HEAVY METAL CONCENTRATIONS IN WHOLE SOFT TISSUES OF *Anodonta implicata* (CLASS: BIVALVIA) FROM MAKERA INDUSTRIAL DISCHARGE POINT OF KADUNA RIVER, NIGERIA

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Abstract

The whole soft tissues of forty sampled *Anodonta implicata* of different mean weight range (15.0±0.7-100±0.1 g) from a major industrial discharge point of Kaduna River were analyzed for four heavy metals by use of Atomic Absorption Spectrophotometer. The mean concentrations (ppm) of lead, zinc, copper and cadmium were 0.0494±0.01, 1.5866±0.10, 0.4792±0.24 and 0.0054±0.00, respectively. The differences in tissue weights of the bivalve did not have any significant influence on the concentrations of the different heavy metals ($p \geq 0.05$). There was no variation in concentrations of the metals between rainy and dry seasons. The strong positive relationship of the different weight categories of *A. implicata* is an indication of a similarity in the pattern by which the heavy metals concentrate in the soft tissues. This similarity could be attributed to interactions by the heavy metals in the uptake/accumulation through synergic and antagonistic actions in the soft tissues of the bivalve. The concentrations of Pb and Cd in the different weight categories of the bivalve can be toxic to the organism even at low concentrations. Human consumption or use of the soft tissues as fish bait could constitute a serious health risk. The persistence of Pb and Cd in the biological system could impair the bivalve from performing its ecological roles and cause negative effects on organisms in the higher trophic levels of the aquatic food chain.

Keywords: heavy metals, *Anodonta implicata*, Kaduna River.

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Introduction

Metals are considered very important and highly toxic pollutants in terrestrial and aquatic environments. Atmospheric and river inputs, dredging spoil, direct discharges, industrial dumping and sewage sludge are some of the important contributors to metal pollution in aquatic environments (Valavanidis and Vlachogianni, 2010). Industrial, agricultural and domestic activities (Ajayi and Osibanjo, 1981) are considered as major sources of pollutants that have led to pollution stress on surface waters. As such, many rivers and other freshwater reservoirs in Nigeria have been polluted principally by discharge of untreated wastes (Alinnor, 2005; Fakayode, 2005). Kaduna River serves as a sink for effluents discharged from a number of companies (Bamgboye, 2004). The river was reported to be contaminated with heavy metals like copper (Cu), chromium (Cr), iron (Fe), zinc (Zn), cadmium (Cd), lead (Pb) and mercury (Hg) due to the discharge of untreated waste products from local textiles, breweries and oil refinery (Arah, 1985; Oladimeji, 1987). Mahre *et al* (2007) reported the presence of different levels

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of Mn, Cu, As, Cd, Fe, Zn, Hg and Pb in water and sediment samples from Kaduna River .

Goodherham and Tsyrlin (2002) reported macro-invertebrates as useful and convenient indicators of ecological health of a water body or river. As sedentary organisms, bivalves are well known for their biological features of concentrating heavy metals and other substances in their tissues. Due to their exceptional vulnerability to water pollution, bivalves have been used in bio-monitoring studies of heavy metal pollution in aquatic ecosystems (Blasco *et al,* 1999; Gundaker, 2000; Bonneris *et al*, 2005). However, they have developed mechanisms to protect themselves from toxic effects of heavy metals. Some bivalves can neutralize the toxicity of heavy metals and store toxic materials at cellular levels of the body tissues (Viarengo, 1985). Bivalves can accumulate Cd in their tissues at levels up to 100,000 times higher than the levels observed in the water in which they live (Avelar *et al,* 2000). In small quantities, certain heavy metals are nutritionally essential for a healthy life. The persistence of heavy metals in the environment may lead to contamination of aquatic organisms (Oshisanya *et al,* 2011). Kraak *et al* (1994) observed that heavy metals reduce the performance of bivalve molluscs. A number of adverse effects of heavy metals on the health and productivity of bivalves have been reported (Viarengo, 1985; Krogh and Scanes, 1996). Boyden (1974) reported on biological factors such as reproductive cycle, age and body size of bivalve species as important factors that influence the accumulation of trace metals. Aside the potential for human exposure and increased health risk, quite a number of abnormalities in animal body functions have been associated with heavy metals. Notable are the studies of Moriber (1974), Ademoroti (1996) and Eletta *et al* (2003).

