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Host condition, parasite interaction and metal accumulation in *Tilapia guineensis* from Iddo area of Lagos lagoon, Nigeria

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Abstract: This paper investigates host condition factor, parasite interaction and metal accumulation in *Tilapia guineesis* and Host-Parasite system in Iddo Area of Lagos Lagoon, Nigeria. Eighty specimens of *T. guineensis* from the Lagoon were caught and dissected for intestinal helminth parasites. Condition factors of all individuals were determined. Median condition (K<2.62 and K>2.62) were used in grouping the individuals. Sediment, water samples, livers and intestinal parasites were analyzed for heavy metals using atomic absorption spectrophotometer. Metal concentrations in the water medium were below detectable limits, but those in sediments were; cadmium (Cd) (mg/l), 11.29±22.59, Manganese (Mn)(mg/l), 0.02±0.02, Iron (Fe) (mg/l), 141.09±15.09, P<0.01, lead (Pb))(mg/l), 123.16±8.41, P<0.01. Livers of infested individuals accumulated more metals than those not infected by the parasites. This was also discovered when they were grouped based on their condition status. Among the infested fish, high condition individuals (>2.62) accumulated more metals than the low condition infested individuals (<2.62), but irregular trend was found among the non-infested individuals. Parasite prevalence among infested individual of low condition factor were; *Heterostomum* sp., a trematode; 0.25, *Procamallanus* sp., a nematode; 0.75, *Acanthogyrus tilapae*, an acanthocephalan, 0.05 and *Proteocephalanus* sp., a cestode was absent. High condition individuals had parasite load; *Heterostomum* sp.; 0.45, *Procamallanus* sp., 0.05, *Proteocephalanus* sp., 0.05 and *Acanthogyrus* sp.; 0.25. Generally, the host liver accumulated more metals than their parasites except iron among parasites of high condition individuals. Relative abundance and diversity in intestinal parasites with varying metal bioaccumulation potentials in high and low condition individuals could have been responsible for the difference in metal accumulation in their host tissue. Therefore, it is recommended that environmental monitoring studies involving the use of fish could be improved upon by including the macro-parasites, considering the significant effect they could have in the fish biology, behaviour and ecology.

Keywords: Parasite interaction, Metal accumulation, Condition factors, Atomic absorption spectrophotometer.

Introduction

Parasites are ubiquitous in nature (Marcogliese 2005) and it has been suggested they are useful indicator species of contaminant-impacted ecosystems (Mackenzie et al. 1995) and impaired food webs (Marcogliese et al. 2006). Numerous studies have shown the sensitivity of parasites to contaminants such as metals (Pietrock & Marcogliese 2003). Over the past few decades, heavy metal contamination of aquatic system has attracted the attention of several investigators both in the developed and developing countries of the world. Many industrial and agricultural processes have contributed to the contamination of aquatic ecosystems causing adverse effects on aquatic biota and human health (Wang 2002). The fact that heavy metals cannot be destroyed through biological degradation and their ability to accumulate in the environment, make these toxicants deleterious to the aquatic environment and consequently to humans who depend on aquatic products as sources of food. Heavy metals can accumulate in the tissues of aquatic animals and as such tissue concentrations of heavy metals can be of public health concern to both animals and humans (Kalay et al. 1999; Ashraf 2005). In the Lagos Lagoon, metals are reported to be well concentrated in the water, sediments and biota (Don-pedro et al. 2004).

Certain parasites may be vulnerable partly due to their ability to accumulate metals (Sures et al. 1997). Cestodes, for example, have been shown to accumulate metals to levels that can exceed host tissue concentrations by several orders of magnitude (Sures et al. 1997). Parasites that are in direct contact with the environment such as monogeneans and the free-living stages of some intestinal parasites may be especially vulnerable to heavy metals (Pietrock & Marcogliese 2003). Endoparasitic infections normally give an indication of the quality of the water since this infections increase in more polluted waters (Avenant-Oldewage 2001). Bush et al. (2001) stated that parasites usually exist in equilibrium with their hosts as a survival strategy.

