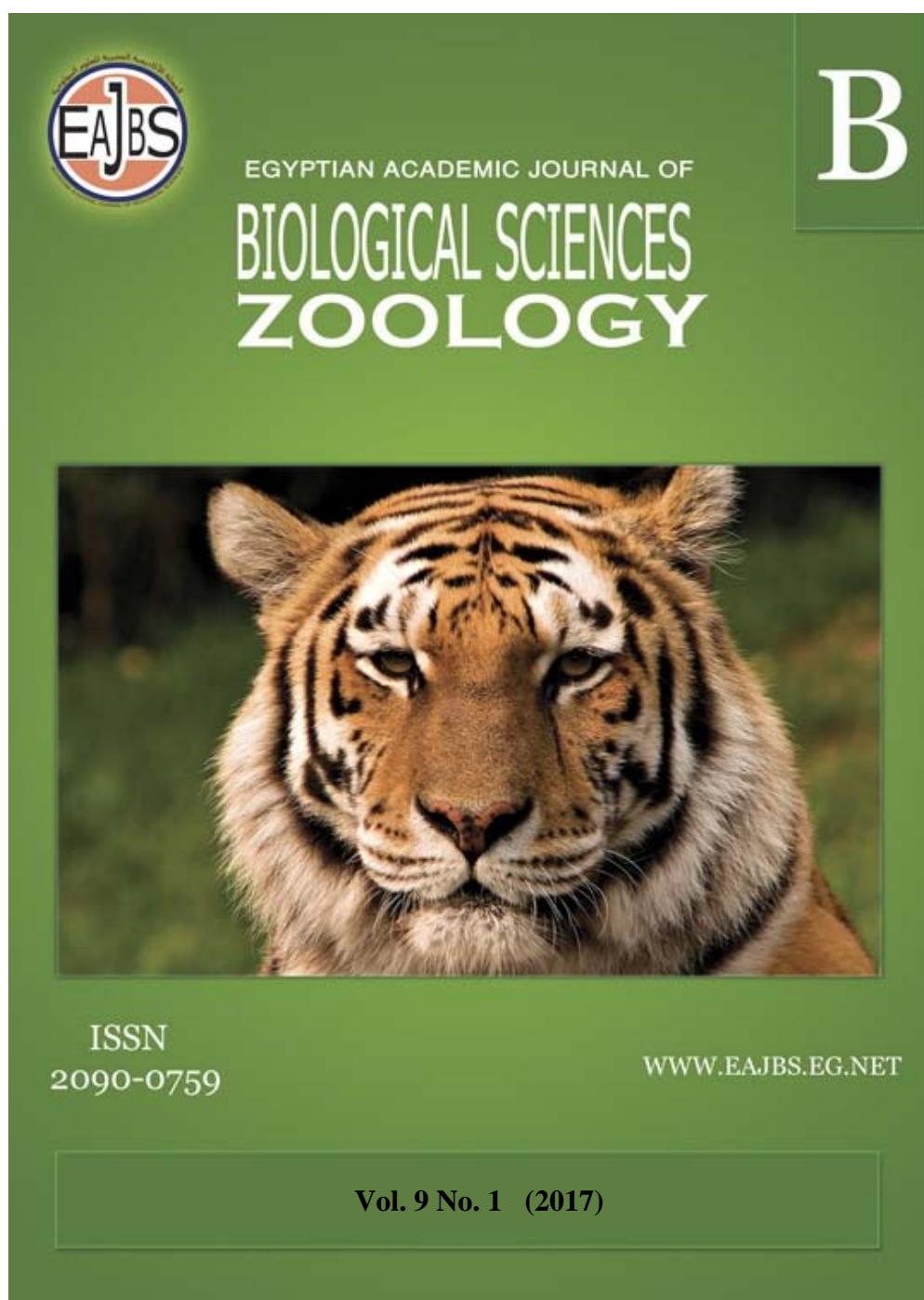


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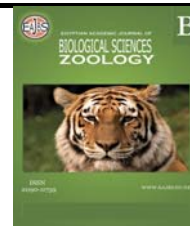


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Parasitic Helminth Fauna and Heavy Metals Analysis in *Macrobrachium macrobrachion* (Herklots, 1851) and *Macrobrachium vollenhovenii* (Herklots, 1857) From Lekki Lagoon, Lagos, Nigeria.

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ABSTRACT

This study was carried out to investigate the uptake and bioaccumulation of heavy metals by intestinal helminths parasites in relation to the muscular tissues of the freshwater prawn (*Macrobrachium species*) obtained from Lekki Lagoon, Epe, Lagos, Nigeria. A total of 120 specimens composed of 82 *Macrobrachium macrobrachion* and 38 *Macrobrachium* species was < 2.0. The trend of the mean metal concentration in the infected specimen ranged from Mn> Zn> Fe> Cu> Pb> Cr> Cd> Ni and Mn> Fe> Zn> Cr> Pb> Cu> Ni> Cd in the parasite. In the non-infected specimens the trend shows; Mn > Fe> Cu> Zn> Cd> Cr, Ni> and Pb. Manganese (Mn) was found to have high concentration compared to other metal analysed. Two of the metals (Pb and Mn) were found in significantly higher concentrations *vollenhovenii* were examined for helminths parasites. Four species of helminths parasites were recovered; *Procamallanus species*, *Cucullanus species*, *Anisakis species* and *Pomphorhynchus species*. Out of 82 specimens of *Macrobrachium macrobrachion* examined, 16 (19.51%) were infected. These infected specimens had 16(19.51%) males and 0(0.00%) female while the non-infected individuals had 49 (59.76%) male and 17(20.73%) females. The Chi-square distribution was significant at 0.01 level ($\chi^2(2) = 5.20$, $p < 0.01$). Sixteen specimens (16) (42.11%) were infected out of the 38 *Macrobrachium vollenhovenii* examined. The infected *Macrobrachium vollenhovenii* had 16(42.11%) males and 0(0.00%) female and the non-infected individuals had 18(47.37%) males and 4(10.53%) females. The Chi-square distribution was significant at 0.01 level ($\chi^2(2) = 5.20$, $p < 0.01$). The condition factor in the study for *Macrobrachium macrobrachion* in female was 1.4 ± 0.40 and in male 1.45 ± 0.49 , while the condition factor (K) of *M. vollenhovenii* in female was 1.18 ± 0.20 and in male 1.63 ± 0.61 . The mean value for the condition factor of both in acanthocephalans compared with the host tissues. While five of the metals (Fe, Zn, Cd, Cu and Cr) were found in significant high concentration in the infected tissue than the parasites. Analysis of all the heavy metal sample recorded for the water and sediment media of Lekki lagoon, indicate Zn to be above the FME permissible limit. However, findings from this study indicate that the acanthocephalans interfere with the uptake and bioaccumulation of heavy metal in relation to the tissue of the freshwater prawn, and can be applied in biomonitoring procedure of environmental pollution.