A. implicata is a taxonomically stable species of bivalve that is restrictive in its distribution in freshwater bodies of northern Nigeria. Due to its relative large size and ease of handling, *A. implicata* is collected by local fishermen, the soft tissues scooped and used on fishing hooks as baits in Kaduna River. This study therefore assesses the heavy metal load of the soft tissues of *A. implicata* of different weights from a major industrial discharge point of Kaduna River and establishes the role of the bivalve as a possible route to which heavy metals may get into the aquatic food chain, Figure 1.

Materials and methods

Study area

Kaduna River is centrally located in Kaduna on Lat. 10.52°N and Long 7.44°E (Kaduna, 2004) and serves as the main source of water supply for Kaduna town and associated industries. It also receives wastewater for industrial and domestic sources (Mahre *et al,* 2007). The Makera Drain is the major discharge point of an oil depot, a brewery and two textile industries with waste water flow rate of 21,700 (m^3/d) (Bamgboye, 2004) representing 86.1% of the total industrial waste water flow into the river. Peasant farmers and dwellers along the river coast use the water extensively for irrigation and various domestic activities such as for washing, swimming and drinking most especially during the dry season (Dadi-Mamud *et al,* 2012).

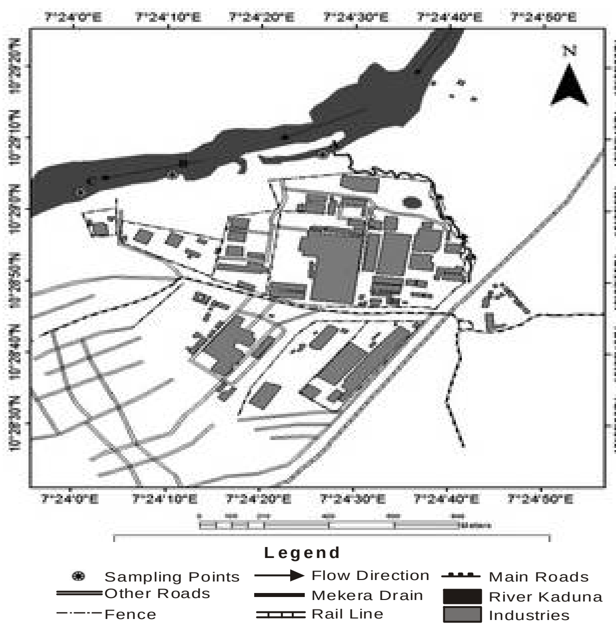

Figure 1. Map of Kaduna River.

Collection of bivalves

Three different sites in a line transect were marked downstream along the bank of Makera discharge point of the river. Samples of *A. implicata* within the mean weight range of 15.0±0.7-100±0.1 g were collected at random using improvised scoop net on the same day every three months, from February 2012 to January 2013, covering rain and dry seasons. The bivalve species from the substratum of each site were sorted by hand-picking, with hands protected in disposable hand gloves. Collected samples were washed at collection sites, placed in labelled plastic containers and identified using standard identification guide (Thompson, 2004).

Sample preparation and metal analysis

The bivalves were weighed and grouped into mean weight-scale (grams) of five categories (15, 28, 45, 61 and 100). The shells of the bivalves were parted open around the hinges and the soft tissues separated from the shell using sharp stainless-steel scalpel. The soft tissues were weighed and macerated using a sharp stainless-steel blade and placed in a crucible and oven dried at 50°C for 48 hours. The dried soft tissues of the bivalves of known weights were digested by wet method in a mixture of 62% Perchloric acid and 70% Nitric acid (1:4) according to Harris (1970). Samples of each weight category were analyzed in triplicate for lead, cadmium, zinc and copper using Atomic Absorption Spectrophotometer (Model: VARIAN AA240FS). The mean concentrations of the heavy metal were calculated.

Statistical analysis

A one-way analysis of variance (ANOVA) was used to determine the variations in concentrations of the heavy metals in the five-weight categories of *A. implicata*. Pearson correlation coefficient was used to establish the relationships of the heavy metals. The student's *t*-test was employed to compare heavy metals concentrations in bivalves for dry season with that of rainy seasons.