The well-being of the host population in general can be determined by analyses of condition factor. Condition factor is a measure of energetics, nutritional status and viability of a host. Fishes are hosts to taxonomically diverse parasites, and infections can significantly affect fish behaviour, metabolism, body condition, fecundity or survival (Barber et al. 2000; Dobson et al. 2008; Lafferty 2008; Seppanen et al. 2009). Condition factor varies directly with nutritional status, and disease has been shown to be negatively correlated with condition (Adams 1999). White suckers have been found to have enhanced condition factors in response to pulp mill effluents. Decreased condition factors have been observed in white suckers in response to elevated concentrations of metals. Nutrition, disease and contaminants are highly interrelated in terms of their effects on fish condition. Avenant-Oldewage (2001), observed that the body condition changes rapidly in response to environmental perturbation and is more indicative of current environmental constraints on growth than measures of past growth. Condition factor also appears to vary seasonally, possibly as a response to changing food resources, metabolism, or gonadal status. Condition indices can also vary from location to location within the species (Goede & Barton 1990). The objective of this investigation was to find out the combined effects of heterogeneous parasite load and host condition factor on metal accumulation in host-parasite system in Iddo area of Lagos lagoon, Lagos, Nigeria.

Materials and Methods

Study Area: The Lagos Lagoon is the largest of the four lagoon systems in the Gulf of Guinea. It stretches to about 256km from Cotonou in the Republic of Benin to the Western edge of Niger Delta. The lagoon borders the forest belt and receives a number of important rivers (Yewa, Ogun, Onu and Oshun) draining more than 103,626km of the country (Ajao 1996; Don-Pedro et al. 2004). The Lagos Lagoon (6°22'-6°38'N/2°48'-4°36'E) levels of these heavy metals could be attributed to lying behind the barrier beach and extending for 210km along the coast. Lagos Lagoon is connected by a long narrow navigable channel with Cotonou Lagoon, Benin Republic. The lagoon is more than 50-km long and 3 to 13km wide, separated from the Atlantic Ocean by a long sand spit 2 to 5-km wide, which has swampy margins on the lagoon side. The Lagos Lagoon empties into the Atlantic via Lagos Harbour, a main channel through the heart of the city, 0.5 to 1km wide and 10km long (Fig. 1).

Fish sample collection: Eighty (80) Samples of *Tilapia guineensis*, ranging between 120 and 210mm standard length, were collected from the Lagos Lagoon, within intervals of one week in July, 2013, to ensure that samples were obtained as close to same period of the year as possible. The size range of fish