INTRODUCTION

The freshwater prawn, *Macrobrachium species* is regarded as the most important candidate species for inland aquaculture, and of high economic importance world-wide. In Nigeria, there are varieties of prawns and shrimps inhabiting the water bodies. The most common prawn species found in Nigerian rivers are the *Macrobrachium macrobrachion* and *Macrobrachium vollenhovenii* (Mantelatto and

Barbosa, 2005). They are important food items and also good items for exports when fully recruited for aquaculture.

Freshwater prawns have been found to harbour more endoparasites than ectoparasites. Several species of cestoda (*Ligula intestinalis*, *Monobothrium wagneri*, *Prochristianella species*, *Parachristianella species*, *Renibulbus species*), nematoda (*Paracamallanus species*, *Procamallanus species*, *Cucullanus species* and *Anisakis species*), and acanthocephalans (*Pomphorhynchus species*, *Acanthocephalus species*) have been found to inhabit *Macrobrachium species* (Monjit *et al.*, 2010).

The contamination of aquatic system by heavy metal has attracted the attention of several investigators both in the developed and developing countries of the world. Anthropogenic disturbance, particularly indiscriminate disposal of sewage, industrial waste and plethora of human activities have contributed to the contamination of aquatic ecosystems causing adverse effects on aquatic biota and human health (Wang, 2002). Contaminants pollution of aquatic systems by anthropogenic activities over the last decades has become a great concern. Ming – H0, 2005 in his report on heavy metals informed these contaminants enters the environment by natural and anthropogenic means such as earth's crust weathering, mining, soil erosion, industrial discharge, urban runoff, sewage effluents, pest or disease control agents applied to plants, and a number of other means. It is reported by Yahaya *et al.*, 2009 that both solid and liquid wastes contain heavy metals and are harmful to both flora and fauna in that habitat.

Furthermore, histopathological alterations can also be used as indicators for the effects of various anthropogenic pollutants on aquatic biota and are a reflection of the overall health of the entire population in the ecosystem (Mohamed, 2009; El-Bakary *et al.*, 2011). Degenerative changes in muscular tissues of aquatic animals have been reported as symptoms of exposure to environmental contaminants such as pesticides or metals (Wang *et al.*, 2004; Koca *et al.*, 2005). Histological changes in freshwater prawn muscular tissue might therefore be a reliable indicator of water pollution.

Shell-fishes have been widely used as bio-indicators to monitor heavy metal concentrations in the aquatic ecosystem. Prawns and shrimps are known to consume sand and mud along with detritus during feeding. This sediment act as sink for the heavy metals in aquatic ecosystems and since these metals are non-biodegradable and can persist in the environment for a very long period, they can be taken up by shell-fishes during feeding (Kumolu-Johnson *et al.*, 2010). Parasites of aquatic organisms can be used primarily as accumulation indicators in the aquatic ecosystems. That is, they are useful as bioindicators of environmental impact as reported by Victor *et al.*, 2009. Changes occur within the host when the dynamic equilibrium between the host and the parasite is lost. These lesions eventually resulted in some pathological changes as reported by Buchman and Lindstrom 2002; Knudsen *et al.*, 2009; Al-Jahdadi and Hassanine, 2010).

Macrobrachium species are of high economic importance world-wide and have been subjected to intense aquacultures especially in Asia and the Americas (FAO, 2006). *Macrobrachium species* occur commonly in the West African region and constitute an important part of the artisanal fishery.

The existing terms of 'shrimp' and 'prawn' without reference to any taxonomic groups are sometimes confusing as they are being used differently for different region. However, the usage of these two terms is inconsistent even within the same region.

Two species of these prawns *Macrobrachium macrobrachion* and *M. vollenhovenii* are important food items and are good items for exports when fully recruited for aquaculture (Bello-Lousier, 2004). *M. macrobrachion* and *M.*

vollenhovenii occur in the interconnecting Lagos and Lekki Lagoons where they support an artisanal fishery carried out mostly by women. The Lagos Lagoon with a surface area of nearly 208 km² is brackish with salinity range of 0.050/00 – 28.70/00 while the Lekki Lagoon with surface area of about 247 km² is mostly freshwater with salinity range of 0.0 – 3.10/00 (Lawal-Are, 2003).

Matsuo (2003) emphasized that heavy metal pollution are the cause of many diseases globally. They may have serious effects and cause ecological imbalance of the affected environment and to an enormous species of aquatic organisms. Some of the heavy metals are important to living organisms and are naturally found in the aquatic ecosystems. Their accumulation however, can reach a toxic level which can eventually demolish the ecological environment (Agusa *et al.*, 2005; 2007; Hajeb *et al.*, 2009; Kiyani *et al.*, 2013). The state and integrity of the environment should be monitored because the world is becoming increasingly concerned with controlling pollution. Walker *et al.*, (2001) reported the need for bio indicators organisms that reflect the environmental impact by responding to habitat alterations with changes in physiology, biochemical composition and behaviour. WHO (2010) reported that heavy metals have a serious impact on the environment and can threaten the ecosystem stability beyond tolerable limits. Otitolaju (2002); Agah *et al.*, (2009) however reported that at high concentrations, when heavy metals are deposited into soil, water and living organisms, in a contaminated environments, they actually resulted in detrimental health impacts. The bioaccumulation rate of heavy metals in a particular organism is a function of the chemical effect and its ability to bind to particular molecules and on the lipid content and composition of the biological tissue (Gbaruko and Friday, 2007).

The present research examined host-parasite bioaccumulation in prawn so as to ascertain the state of contaminant levels in the study locations.

MATERIALS AND METHODS

Study Area:

Lekki lagoon, located in Lagos State, South- Western Nigeria. Lekki Lagoon is one of the major lagoons in Lagos state, Nigeria (Kumolu-Johnson *et al.*, 2010). The other lagoons found in Lagos state includes: Ologe Lagoon and Lagos Lagoon. Lekki Lagoon is situated to the east, Lagos Lagoon in the central and Ologe Lagoon is situated to the west. Lekki Lagoon lies between latitudes 6°29'N and 6°38'N; and longitudes 3°30'E and 4°05'E. It has a surface area of about 247 km² with a maximum depth of 6.4 m; a greater part of the lagoon is shallow and less than 3.0 m deep. The Lekki Lagoon is part of an intricate system of waterways made up of lagoons and creeks that are found along the coast of South-Western Nigeria from the Dahomey border to the Niger Delta stretching over a distance of about 200 km (Agboola and Anetekhai, 2008). Lekki lagoon is very important because of the nursery and breeding ground it provides for a large variety of fish. Due to increase human activities and speedy industrialization there has been high discharge of industrial effluent containing heavy metal into the Lagoon. Map of Lekki lagoon is shown in Fig. 1.