Results

Table 1 shows the five weight categories of *A. implicata* within the mean range of 15.0±0.7-100±0.1 g used in this study. The oldest and youngest bivalves belong to weight categories W1 and W5 respectively. Highest percentage of soft tissue was observed in W5 (66.7 %) while W3 was the least with 36.4 % (Table 1). The soft tissues of the bivalve in this study were found to accumulate varying concentrations of the four heavy metals measured. The variations in concentrations of the four heavy metals in the five-weight categories were not significantly different ($p > 0.05$) (Table 2). Relatively high concentrations of Pb (Figure 2) and Cu (Figure 4) were found in the highest weight category of the bivalve (W1). The concentrations of Zn in the different weight groups did not differ significantly in the different weight groups (Figure 3). High accumulation of Cd was observed in magnitude of W5>W1>W4 (Figure 5). With the exception of Zn, the other three heavy metals (Pb, Cu and Cd) showed variability in their mean concentrations for the different weight categories. In this study zinc and cadmium had the highest and lowest concentrations respectively in *A. implicata* (Figure 6). Zinc and Cu showed high mean concentrations (Figure 6) in the whole soft tissues of the bivalves.

The correlation matrix showed both positive and negative interactions among the heavy metals and strong positive relationships among the weight categories (Tables 3 and 4). The *t*-test (t=0.420) shows that the concentrations of heavy metals in the bivalve of different weights did not differ significantly ($p \geq 0.05$) between rain and dry seasons.

Table 1. Mean weights of the five categories of *A. implicata* (n = 40) analyzed for heavy metals.

Weight category	Mean weight (g)	Shell weight (g)	Soft tissue weight (g)	Dried soft tissue weight (g)	Percentage soft tissue (%)
W1	100.0± 0.1	72.0	28.0	26.0	38.9
W2	61.0± 0.2	43.0	18.0	15.0	41.9
W3	45.0± 0.0	33.0	12.0	11.0	36.4
W4	28.0± 0.1	20.0	8.0	7.0	40.0
W5	15.0± 0.7	9.0	6.0	5.0	66.7

Table 2. Variation in concentrations of the metals of the weight categories of *A. implicata*.

Source of variation	Sum squares	*df*	Mean square	*f*-statistic	*p*
Weight Categories	0.3149	4	0.0787	0.13	0.9697
Residual	9.1938	15	0.6129		
Total	9.5087	19			

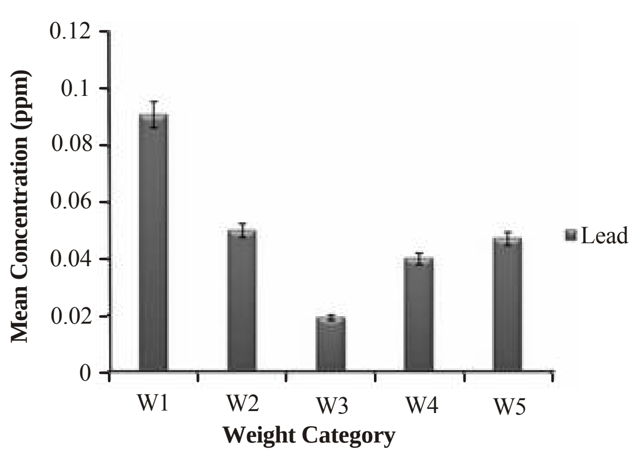

Figure 2. Mean concentrations of lead in the soft tissues of the weight categories of *A. implicata* from Kaduna River.

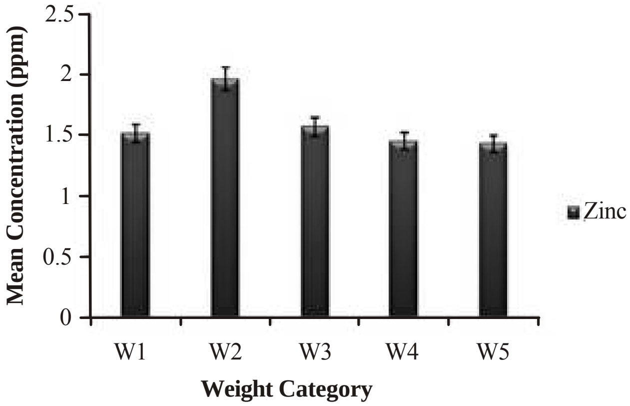

Figure 3. Mean concentrations of zinc in the soft tissues of the weight categories of *A. implicata* from Kaduna River.