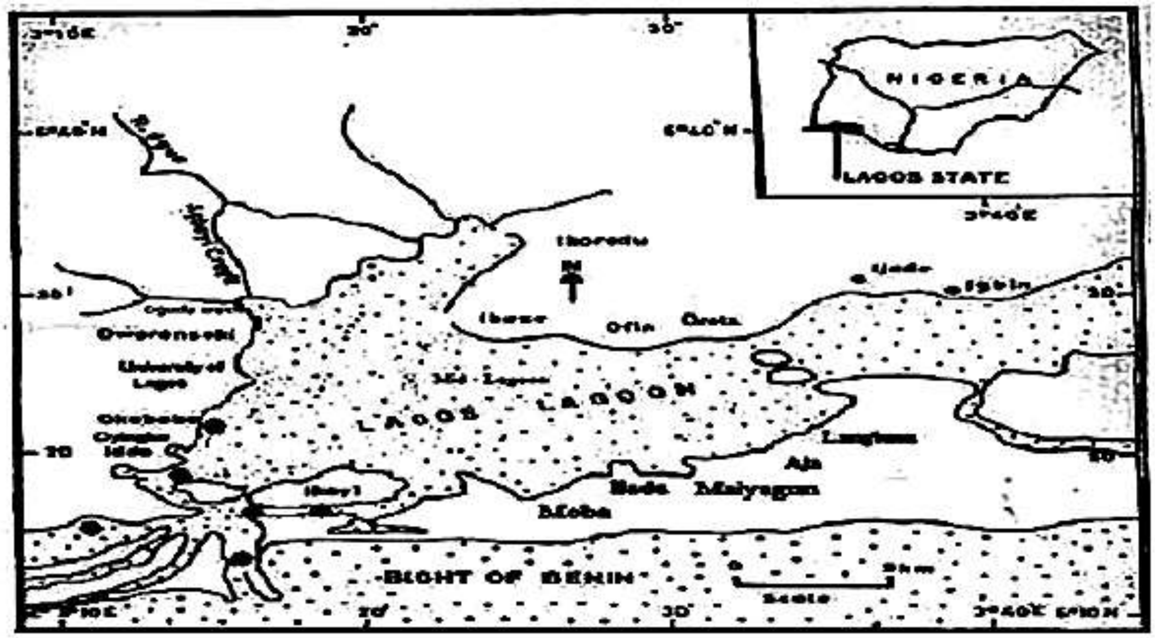

Fig.1. Map of Lagos Lagoon showing the sampled sites; Iddo area of the Lagos lagoon, Lagos, Nigeria.

used were likely to be of approximately the same age. Fishing was done by means of cast nets of 45 and 50mm mesh sizes. The specimens were transported in ice chest to the laboratory, where measurements started immediately to avoid shrinkage.

Laboratory analysis for the parasites and metals: The eighty (80) individuals of *T. guineensis* dissected for intestinal parasites. Sexes were identified. The sex ratio, parasite prevalence, load, intensity, and Fulton Condition Factor (K) were estimated using the formulae below;

Sex Ratio = No of Male individuals / No of Female individuals

Prevalence $= \frac{\text{Number of infested fish}}{\text{Total number of fish examined}}$

Bio-load $= \frac{\text{Number of collected parasites}}{\text{Number of infected fish}}$

Abundance $= \frac{\text{Number of collected parasites}}{\text{Number of fish examined}}$

Condition Factor (K): The condition factor also known as the Ponderal index or the Fulton Coefficient of condition computed using the formula described by Worthington and Ricardo (1936):

$K = \frac{100W}{L^b}$ (Le Cren, 1951)

b =Value obtained from the growth exponent in the length (b = 3)

W=Total weight of fish (g)

L=Standard length (cm)

K=Condition factor

Condition factor (K) was used as a base for grouping individuals into low condition status and high condition status (Carlander 1969, 1977; Friedmann et al. 2002; Amber et al. 2007; Goede & Barton 1990). Median Condition factor was calculated using Ranking cases; order of magnitude of condition factors of eighty (80) individuals of *T. guineensis* in the sample 50th percentile, median condition (K<2.62 and K>2.62). Individuals with Condition factor less than 2.62 were grouped into low condition status while individuals with Condition factor greater than 2.62 were grouped into high condition status.

Examination and Identification of Parasites: The livers and a fraction of the teased intestine were collected in EDTA bottles. The recovered helminth parasites were preserved in 70% alcohol. The

Table 1. Mean of heavy metals in water and sediment media, organs of *Tilapia guineensis* and its intestinal parasites in Iddo Area of the Lagos lagoon, Lagos, Nigeria.

Medium/Host	Number	Cadmium (mg/l)	Manganese (mg/l)	Iron (mg/l)	Lead (mg/l)
Water	4	0.00±0.00	0.00±0.00	0.01±0.02	0.00±0.00
Sediment	4	11.29±22.59	0.02±0.02	141.09±15.09**	123.16±8.41**
Infested liver	20	1203.62±1888.69**	638.38±754.83**	53.72±81.79**	1551.63±2021.93**
Non-infested liver	20	495.83±562.49**	214.51±210.79**	34.87±23.32**	1078.33±652.89**
Parasite	20	752.21±1804.34	393.11±496.09**	248.14±1049.49	1115.29±2131.21**

*Mean values significant at P<0.01 level, **Mean values significant at P<0.05 level.

recovered helminth parasites were sorted out into various groups using standard parasitological guidelines. Samples of the parasites were transferred to vials thoroughly sealed and appropriately–labelled. Samples were later given detailed identification in Zoology Laboratory, University of Lagos. Parasites identified were *Proteocephalanus* sp., a cestode, *Heterostomum* sp., a trematode, *Acanthogyrus* sp., an acanthocephalan and. *Procamallanus* sp., a nematode.