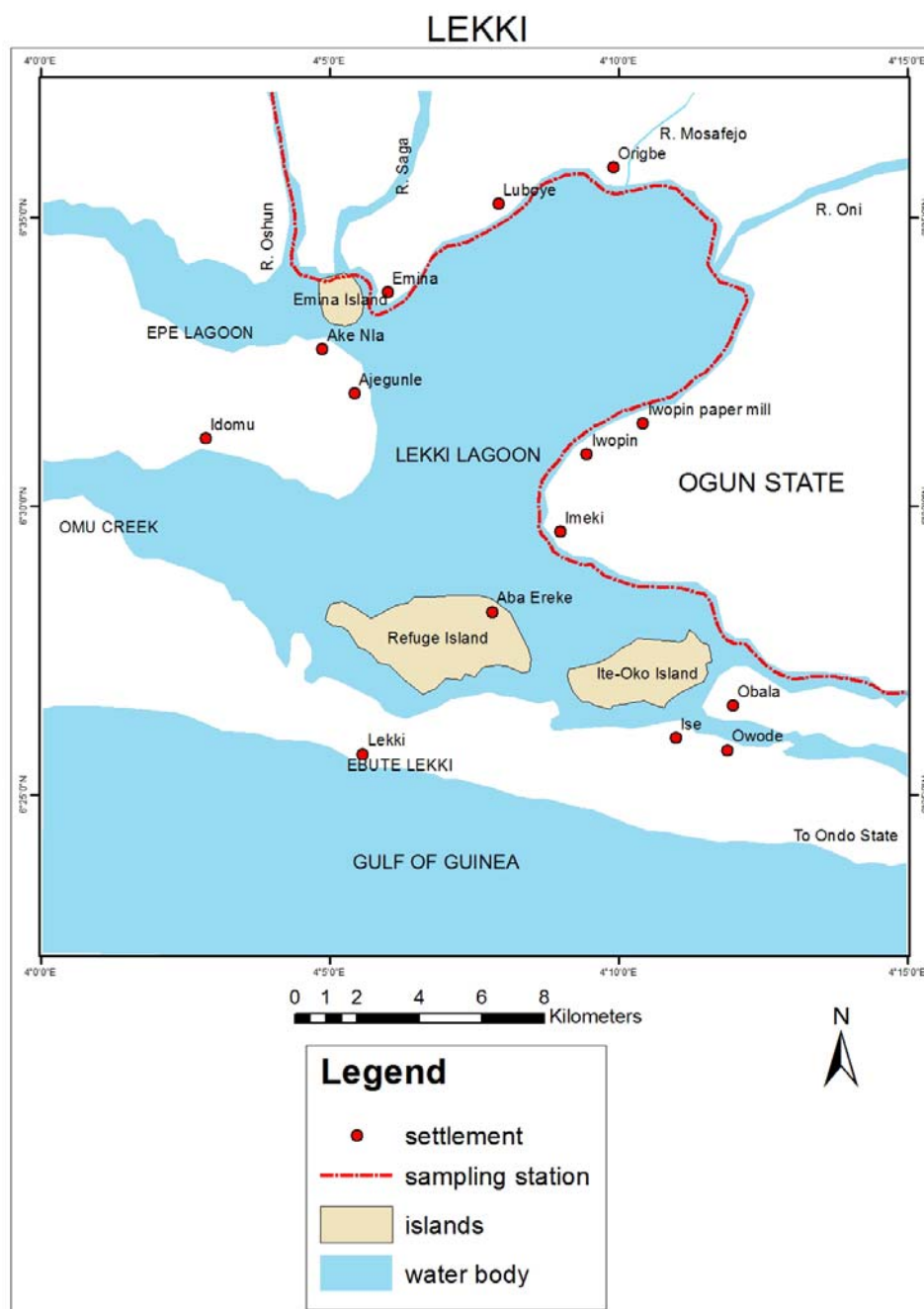


Fig. 1: Map of Lekki Lagoon and its environs

Specimens Collection

A total of 120 specimens of freshwater prawn (*Macrobrachium species*) freshly obtained from Lekki Lagoon were randomly purchased, twice weekly during the wet season at Oluwo fish market located along the Lekki Lagoon, Epe Lagos, Nigeria.

The specimens was immediately preserved in an ice-chest with ice-blocks and later transferred into the deep freezer (-20°C) within 24 hours to the laboratory prior analysis.

Length- Frequency Distribution:

The size distribution of the specimen was investigated using the length-frequency distribution method. The length- frequency distribution is based on the fact that the length of prawn of a particular size tends to form a normal distribution.

Laboratory Analysis:

Morphometric Parameters of Freshwater Prawn (*Macrobrachium species*). The body size of each specimen was determined length-wise (± 0.1 cm) by measuring from the tip of the telson to the tip of the dorsal teeth using meter ruler and measuring board, while the weights-wise (± 0.1 g) was determined using digital Camry weighing balance (model EK-1A series).

Prawn Identification: The specimens (*Macrobrachium species*) were identified using morphological characters reported by (Holthius, 1980) and (Powell 1982). Sex was determined using morphometric features, such as presence of an appendix masculina on the second pleopods and an appendix internal in the male while only the appendix internal was present in the female. There is also a lump on the ventral view of the first segment of the male abdominal region which could be felt by the tip of finger while it is absent in the female (Anetekhai, 1990; Abowei, 2008).

Parasite examination:

Each specimen was dissected and their guts removed and cut open with the aid of forceps. The stomach contents were removed by means of spatula into Petri-dishes containing physiological saline. The intestine was carefully split open longitudinally to aid the emergence of the gastrointestinal helminthes parasite. The infected guts was remove and the helminthes parasite obtain was fixed in 70% alcohol. The helminths parasite obtains was washed in water for proper identification and the parasites were analysed for heavy metal. The gastrointestinal helminthes parasite obtain, was sorted out into their various groups using standard parasitological guideline. Samples of the parasites were labelled in a sealed specimen bottle. Parasites identified were; nematodes (*Anisakis sp*, *Cucullanus sp*, *Procamallanus sp*) and acanthocephalan (*Pomphorhynchus sp*), using descriptions from; Chandler and Read (1961), Hoffman (1998) and FAO (2012).

