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PROXIMATE ANALYSIS AND HEAVY METALS IN MOLLUSCS, *Tympanotonus fuscatus* (LINNAEUS, 1758) AND *Pachymelina aurita* (MÜLLER, 1774) FROM THE RIPARIAN AREA OF LAGOS LAGOON, SOUTH - WESTERN NIGERIA.

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Abstract

An investigative study was carried out to determine the proximate composition, heavy metals and ratio of foot weight to shell weight of Tympanotonus fuscatus and Pachymelina aurita in the riparian area of the University of Lagos between March and May, 2017. The shell length, foot weight and shell weight of T. fuscatus ranged from 1.80-6.00 cm, 0.10-2.10 g and 0.30-6.40 g respectively and has means of 3.48±0.98 cm, 0.63±0.41 g and 2.21±1.37 g respectively. Also, the shell length, foot weight and shell weight of P. aurita ranged from 1.70-5.70 cm (3.44±0.85 cm), 0.10-1.70 g (0.62±0.37 g) and 0.30-6.40 g (2.13±1.17 g) respectively. The proximate compositions obtained were: %Protein (T. fuscatus: 47.53±20.79 and P. aurita: 45.83±19.74), %Fat (T. fuscatus: 4.75±4.24 and P. aurita: 2.00±0.00), %Fibre (T. fuscatus: 1.13±0.88 and P. aurita: 2.38±0.18), %Ash (T. fuscatus: 15.25±1.77 and P. aurita: 17.75±17.32) and %Moisture (T. fuscatus: 17.75±17.32 and P. aurita: 19.50±14.14). There was no significant difference in the means of the proximate compositions of the species when compared with a Student t test. % Foot Weight: % Shell Weight of T. fuscatus was 22.16:77.85 and % Foot Weight: % Shell Weight of P. aurita was 22.56:77.45. All the heavy metals analyzed were below the limit recommended by FAO/WHO except chromium in P. aurita and Iron that were above the safe level in both species. It should therefore be noted that T. fuscatus and P. aurita found around the riparian area of the Lagos Lagoon, though being good protein sources of food, may pose risk of chromium and iron induced illnesses.

Keywords: *Tympanotonus fuscatus, Pachymelina aurita*, Proximate, Heavy metal, lagoon.

Introduction

Periwinkles are invertebrates belonging to the phylum Mollusca, class Gastropoda and subclass Prosobranchia (Dorit *et al*., 1991). These gastropods belong to a successful group of organisms which live in a variety of habitats with the majority being aquatic (Adebayo-Tayo *et al*., 2006). They are capable of burrowing into the mud in their habitats and are basically marine mollusks usually found dominating the brackish water environment (Adebayo-Tayo *et al*., 2006). In Nigeria, they occur in lagoons, estuaries and mangrove swamps and are represented by two genera (*Tympanostonus and Pachymelania*) (Olaniyan, 1968; Adebayo-Tayo *et al*., 2006; Emmanuel and Ogunwenmo, 2010) and are usually harvested by hand-picking.

Periwinkles constitute one of the relatively easily harvested shell fishes occurring widely within the upper intertidal zone of coastal areas (Castrol & Huber, 2005).

The use of icthyofauna of water bodies especially fish and shellfishes for monitoring anthropogenic pollution sources in different environmental matrices and toxicity through ingestion has received tremendous scientific attention in Nigeria. However, there is still limited toxicological evidence on some of these metals, especially their health risk indices and organ affinity from consumption in tropical countries (Freeman and Ovie, 2017).

Heavy metals are intrinsic, natural constituents of our environment. They are generally present in small amounts in natural aquatic environments (Aderinola *et al*., 2009). Apart from the natural sources, several anthropogenic ones also contribute to metal concentrations in the environment. In recent times, industrial activities have raised natural concentrations causing serious environmental problems. Marine coastal ecosystems could therefore be endangered by pollutants, such as heavy metals, pesticides and antifoulants that could be easily detected in the water column or in the sediment of harbours and estuaries (Antizar, 2008; Bellas, 2005).

Heavy metals, such as cadmium, lead, chromium, mercury and metalloid of arsenic have been reported with high toxicity and in carcinogenicity (Tchounwou *et al*., 2014). These heavy metals are of severe environmental and public health significance and they connote systemic toxicants that are known to induce

According to WHO (1971) and Staniskiene *et al.* (2006), the monitoring of eight elements (Hg, Cd, Pb, As, Cu, Zn, Fe, Sn) in fish is obligatory. Hence, the determination of harmful and toxic substances in the shell fishes *Tympanotonus fuscatus* and *Pachymelina aurita.*

In view of the wide consumption of these two periwinkle species in the coastal regions of Nigeria, there is need to assess the proximate and heavy metals of the species to ascertain their nutritional value for the purposes of proper utilization for human and animal nutrition and also to know how safe it is to consume them.