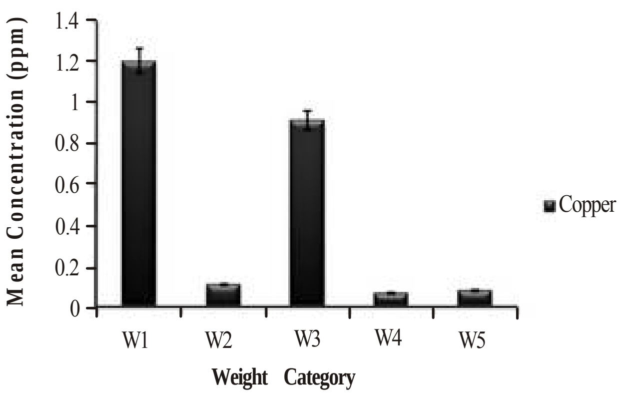

Figure 4. Mean Concentrations of copper in the soft tissues of the weight categories of *A. implicata* from Kaduna River.

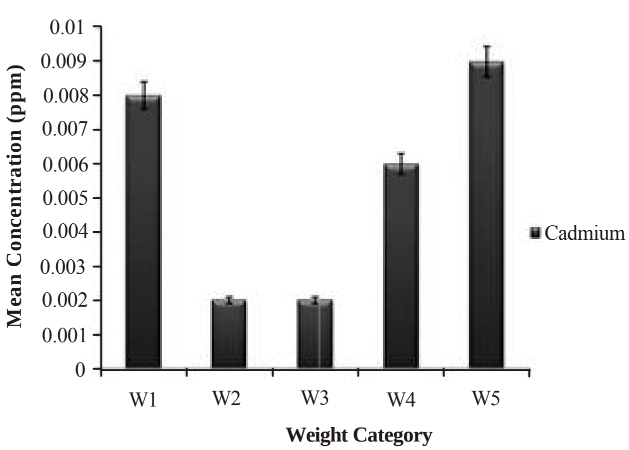

Figure 5. Mean concentrations of cadmium in the soft tissues of the weight categories of *A. implicata* from Kaduna River.

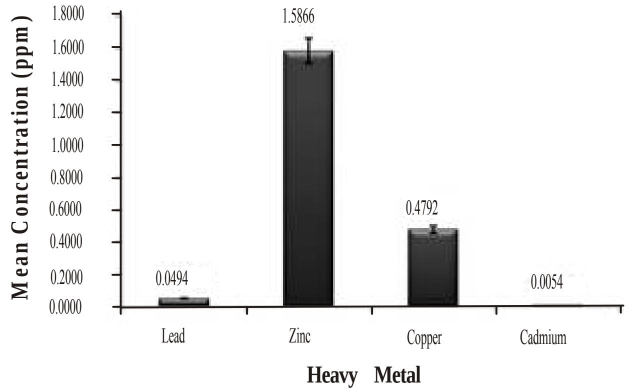

Figure 6: Mean concentrations of the heavy metals in the soft tissues of *A. implicata* sampled (n = 40) from Kaduna River.

Table 3. Correlation matrix of the heavy metal loads of *A. implicata* of different weights from Kaduna River.

	Pb	Zn	Cu	Cd
Pb	1.00			
Zn	-0.024	1.00		
Cu	0.378	-0.169	1.00	
Cd	0.566	-0.717	-0.003	1.00

Table 4. Correlation matrix of the weight categories of *A. implicata* from Kaduna River.

	W1	W2	W3	W4	W5
W1	1.00				
W2	0.73	1.00			
W3	0.98	0.85	1.00		
W4	0.72	1.00	0.84	1.00	
W5	0.73	1.00	0.85	1.00	1.00

Discussion

This study revealed that the whole soft tissues of *A. implicata* of different weights accumulate different concentrations of Pb, Zn, Cu and Cd. Since metals exist in water in different forms (colloidal – as free ions, particulate, and dissolved forms), the bivalves could have been predisposed to continuous metal intake from the surrounding water. The uptake from solution (Bryan and Uysal, 1978) through the gills, feeding on contaminated algae and coming in contact with the metal contaminated-sediments at the discharge point of the river could have exposed these *A. implicata* species to these heavy metals. Animal tissues accumulate heavy metals as a result of competing rates of chemical uptake and excretion (Otitoloju and Don-Pedro, 2006). The concentrations of the heavy metals in this study were lower than the values reported from Timsah Suez Lake and Canal by Khalid and Mohammed (2005) in two species of bivalves, *Paphia undulata* with concentrations (µg/L) 0.19 (Cd), 0.47 (Pb), 1.0 (Cu) and 8.71 (Zn) respectively. The second species, *Gafrarium pectinatum,* had corresponding concentrations of 0.22, 0.67l, 1.63 and 8.71 µg/L. Higher values of Cd, Pb and Cu were also reported in the tissues of *Etheria radiata* from creeks contaminated with heavy metals in Delta State Nigeria (Nwabueze, 2011).