Histopathological Examination:

Tissues sample (both infected and non-infected) were obtained from each prawn (*Macrobrachium species*) and fixed in separate bottles containing Bouins fluid for 6 hours This was thereafter decanted and 10 % buffered formalin was added to preserve the tissue. Random selection was made from the preserved tissues based on single or multiple infection and light, heavy or no infection.

The tissues were routinely dehydrated in an ascending series of alcohol (70%, 95% and then twice in absolute alcohol at 30 minutes duration), cleared in xylene and embedded in paraffin wax. Sections of 4-6 μ m thick were cut, processed and stained with hematoxylin and eosin (H&E). The stained tissues were washed off in tap water and the over stained ones destained in 1% acid alcohol. The tissues were mounted using DPX mountant dried and examined under the microscope.

Physico-chemical parameters measurement:

Water and sediment samples used for this study were collected from two (2) point along the Lekki lagoon using a 1litre water sampling containers. The containers were then preserved in a cool dry place and transported to the laboratory for further analysis of water quality parameters that could not be measured on site.

Parameters measured in-situ using a handheld multi-parameter probe, includes; Salinity, Hydrogen ion concentration (pH), Conductivity, Dissolved Oxygen while others were measured in the laboratory.

Bioconcentration Factor (BCF) :

In order to express the accumulation capacity of parasite the ratio were computed according to Sures *et al.* (1999):

$$\text{Bio concentration factors} = \frac{\text{Concentration of metal in parasite}}{\text{Concentration of metals in host tissue}}$$

Heavy metal analysis:**Tissue sample:**

Ten prepared samples, five infected and five non-infected were selected randomly for metal analysis (Pb, Fe, Zn, Cd, Mn, Ni, Cu and Cr). The head regions of each specimen was separated from the abdomen/tail and digested according to APHA (1985) and FAO/SIDA (1986). The tissue sample (10g) for metal analysis was obtained from the left dorso-ventral tissue of the Prawn. 100ml of thoroughly well mixed water sample was transferred into a beaker and 5ml concentrated nitric acid was added. The beaker was placed on a hotplate and evaporated to dryness.

The beaker cooled and another 5ml nitric acid was added. Heating was continued until a light-colourless residue was observed. Then 1ml concentrated nitric acid was added and the beaker was warmed slightly to dissolve the residue. The walls of the beaker were then washed with distilled water. The volume was adjusted to 50ml. Pb, Fe, Zn, Cd, Mn, Ni, Cu and Cr were then determined in the digested samples. Each digested prawn sample was analyzed for metal concentration using an Atomic Absorption Spectrophotometer (Analyst 200 Perkin Elmer model).

Sediment sample:

5 grams of dried sediment sample (soil) was digested with nitric acid (400ml). The sample is then heated until the brown fumes disappear. The sample is allowed to cooled, distilled water is added and make up to 50ml in a standard volumetric flask. The filtrate was filtered off and analysed for Pb, Fe, Zn, Cd, Mn, Ni, Cu and Cr using an Atomic Absorption Spectrophotometer (Analyst 200 Perkin Elmer model).

Statistical Analysis:

Data generated from the investigation was entered into Microsoft excel data sheet (2013) and then later subjected to two way analysis of variance (ANOVA). Chi-square was used to determine differences in parasite load between the sexes. Six (6) regression model were used to determine the relationship between length, weight and the total number of helminths parasites in the infested prawn using social science software (SPSS version 16 software

RESULTS

A total number of 120 freshwater prawns (*Macrobrachium species*) composed 82 *Macrobrachium macrobrachion* and 38 *Macrobrachium vollenhovenii* were examined for helminths parasites. Out of 82 specimen of *Macrobrachium macrobrachion* examined, 16 (19.51%) were found to infected and 38 specimen of *Macrobrachium vollenhovenii* were also found to be infected 16 (42.11%). The mean intensity was found to range between 3.44 and 4.25 (Table 1).

In the study, total number of 32(61.62%) freshwater prawn was found to be infected with intestinal helminths parasites. Helminths parasites encountered were in the phyla: Aschelminthes (nematodes) and *Acanthocephala species*. The rate of infection of each helminths recovered was *Paracamallanus species* 65(52.54%),

Cucullanus species 15(12.19%) *Anisakis species* 33(26.82 %) and *Pomphorhynchus species* 10 (10.00%) (Fig. 2).

Table 1: Parasite prevalence in both *Macrobrachium macrobrachion* and *Macrobrachium vollenhovenii*

Species	Number examined	Number infected	Prevalence (%)	Total number of parasite recovered	Mean Intensity
<i>Macrobrachium macrobrachion</i>	82	16	19.51	68	4.25
<i>Macrobrachium vollenhovenii</i>	38	16	42.11	55	3.44
Total	120	32	61.62	123	7.69

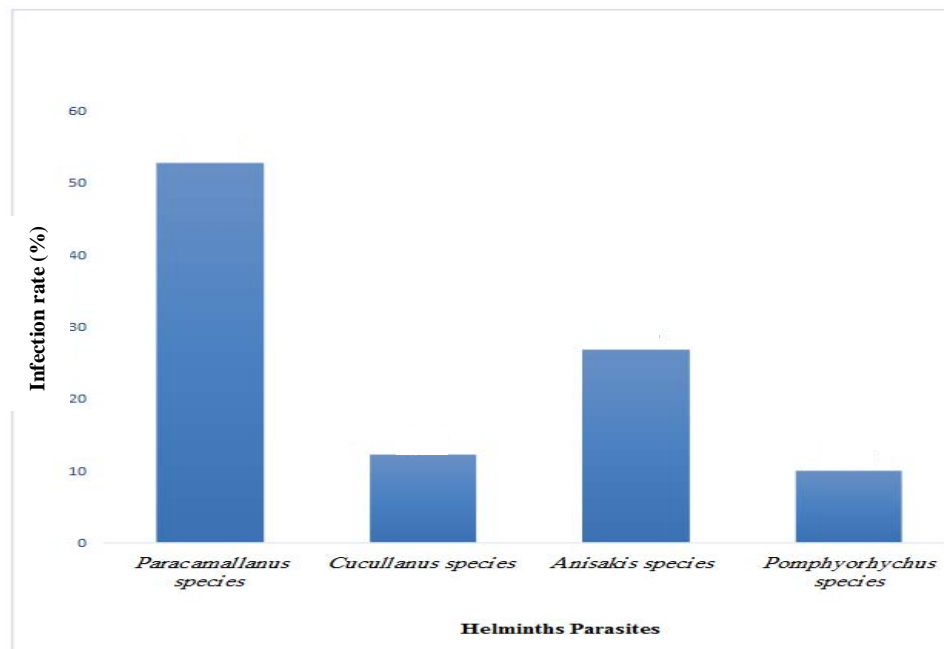


Fig. 2: Degree of helminths infection in relation to the genera of the helminth parasites.