Heavy Metal Analysis: Individuals were also grouped based on infestation status. The livers of twenty each of infested and non-infested individuals, making a total of forty (40) individuals, including the identified parasites from each of the infected host, were selected for heavy metal analysis.

Digestion of Samples: Water samples collected from the experimental stations were filtered and digested using standard digestion procedure (APHA/AWWA/ WPCF, 1995). Sediment samples were dried, sieved through a 200µm sieve to normalize for particular size, and digested using the method provided by Agemian & Chau (1976). The livers and the parasites were homogenized and digested using the method described by Jensen et al. (1983).

Atomic Absorption Spectrophotometer (AAS): The heavy metal concentration in each digested sample was determined by comparing their absorbance with those of standards (Solutions of known metal concentration) using AAS, Atomic Absorption Spectrophotometer, AES, 2000 series. An acetylene air mixture was used as the flame. The working standard for each of the metals was aspired into the flame in the order of 0.00 0.8 and 1.6ppm. The values were used to plot a standard curve. The tissues were then aspirated into the flame and the values were obtained by extrapolation from the standard curve. For data quality, particular attention was paid to the cleanliness of all wares. In addition, factory prepared AAS standard solutions were run as sample for accuracy check after every five measurements.

Statistical Analysis: Multi-Variance Analysis and Analysis of Variance (ANOVA) was used to compare means of metal concentrations in media (fish parasite, fish tissue, water and sediment) of the lagoon ecosystem, using Microsoft Excel 2010 and SPSS 20 software.

Result

Heavy metals in water sediment, fish organs and intestinal parasites: Metal concentrations in the water medium were below detectable limits, but those-in the sediments, liver of infested individuals, non-infested individual and the parasites were significant (Table 1). The trend of metal accumulation for cadmium concentrations was: water < sediment < non-infested (liver) < parasite < infested (liver), for manganese concentration was: water < sediment < non-infested (liver) < infested (liver) <parasite, for Iron concentration was: water < non-infested (liver) < infested (liver) < sediment<parasite-and for Lead concentration was: water < sediment < non-infested (liver) < parasite < infested (liver).

Mean concentration of heavy metals in fish organs and parasites based on host Condition Factor (K): Heterogeneous infested individuals of low condition

Table 2. Parasite diversity and load based on Host Condition Factor. Median Condition Factor (mK)=2.62.

Condition Factor	Infestation Status	*Heterostomum* sp.	*Proteocephalanus* sp.	*Procamallanus sp.*	*Acanthogyrus* sp.
Low Condition Factor K<2.62	Infested Individuals	0.25	Nil	0.05	0.05
	Non-infested Individuals	Nil	Nil	Nil	Nil
High Condition Factor K>2.62	Infested Individuals	0.45	0.05	0.75	0.25
	Non-infested Individuals	Nil	Nil	Nil	Nil

Table 3. Mean concentration of Cadmium and Magnesium in organs of *Tilapia guineesis* and parasite based on Host Condition Factor (K). Median Condition Factor (mK)=2.62.

Condition Factor (K)	Infestation Status	Cadmium (mg/l)		Magnesium (mg/l)	
		Liver	Parasite	Liver	Parasite
Low Condition Factor K<2.62	Infested Individuals	1043.05±1294.04	786.64±1473.15	319.61±518.29	360.72±450.42
	Non-infested Individuals	524.19±599.37**	Nil	211.14±220.51**	Nil
High Condition Factor K>2.62	Infested Individuals	1272.43±21.28**	737.45±1980.60	774.99±813.89	406.99±530.06*
	Non-infested Individuals	429.68±510.67	Nil	222.38±205.68*	Nil

*Means values significant at P<0.01 level, **means values significant at P<0.05 level.

status (K<2.62) harbored *Heterostomum* sp., a trematode, *Procamallanus* sp., a nematode, and *Acanthogyrus* sp., an acanthocephalan with prevalence of 0.25, 0.05 and 0.05, respectively (Table 2). Cadmium and magnesium concentrations in the host organs of low condition status are shown in Table 3. These concentrations were lower among non-infested, low condition individuals. Host individuals of high condition status, (K>2.62) harbored *Heterostomum* sp., *Proteocephalanus* sp., a cestode, *Procamallanus* sp. and *Acanthogyrus* sp. having prevalence of 0.45, 0.05, 0.75 and 0.25, respectively. These infested individuals had tissue cadmium and magnesium concentrations as indicated in Table 3. Among these individuals, the liver accumulated more cadmium and magnesium than the parasites.