The aim of this study is to provide information on the proximate composition, heavy metals and ratio of foot weight to shell weight of *Tympanotonus fuscatus* and *Pachymelina aurita* in the riparian area of the Lagos Lagoon, South-Westhern Nigeria.

Materials and Methods

Collection of Ssamples

Four hundred (400) matured samples of each species of *Tympanotonus fuscatus and Pachymelina aurita* were handpicked from riparian areas of Lagos lagoon around the University of Lagos, Nigeria (Figure 1) twice within a period of three months (March to May) for proximate analysis and heavy metals analysis. University of Lagos, Nigeria is located on latitude 6.51679°N and longitude 3.39583°E. *Tympanotonus fuscatus* was collected from the creek (6º30'49.0" N, 3º23'59.9" E), where the sediment is mud and no dump site can be sighted around the vicinity to alienate the samples from impurity. *Pachymelina aurita* was collected from the open lagoon (6º31'4.5" N, 3º24'11.3" E), where the sediment is coarse in texture and a dump site could be sighted around the vicinity. *P. aurita* were found to adapt to coarse sediment texture (Emmanuel and Ogunwenmo, 2010). The samples were collected in plastic buckets and immediately transported to the Research laboratory in the Department of Marine Sciences, Faculty of Science, University of Lagos for morphometric measurement. The samples were then divided into halves and a part transported to Livestock Feed Lab, Ikeja, Lagos, Nigeria for proximate analysis and the other part transported to Central Research Lab for digestion before being taken to Agilent Technologies, Lekki, Lagos, Nigeria for heavy metals analysis.

(Sampling sites in red)
Figure 1: Map of the study area

Proximate Composition

The proximate composition of collected fish samples was determined according to the AOAC (2005) methods. The moisture content was determined by drying samples overnight at 105 °C for 2 hr 30 mins. Crude protein content was determined using the Kjeldahi method. Fat content was determined as per using soxhlet method and the ash content was by ashing at 500°C until fully ashed. Carbohydrate content was determined by calculating the difference i.e. the sum total of moisture; protein, fat and ash content were subtracted from 100 %.

Heavy Metal Analysis

Sample Digestion and Determination of Heavy Metals

Sample digestion was done by using microwave digester Central Research Laboratory, Lagos, Nigeria. The concentration of heavy metals (Cadmium, Cobalt, Chromium, Lead, Copper, Iron and Zn) in each of the samples was determined using atomic absorption spectrophotometer (AAS) (AOAC, 2005) UNICAM SOLAAR32 model at Agilent Technologies, Lagos, Nigeria.

Statistical Analysis

The results of proximate analyses obtained were subjected to Student *t*-test using the Statistical Package for Social Sciences (SPSS 16.0, 2015) to test for significant differences. Statistical significance was accepted at 5% probability level. Linear Regression equations and plots were developed to assess the relationship among the morphometric properties of each mollusc.

Results

Morphometric Analysis of *Tympanotonus fuscatus* and *Pachymelina aurita* from Riparian Areas of Lagos Lagoon.

The shell length (SL), foot weight (FW) and shell weight (SW) of each *Tympanotonus fuscatus* and *Pachymelina aurita* were measured and the summary is presented in Table 1. The shell length of *Tympanotonus fuscatus* ranged from 1.80-6.00 cm and has a mean of 3.48±0.98 cm, a foot weight range of 0.10-2.10 g with a mean of 0.63±0.41 g and a shell weight range of 0.30-6.40 g with a mean of 2.21±1.37 g. The shell length of *Pachymelina aurita* ranged from 1.70-5.70 cm and has a mean of 3.44±0.85 cm, a foot weight range of 0.10-1.70 g with a mean of 0.62±0.37 g and a shell weight range of 0.30-6.40 g with a mean of 2.13±1.17 g. The shell length's relationship with *T. fuscatus* foot weight, *T. fuscatus* shell weight, *P. aurita* foot weight and *P. aurita* shell weight are presented in figures 3, 4, 5 and 6 respectively. The linear regression models of these relationship, R and the R^2 values are presented in Table 2.