Prevalence of intestinal helminths parasite in relation to sex of *Macrobrachium macrobrachion* in Lekki Lagoon

Out of the 82 *Macrobrachium macrobrachion* examined, 16(19.51%) were infected and 66(80.49%) were not infected. The infected individuals had 16(19.51%) males and 0(0.00%) female while the non- infected individuals had 49 (59.76%) male and 17(20.73%) females. The Chi-square distribution was significant at 0.01 level ($\chi^2(2) = 5.20$, $p < 0.01$). This is shown in Table 2.

Table 2: Prevalence of intestinal helminths parasite in relation to sex in *Macrobrachium macrobrachion* in Lekki lagoon.

SEX	NON-INFECTED	INFECTED	TOTAL NUMBER EXAMINED
Female	17 (20.73%)	0 (0.00%)	17 (20.73%)
Male	49(59.76%)	16(19.51%)	65(79.27%)
TOTAL	66(80.49%)	16(19.51%)	82(100%)

Chi-square Value χ^2 Asymp. Sig. (2-sided) = 5.20

P-Value significant at -0.01 level

Prevalence of intestinal helminths parasite in relation to sex of *Macrobrachium vollenhovenii* in Lekki Lagoon:

Out of the 38 *Macrobrachium vollenhovenii* examined, 16(42.11%) were infected and 22(57.89%) were not infected (table 3). The infected individuals had 16(19.51%) males and 0(0.00%) female while the non- infected individuals had 18(47.37%) males and 4(10.53%) females. The Chi-square distribution was significant at 0.01 level ($\chi^2(2) = 5.20$, $p < 0.01$).

Table 3: The prevalence of intestinal helminths parasite in relation to sex in *Macrobrachium vollenhovenii* in Lekki lagoon.

SEX	NON-INFECTED	INFECTED	TOTAL NUMBER EXAMINED
Female	4(10.53%)	0(0.00%)	4(10.53%)
Male	18(47.37%)	16(42.11%)	34(89.47%)
TOTAL	22(57.89%)	16(42.11%)	38(100%)

Chi-square Value χ^2 Asymp. Sig. (2-sided) = 3.25

P-Value significant at -0.01 level

Prawn tissues histopathology results:

The microscopic examination of the tissues affected by the helminths parasites (Fig.3) shows different pathological changes such as degeneration of the mucosal epithelium, presence of debris in the lumen and mild infiltration of fat into the lumen (Plates 1-5).

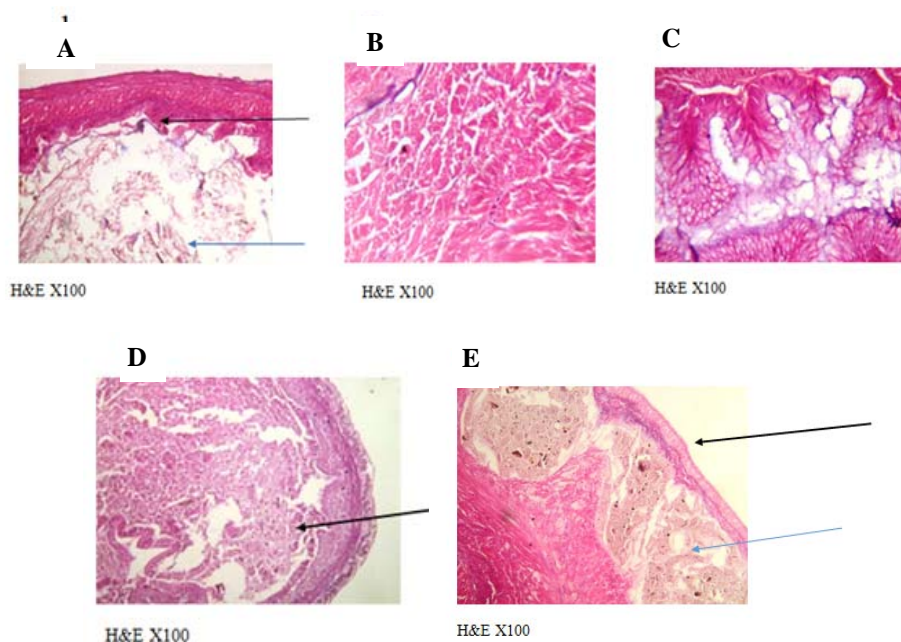


Fig. 3: Prawn tissues histopathology results

- Plate A: Photomicrographs of muscle tissue show villous structures (black arrow) completely obliterated and presence of debris in the lumen (blue arrow). (Infected muscle tissue).
- Plate B: Photomicrographs of muscle tissue show no significant lesion. (Non- Infected muscle tissue)
- Plate C: Photomicrographs of intestine show mild infiltration of fats into the lumen. No parasite seen. (Infected Intestinal tissue).
- Plate D: Photomicrographs of intestine show severe degeneration (black arrow) of the mucosal epithelium. The villi are unremarkable. No parasite seen. (Infected tissue)
- Plate E: Photomicrographs of intestine show the villous structures (black arrow) completely obliterated and presence of debris in the lumen (blue arrow). (Non-infected intestine)

Morphometric of *Macrobrachium macrobrachion* and *Macrobrachium vollenhovenii* in Lekki lagoon:

A total of 120 freshwater prawn were measured to obtain their length and weight parameters. The total length (TL) ranged from 8.00 to 15.00 cm and body weight (W) ranged from 5.00g to 88.00g. From (Fig. 4) the length-weight linear relationship ($y = 0.0888x + 9.0591$, $r = 0.7603$, $p < 0.001$) was obtained by plotting weight (g) and total length (cm) for the combined species, which shows a high relationship exist between length and weight and also the suitability of fitting the exponential formula $W = aL^b$ to the data.

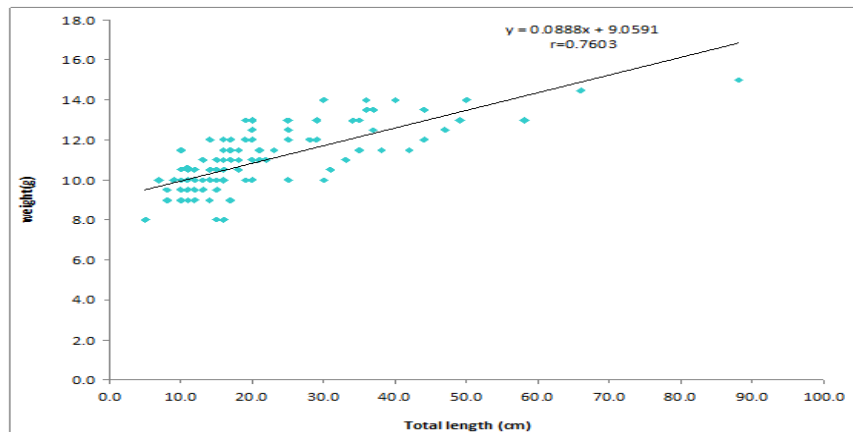


Fig. 4: Morphometric relationship between Length and weight of *Macrobrachium* species in Lekki lagoon.