Mean concentration of Iron and Lead in organs of *T. guineensis* and parasites based on Host Condition Factor are shown in Table 3. Infested accumulated higher iron and lead concentrations than non-infected fish. Among these individuals the parasites accumulated more iron (Table 4).

Discussion

Heavy metals such as cadmium and manganese are contained in industrial waste, metal plating and generated through microbial actions (Nwankwo 2004). The properties of metals ionized in water depend largely upon the nature and behavior of the metal species (Bhatia 2010). Speciation and complexation of these metal species depend on the physiochemical properties of the aquatic environment which are influenced by seasonal variation and other factors. These processes determine their bioavailability, transport, accumulation and biological effect (Bhatia 2010). Complexation may cause changes in oxidation states

Table 4. Mean Concentration of Iron and Lead in organs of *Tilapia guineensis* and Parasite based on Host Condition Factor (K). Median Condition Factor (mK)=2.62.

		Iron (mg/l)		Lead (mg/l)	
Condition Factor (K)	Infestation Status	Liver	Parasite	Liver	Parasite
Low Condition Factor K<2.62	Infested Individuals	34.01± 15.33*	14.06± 6.72*	1126.63±933.25	798.28±589.56
	Non-infested Individuals	30.12± 25.60**	Nil	1106.18±724.25**	Nil
High Condition Factor K>2.62	Infested Individuals	65.54± 95.73	348.18±1254.53	2060.91±2214.05	1485.41±2468.14
	Non-infested Individuals	43.70± 16.46**	Nil	970.91±529.20**	Nil

*Means values significant at P<0.01 level, **Means values significant at P<0.05 level.

of the metals and result in metals becoming ionized, its interaction with organic species is a function of hydrogen ion concentration (Bhatia 2010). The formation of insoluble complex compound removes metal ions from the water medium.

In the lagoon, the metal concentration in the sediment was more than that in the water medium. The various metal concentrations in the water medium and sediment were very low, but greater concentrations were found in the fish. Some metals are available for uptake into organisms from solution only as free ions, whereas others are transported over biological membranes as inorganic complexes (Bienvenue et al. 1984). Variation in the behavior of metallic species, properties and complexation is responsible for the variation in the bioaccumulation gradient in the fish and the parasites. Experimental studies have shown that there are variations in the trend of accumulation of metals in tissues and is dependent on species, sex, age, diseases, nutritional and genetic factors (Farombi et al. 2007).

Studies from the field and laboratory experiment also showed that accumulation of heavy metals in the tissues is mainly dependent upon water concentration of metals and exposure period, although some other environmental factors such as salinity, pH. Hardness and temperature play significant roles in metal accumulation (Authman, 2008). Ecological needs, sex, age, size, feeding habitat as well as biological conditions of the fish affect the heavy metal accumulation in their tissues. The presence of a given metal at high concentration in water and sediments does not involve direct toxicological risk to the fish, especially in the absence of significant bioaccumulation.

