and long-term consumption of the fish from the area may pose a serious health risk to regular fish consumers due to possible bioaccumulation of the heavy metals, especially chromium and lead. Similarly, the water and soil sediment samples showed high concentrations of these trace metals, indicating severe pollution of the river ecosystem.

The findings suggest that the contamination of the river and its inhabitants poses a significant risk to human health, particularly for communities relying on the river for fishing and drinking water. The study highlights the need for urgent measures to address the sources of pollution, such as industrial and agricultural activities, and to implement effective monitoring and regulation of heavy metal levels in the environment.

To harvest fish and fish products that are safe for human consumption, there should be continuous monitoring of heavy metal levels to prevent bioaccumulation above permissible levels. The water standards and concentrations of heavy metals in the River Rukubi, Doma should be monitored routinely to ascertain the heavy metal transfer factor to fish from its environment, as well as the sources to enable sustainable aquatic protection and management practices in the area.

The quality of the fish (both wild catch and aquaculture) for human consumption and the health risks associated with the aquatic products need to be given priority. Various human activities, such as the location of automobile workshops, dumping of domestic sewage, and other activities that are inimical to the safe use of the river, should be discouraged. Appropriate measures, such as legislative provisions and other tools for effective environmental monitoring, should be mounted and used to protect and enhance the quality of the river.

Conflict of interest: Authors have declared that there is no conflict of interest reported in this work.

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