The Mean morphometric and Condition factor analysis of *Macrobrachium macrobrachion* and *Macrobrachium vollenhovenii* in Lekki lagoon:

The mean value of the total length of *Macrobrachium macrobrachion* in female and male are $M \pm SD$, 10.33 ± 1.08 , $M \pm SD$, 10.88 ± 1.44 ($p < 0.01$) respectively, while the total length of *Macrobrachium vollenhovenii* in female and male are $M \pm SD$, 11.63 ± 1.11 , $M \pm SD$, 11.07 ± 1.80 ($p < 0.01$) respectively. Also, the mean value of the weight of *M. macrobrachion* in female and male are $M \pm SD$, 16.18 ± 4.94 , $M \pm SD$, 19.91 ± 11.52 ($p < 0.01$), meanwhile *M. vollenhovenii* in female and male are $M \pm SD$, 18.25 ± 2.06 , $M \pm SD$, 24.12 ± 17.50 ($p < 0.01$). The mean condition factor (K) of *Macrobrachium macrobrachion* in female was 1.4 ± 0.40 and in male 1.45 ± 0.49 also the condition factor (K) of *M. vollenhovenii* in female was 1.18 ± 0.20 and in male 1.63 ± 0.61 (Table 4).

Table 4: Mean morphometric and Condition factor analysis of *Macrobrachium macrobrachion* and *Macrobrachium vollenhovenii* in Lekki lagoon.

Species	Sex		Total Length (cm)	Weight (g)	Condition factor (K)
<i>Macrobrachium macrobrachion</i>	Female	Mean	10.33	16.18	1.47
		Std. Deviation	1.08	4.94	0.40
	Male	Mean	10.88	19.91	1.45
		Std. Deviation	1.44	11.52	0.49
<i>Macrobrachium vollenhovenii</i>	Female	Mean	11.63	18.25	1.18
		Std. Deviation	1.11	2.06	0.20
	Male	Mean	11.07	24.12	1.63
		Std. Deviation	1.80	17.50	0.61
Total	Female	Mean	10.58	16.57	1.42
		Std. Deviation	1.18	4.57	0.39
	Male	Mean	10.94	21.35	1.51
		Std. Deviation	1.56	13.92	0.54

P<0.01

Regression Model in Length-Weight relation of *M. vollenhovenii* in Lekki lagoon:

Table 5 shows six regression models used to show the length- weight relationship of *Macrobrachium vollenhovenii* between infected and Non-infected in Lekki lagoon. There were high correlations among the Infected compared to the Non-Infected. The correlation coefficients were significant at ($P < 0.001$) level between the Infected and Non-Infected freshwater prawn examined.

Table 5: Regression model in Length–Weight relationship of *M. vollenhovenii* in Lekki lagoon

Regression model	R-square		B-value	
	Infected	Non-Infected	Infected	Non-Infected
Linear	0.666**	0.215**	0.829**	0.503**
Logarithm	0.793**	0.303**	0.891**	0.550**
Growth	0.607**	0.208**	0.796**	0.496**
Exponential	0.634**	0.246**	0.796**	0.496**
Logistic	0.796**	0.496**	0.451**	0.609**
Quadratic	0.887**	0.600**	-1.189*	-1.612*

P – Value significant at *- 0.005 level, * *- 0.001 level

Regression Model in Length-Weight relation of *M. macrobrachion* in Lekki lagoon:

Table 6 shows six regression models used to show length-weight relationship between Infected and Non-Infected *M. macrobrachion* in Lekki lagoon. Low, Moderate and High correlation were recorded among the model used. Moderate to high correlation were recorded among the Infected and were all significant at $p < 0.001$. While Low to moderate correlation were recorded among the Non-Infected at $p < 0.001$ significant level.

Table 6: Regression model in Length – Weight relationship of *M. macrobrachion* in Lekki lagoon.

Regression model	R-square		B-value	
	Infected	Non-Infected	Infected	Non-Infected
Linear	0.519**	0.324**	0.720**	0.578**
Logarithm	0.655**	0.393**	0.810**	0.627**
Growth	0.504**	0.329**	0.733**	0.582**
Exponential	0.537**	0.339**	0.733**	0.582**
Logistic	0.733**	0.582**	0.481**	0.559**
Quadratic	0.847**	0.662**	-1.972*	-1.579*

P – Value significant at *- 0.005 level, * *- 0.001 level

Metal concentration in the infected and non-infected tissues of *Macrobrachium* species and the parasite:

The analysis of heavy metals in the water and tissue of the freshwater prawn showed that there was a significant difference (Anova, $P < 0.05$) which was an indication that the metal concentration were significantly impacting the aquatic ecosystem. The trend of the mean metal concentration in the infected specimen ranged from $Mn > Zn > Fe > Cu > Pb > Cr > Cd > Ni$ and were $Mn > Fe > Zn > Cr > Pb > Cu > Ni > Cd$ in the parasite, while the Non –infected specimen $Mn > Fe > Cu > Zn > Cd > Cr, Ni > Pb$. Manganese (Mn) was found to have high concentration compared to other metal analysed (Table 7).

Two of the elements (Pb and Mn) were found in significantly higher concentrations in acanthocephalans compared with to their host tissues. While five of the elements (Fe, Zn, Cd, Cu and Cr) were found in significant high concentration in the infected tissue than the parasites.

Table 7: Showing the mean concentration of Lead, Iron, Zinc, Cadmium, Manganese, Nickel, Copper and chromium concentrations in muscle tissues and parasites of freshwater prawn purchased from Oluwo fish market, Epe.