It is important to examine what happens when stressors to a population of fish overlap with parasitism. There are several possible qualitative outcome when parasite interact with other stressors (heavy metal pollution, or/and water temperature). The most obvious concept is that some stressors may make hosts more susceptible to parasitism. This appears to be due to an increase in susceptibility because toxic conditions compromise a fish's immune system (Paperna 1996). However, one obvious prediction is that pollutants, as heavy metals may reduce immunological capabilities of host, rendering them more susceptible to parasites. Bioaccumulation is a measure of exposure response to stress in fishes (Paperna 1996). Infested individuals of *T. guineesis* accumulated more metals in the organs compared to non-infested individuals. Certain parasites could help the fish accumulate metals in its tissues but this is not true for all metals. Infested individuals of *T. guineesis* that harbor *Acanthogyrus* sp. had reduce metal accumulation due to the high bioaccumulation potential of the parasite. Some of these individuals harbor both parasites under heterogeneous infestation, this had resulted to regularity in host tissue accumulation when compared with the non-infested individuals. Parasites with low bioaccumulation potential such as *Heterostomum* sp. and *Procamallanus* sp. could increases metal accumulation in the host while parasites with high bioaccumulation potential such as *Acanthogyrus* sp. could decreases metal accumulation except under heterogeneous infestation. This is dependent on prevalence of the individual parasites, load and intensity of infestation. These also vary between sexes within the population. There are reports on higher parasite infectivity in females as compared to males. Ibiwoye et al. (2004) recorded that female fishes were more prone to parasite infestation than males. Singhal & Gupta (2009) reported comparatively higher trematode infestation in female fish hosts (10.75%) than the male hosts (9.40%). Findings of *T. guineesis* of present investigation do not disagree with the views of above authors. The rate of infestation is dependent on condition status of the individuals, their sexes and other physiological variations among conspecific and congeneric individuals. Condition factor varies directly with nutritional status, and disease has been shown to be negatively correlated with condition (Adams et al. 1992). Decreased condition factors have been observed in white suckers in response to elevated concentrations of metals (Carlander 1969). Nutrition, disease and contaminants are high interrelated in terms of their effects on fish condition (Carlander 1969). Once contaminants directly or indirectly affect host condition, it could cause changes in parasite population dynamics either through reduction in relative abundance of intermediate host or free-living stage of the parasites. No significant differences in infection rate of male and female hosts were observed by Jarkovsky et al. (2004). Factors responsible for causing variations in parasite infectivity due to seasonal variations and host sex are debatable. Changes in the fish feeding behavior and annual temperature regime have been considered as the principal factors responsible for the seasonal incidence and intensity pattern of parasites. Higher prevalence of parasites in female hosts may be due to the fact that they are equipped with a positive stimulus which may preferentially be attracting the cercariae and other helminth parasites. Conversely, the male fish may be having a stronger in-built resistance to the infestation, leading to the establishment of fewer parasites in them. This is not always true, within populations, individuals differ in their ability to compete for limited resources (Begon et al. 1990) and the resulting unequal division of nutrients leads to variation in growth rates, body size and nutritional condition (e.g. Rubenstein 1981; Metcalfe 1986; Westerberg et al. 2004). Unequal nutrient intake by competitors is also likely to have consequences for any parasites they may harbour, though it is difficult to predict the direction of such effects. These parasites are completely dependent on host derived energy for growth and development (Bush et al. 2001), infecting better competitors might benefit parasites, particularly those with significant energetic requirements. Alternatively, if the best competitors are either in better nutritional condition as a result of their competitive superiority, or of intrinsically higher genetic quality, then they may be poor hosts for parasites if they have better immune systems or are able to limit the availability of nutrients to growing parasites. This was found among female individuals of *T. guineesis* with low condition factor compared to their high condition females. Toxicants have high potential of reducing immunological response. These pollutants have shown to cause reduction in reproductive potential, bioenergetics and impaired growth in fish host (Dick 2009).

Thus, it can be concluded that the present work showed noticeable variations within the host population dynamics that examined host relative condition inclusive to that of the other researchers examine difference in host species, feeding biology of the host, diversity of the climatic conditions and the availability and infectivity of the intermediate host. Generally, the host-parasite accumulation had shown that increase in host tissue metal concentration resulted to subsequent increased in the parasite concentration and this is dependent on their condition factor at the time of exposure and after. Parasitic infestation within fish population could either increase or decrease metal accumulation in the fish, depending on the fish condition, sex and the type of parasite harbored. Low condition host and parasites accumulate less heavy metals than the high condition host and parasites. Difference in condition status within the host population under heterogeneous parasitic infestation affects or alters parasite prevalence, load and host-parasite metal accumulation. Therefore, it is recommended that environmental monitoring studies involving the use of fish could be improved upon by including the macro-parasites, considering the significant effect they could have in the fish biology, behavior and ecology.

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