The lower metal concentrations from this study could be attributed to continued dilution of the effluents at the point of discharge by the upstream flowing river. Heavy metals are reported to become toxic when they are not metabolized by the body but accumulate in the soft tissues (Ekpo *et al,* 2008). The heavy metals considered in this study (Pb, Zn, Cu and Cd) are reported among the elements commonly used in industry that are genetically toxic to animals (Scott and Smith, 1981). Though bivalves can store toxic materials at cellular levels of the body tissues to reduce the metal toxicity (Viarengo, 1985), elevated metal concentrations in their surrounding can result in abnormal developments in the adult and larvae (Mance, 1987), reduced heart, stoppage of filtration rate (Grace and Gainey, 1987; Kraak *et al*, 1994) and in greening and thinning of shells (Nielsen and Nathan, 1975). Shell-thinning can have a significant impact on the ecological distribution of bivalves, by rendering them more susceptibility to predation (Frazier, 1976). This could in turn have negative effects on the ecological balance of the aquatic ecosystem and may limit the diversity of aquatic organisms.

The differences in tissue weights of the bivalve did not have any significant influence on the concentrations of the different heavy metals. This is in contrast to report of Gopinathan and Sobhana-Amma (2006) who observed that the concentration of metals increases with the age and growth in edible soft tissues of the green mussel *Perna viridis*. This result implies that *A. implicata* of any weight and age could be used as bio-indicator of heavy metal pollution in a freshwater environment.

The high mean concentrations of Zn and Cu by the weight groups could be indication of the biological significance of these heavy metals as essential elements in the bivalve (Frontasyeva, 2011) or that they are easily assimilated by the soft tissue of the organism. Similarly, Amiard *et al* (1987) demonstrated that the marine mussel *Mytilus edulis* was capable of regulating the body concentration of the essential metals Zn and Cu. Zinc is an essential element to normal cell functions and stabilizes membranes (Viarengo, 1985) while copper enhances functioning of critical enzyme systems. The whole soft tissue load of the non-essential heavy metals, Pb and Cd, did not exceeded the average of 32.30 and 26.37 mg/Kg (Egwaikhide *et al,* 2013) reported in the sediment of River Kaduna**.** Pb and Cd are particularly toxic to animals even at low concentration. Therefore, human consumption of the soft tissues of the bivalve in whatever form could constitute a serious health risk. These two metals are of major interest in bio-availability studies (McKinney and Rogers, 1992). The persistence of these heavy metals could have negative effects on organisms in the higher trophic levels of the aquatic food chain. EPA (2000) reported on the tendencies of Cd and Pb to accumulate in the food chain and can cause damage to living organisms even at low concentrations.

The interactions of heavy metals as reflected in the correlation values could indicate that the heavy metals may influence uptake/accumulation through synergic action or antagonize each other in the soft tissues of the bivalve. The strong positive relationship of the different weight categories of *A. implicata* is an indication of a similarity in the pattern of the concentrations (deposition) of the heavy metals in the soft tissues of the bivalve regardless of their weights. This finding is contrary to the report of Boyden (1974) that age and body size of bivalve species are important factors that influence the accumulation of trace metals.

This study has shown that the metal contents of the effluents from Makera discharge point of Kaduna River among other factors have possibly predisposed the inhabiting *A. implicata* to accumulate different concentrations of lead, zinc, copper and cadmium of biological and public health importance. It also shows that *A. implicata* of any weight or age could be used to monitor the heavy metal pollution of a water body. Aside the biological importance of Zn and Cu to the bivalve, low concentrations of Pb and Cd could be damaging and may impair the bivalve from performing its ecological roles in the aquatic ecosystem. Similarly, *A. implicata* could serve as one of the routes by which these heavy metals could get to higher trophic levels of the food chain.

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