		Pb (mg/kg)	Fe (mg/kg)	Zn (mg/kg)	Cd (mg/kg)	Mn (mg/kg)	Ni (mg/kg)	Cu (mg/kg)	Cr (mg/kg)
Infected tissue (n=5)	Mean	1.1	2.23	3.35	0.32	7.68	0.12	1.81	0.76
	Std.dev	0.32	1.27	1.92	0.22	3.66	0.18	1.28	0.32
	Min.	0.62	1.1	0.93	0.05	2.63	0.02	0.06	0.42
	Max.	1.44	4.14	5.31	0.53	12.14	0.44	3.62	1.13
Non-infected tissue (n=5)	Mean	0.01	0.7	0.29	0.05	0.99	0.03	0.35	0.03
	Std.dev	0.01	0.57	0.32	0.04	1.46	0.03	0.51	0.02
	Min.	0	0.08	0.04	0	0.02	0	0	0.01
	Max.	0.03	1.22	0.73	0.1	3.41	0.07	1.23	0.05
Parasites (n=4)	Mean	0.6	1.7	1.61	0.01	5.62	0.09	0.5	0.37
	Std.dev	0.7	1.19	1.69	0.02	4.37	0.15	0.49	0.36
	Min	0	0.44	0.56	0	0.02	0	0.15	0.01
	Max.	1.32	3.02	4.13	0.03	10.19	0.32	1.2	0.81
Total (n=14)	Mean	0.66	1.73	2.35	0.02	5.11	0.16	0.68	0.41
	Std.dev	0.34	1.01	1.31	0.09	3.16	0.12	0.76	0.23
	Min.	0.21	0.54	0.51	0.02	0.89	0.01	0.07	0.15
	Max.	0.93	2.79	3.39	0.22	8.58	0.28	2.02	0.66

P-Value significant at -0.05 level

Metal Concentration in Water and Sediment Media of Lekki Lagoon:

Sample of water and sediment for metal concentration analysis were collected from two position and the mean concentration (mg/kg) was calculated in the Table 8. From the Table, the metal concentration in sediments was higher compared to metal concentration in the water sample. Zn was found above the FME permissible limit while Mn was not detected (ND) in the water sample. Mn was detected in the sediment and Fe was below the FME limit in both in water and sediment sample

Table 8: Mean Metal concentration in water and sediment media of Lekki lagoon.

		Pb (mg/kg)	Fe (mg/kg)	Zn (mg/kg)	Cd (mg/kg)	Mn (mg/kg)	Ni (mg/kg)	Cu (mg/kg)	Cr (mg/kg)
Water (n=2)	Mean	ND	0.11	1.53	ND	ND	0.03	0.77	ND
	Std. dev	ND	0.05	0.72	ND	ND	0.01	0.36	ND
	Min.	ND	0.07	1.02	ND	ND	0.02	0.51	ND
	Max.	ND	0.14	2.04	ND	ND	0.04	1.02	ND
Sediment (n=2)	Mean	ND	4.71	6.18	ND	0.14	ND	3.05	ND
	Std. dev	ND	2.22	2.91	ND	0.06	ND	1.44	ND
	Min.	ND	3.14	4.12	ND	0.09	ND	2.03	ND
	Max.	ND	6.28	8.24	ND	0.18	ND	4.06	ND
FME limit		Less than 1	20	Less than 1	Less than 1	NA	Less than 1	Less than 1	Less than 1

FME = Federal Ministry of Environment, FME source: FEPA, 1999

NA= Not available, ND= Not detected

Bio accumulation Factor of Freshwater Prawn and Parasites in Lekki Lagoon:

The Bioaccumulation factor (BAF) of the freshwater prawn tissue in relation to Sediment was calculated (**Unit =mg/kg**) according to Kalfakakour and Akrida-Demertzi (2000) and Rasheed(2001) , while the Bioconcentration of the parasite in relation to the infected host tissue was calculated according to Sures *et al.*(1999). The bioaccumulation factor were higher in the infected than the non-infected prawn

tissues. While the concentration of metals were higher in acanthocephalans than nematodes, with Mn (1.99) and Pb (2.4) more concentrated (Table 9).

Table 9: Bioaccumulation Factor (BAF) of freshwater prawn (in relation to sediment) and parasite in Lekki lagoon.

	Tissue		Parasites	
	Infected	Non-infected	Nematodes	Acanthocephalans
	Metal C. (tissue)/ Metal C. (sediment)		C (parasites) /C (Host tissue)	
Pb	0.00	0.00	0.33	2.4
Fe	0.5	0.15	0.95	0.35
Zn	0.00	0.05	0.58	0.33
Cd	0.00	0.00	0.06	ND
Mn	54.86	7.07	0.64	1.99
Ni	0.00	0.00	1.00	ND
Cu	0.59	0.11	0.33	0.21
Cr	0.00	0.00	0.58	0.36

N.D: Not determined as the concentration in the parasite was below or around the detection limit

DISCUSSION

One hundred and twenty specimens which are composed of 82 *Macrobrachium macrobrachion* and 38 *Macrobrachium vollenhovenii* were examined for this study. Out of the eighty two specimens of *M. macrobrachion* examined, 16(19.51%) were found to be infected while 16(42.11%) of *Macrobrachium vollenhovenii* were found to be infected. The infected individuals had 16(19.51%) males and 0(0.00%) female while the non- infected individuals had 49 (59.76%) male and 17(20.73%) females. The Chi-square distribution was significant at 0.01 level ($\chi^2(2) = 5.20$, $p < 0.01$), the prevalence of the helminths parasites recovered from the study was found to be higher *Macrobrachium vollenhovenii* (42.11%) compared to *Macrobrachium macrobrachion* (19.51%).

This study for the first time documented *Paracamallanus species*, *Cucullanus species*, *Anisakis species* and *Pomphorhynchus species* in freshwater prawn from Lekki Lagoon, Lagos, Nigeria. The rate of infection of each helminths recovered was *Paracamallanus species* 65(52.54%), *Cucullanus species* 15(12.19%) *Anisakis species* 33(26.82 %) and *Pomphorhynchus species* 10 (10.00%). Conversely, Akinsanya and Otubanjo (2006) recorded low prevalence of *Paracamallanus spp.* in Lekki lagoon in Lagos Nigeria, suggesting that this nematode is a fresh and brackish water parasite in Nigeria. There is therefore similar parasitic infections in both prawn and fish as reported by Akinsanya (2007) and (2008) in parasites of *Synodontis clarias* and *Malapterurus electricus* respectively.

The muscles are the most edible part of the freshwater prawn body, it is the most part exposed to damage by several types of pollution (Okayi *et al.*, 2013). Histopathological alteration observed in the tissues of freshwater prawn (*Macrobrachium species*) collected from Lekki Lagoon includes; degeneration of the mucosal epithelium, presence of debris in the lumen and mild infiltration of fat into the lumen. Although, no parasite was seen, contrary to the findings presented by Saad *et al.*(2012) associating fish epithelial lesion in polluted water with the invasion by microorganisms.

The length-weight relationship of the freshwater prawn is an important management tool. The average weight at a given length is used in assessing the relative wellbeing of a freshwater prawn population (Beyer, 1987). In this study, prawn specimens were randomly selected and their length and weight measured. The mean value of the total length of *Macrobrachium macrobrachion* in female and male were $M \pm SD$, 10.33 ± 1.08 , $M \pm SD$, 10.88 ± 1.44 ($p < 0.01$) respectively, while the total length of *Macrobrachium vollenhovenii* in female and male are $M \pm SD$, 11.63 ± 1.11 , $M \pm SD$, 11.07 ± 1.80 ($p < 0.01$) respectively. Marioghae (1990) reported similar variation pattern in the maximum attainable size for the two species in Lagos Lagoon. Willfuhr-Nast *et al.* (1993) cited *M.vollenhovenii* as the main targeted prawn species in the West African Tropical water bodies and attributed it to its much larger attainable size than others

Six regression models were used to show the length- weight relationship of *M. vollenhovenii* and *M macrobrachion* between Infected and Non-infected Prawns in Lekki lagoon. There were stronger correlation among the Infected compared to the Non-infected. The correlation coefficients were significant at 0.001 level between the Infected and Non-Infected of *M. vollenhovenii*. In *M macrobrachion* moderate to high correlation were recorded among the Infected and were all significant at $p < 0.001$. While Low to moderate correlation were recorded among the Non-Infected at $p < 0.001$ significant level. The regression exponent ($b < 3$) for both infected (0.733) and non-infected (0.582) of *Macrobrachium macrobrachion* and *Macrobrachium vollenhovenii* infected (0.789) and non-infected (0.496) indicate an isometric growth pattern of prawns in Lekki Lagoon, while the prawns exhibited negative allometric growth pattern. Similar findings by Lawal-Are and Owolabi (2012) confirms negative allometric growth pattern of freshwater prawns in Lekki Lagoon with values of regression coefficient (b) ranging between 2.4148 and 2.8666 for *M. macrobrachion* and between 2.2788 and 2.7117 for *M. vollenhovenii*.

The condition factor (K) indicates the state or overall wellbeing of the prawns as reported by Abohwerere (2008) which decreases with increase in length (Abowei, 2010). The condition factor (K) in this study were 1.47 ± 0.40 for female and 1.45 ± 0.49 for male in *M. macrobrachion* and 1.18 ± 0.20 for female and 1.63 ± 0.61 for male in *M vollenhovenii*. The mean value for the condition factor of both *M.macrobrachion* and *M.vollenhovenii* were < 2.0 . These results were comparable to the result documented for freshwater prawn by Abohweyere (2008) and Abowei (2008) in Lekki Lagoon. This implied that the condition (K) of Lekki lagoon, Nigeria in comparison with other fresh water bodies might be favorable for *M.macrobrachion* and *M.vollenhovenii*

The findings presented in this study, showed more heavy metal concentration in sediments sample than heavy metals present in the water sample, and these confirms previous observations by Don-Pedro *et al.*, (2004) in the Lagos Lagoon. Due to the static nature of sediments, they tend to accumulate more toxicant than water which may flow away, drain off or even evaporate. Fe, Zn and Cu were observed to be high in water and sediments samples when the two point were compared (**Table 8**). However, Zn was found above the recommended FME permissible limit for both the water and sediments sample while Cu was found to be above FME permissible limit for the sediments sample (Table 8). Furthermore, high concentration of metals in sediment may be attributed to human activities such as wastes from burning of fossil fuel, discharge of untreated sewage and industrial effluents containing metals into water bodies, as well as the natural ability of the sediment to act as sink (Kakulu and Osibanjo, 1988). This is in agreement with Odiete (1999), who concluded that

sediment is the major depository of metals, in some cases, holding more than 99% of the total amount of a metal present in the aquatic.

Recent published studies have reported that parasites as bioindicators of environmental impact and bioaccumulation of heavy metal in tissues of organisms (Sures *et al.*, 1999, Sures, 2001; 2003). It is also of note that parasites can act as metal sink for its fish hosts (Sures 2007). This is as a result that parasites physiological response are quick to polluted ecosystems (Sures, 2008). Knowledge of shell-fish parasites is of particular interest in relation not only to the health of the freshwater prawn but also to understand ecological problems. In this study, the bioaccumulation factor were higher in the infected than the non-infected freshwater prawn tissues. While the bio concentration of metals were higher in acanthocephalan (*Pomphorhynchus species*) than nematodes (*Paracamallanus species*, *Cucullanus species* and *Anisakis species*). Pb (2.4) were high when compared with the infected prawn tissue Pb (0.00), meanwhile Mn (54.86) was recorded to be very for the infected prawn tissue (Table 9) but not detected in water sample and very low in sediment (0.14) (Table 8). Also, Pb and Cr were not detected in the sediment sample analysed (Table 8) but were found to be concentrated in the acanthocephalan (*Pomphorhynchus species*). This result confirms the fact that acanthocephalans can be used in environmental impact studies, to detection very low concentrations of metals in the environment owing to the enormous accumulation capacity of these worm (Sures and Riemann, 2003).

The distribution of metal in the infected specimen ranged from Mn > Zn > Fe > Cu > Pb > Cr > Cd > Ni and were Mn > Fe > Zn > Cr > Pb > Cu > Ni > Cd in the parasite, while the Non-infected specimen Mn > Fe > Cu > Zn > Cd > Cr, Ni > Pb. While Pb, Cd and Cr were not detected in the water and sediment sample analysed, while Mn was only detected in sediment sample at a very low concentration. Meanwhile Fe, Zn and Cu were detected (Table 8). From this study Zn was found to be the most abundant metal in the Lekki Lagoon (water = 1.53 ± 0.72 ; sediment = 6.18 ± 2.91). This was also in confirmation with previous observations by Don-Pedro *et al.*, (2004) in Lagos Lagoon.

The outcome of this study indicate that the of infection have affect the uptake and accumulation of heavy metal in the prawn tissues, this confirms previous reports that have been published on the uptake and accumulation of heavy metals by helminths parasites (Sures, 2001; Sures, 2003)

CONCLUSION

This study showed that *Macrobrachium species* from Lekki Lagoon in Lagos, Nigeria contains metals in detectable quantities. It was found that helminths parasites affects the uptake and bioaccumulation of heavy metals in tissues of freshwater prawn. Although the use of shellfish such as freshwater prawn to assess the impact of environmental pollution lacks considerable amounts of information on the effects of the parasites on common bioindication procedures such as analysis of biomarkers and the use of parasites as indicator. Hence, the use acanthocephalan (*Pomphorhynchus species*), and nematodes (*Paracamallanus species*, *Cucullanus species*, *Anisakis species*) can be applied as bioindicator for monitoring heavy metals pollution in the aquatic ecosystem.

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