Table 1: Morphometric analysis of *T. fuscatus* and *P. aurita* from riparian areas of Lagos lagoon

Parameter		Species: *Tympanotonus fuscatus*	Species: *Pachymelina aurita*
	N	400	400
Shell Length (cm)	Range	1.80-6.00	1.70-5.70
	Mean	3.48±0.98	3.44±0.85
Foot Weight (g)	Range	0.10-2.10	0.10-1.70
	Mean	0.63±0.41	0.62±0.37
Shell weight (g)	Range	0.30-6.40	0.30-6.40
	Mean	2.21±1.37	2.13±1.17

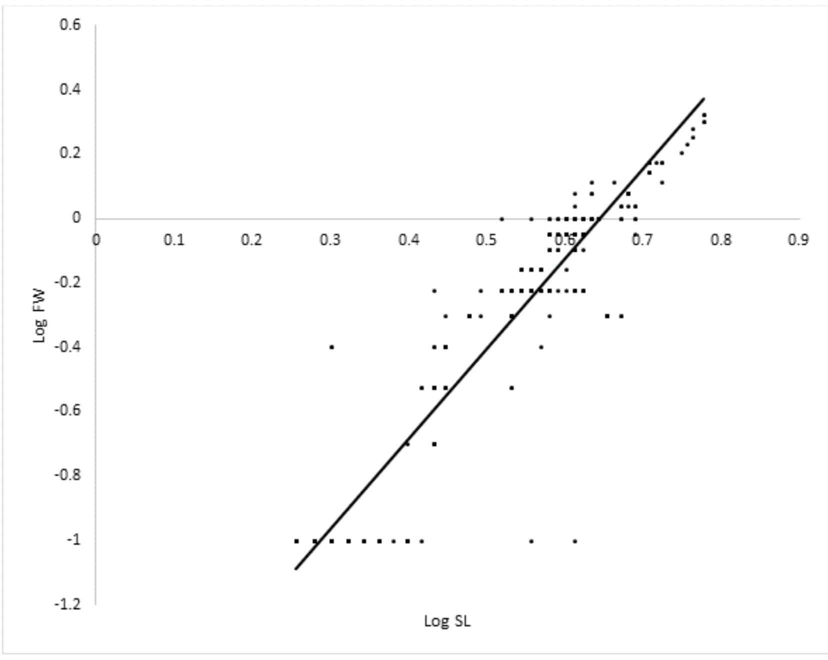


Figure 2: Shell length-foot weight relationship of *Tympanotonus fuscatus* from riparian areas of Lagos lagoon.

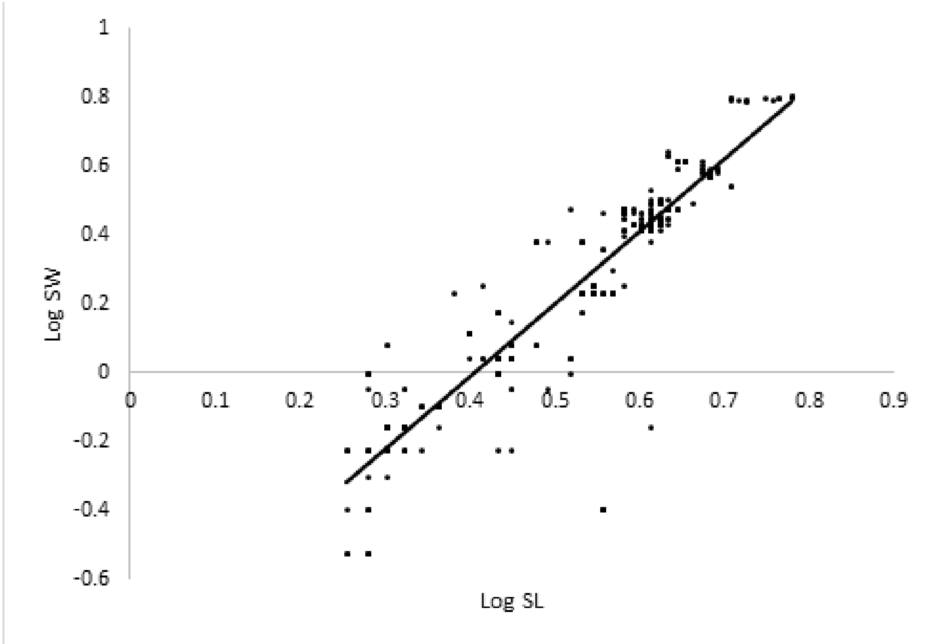


Figure 3: Shell length-shell weight relationship of *Tympanotonus fuscatus* from riparian areas of Lagos lagoon.

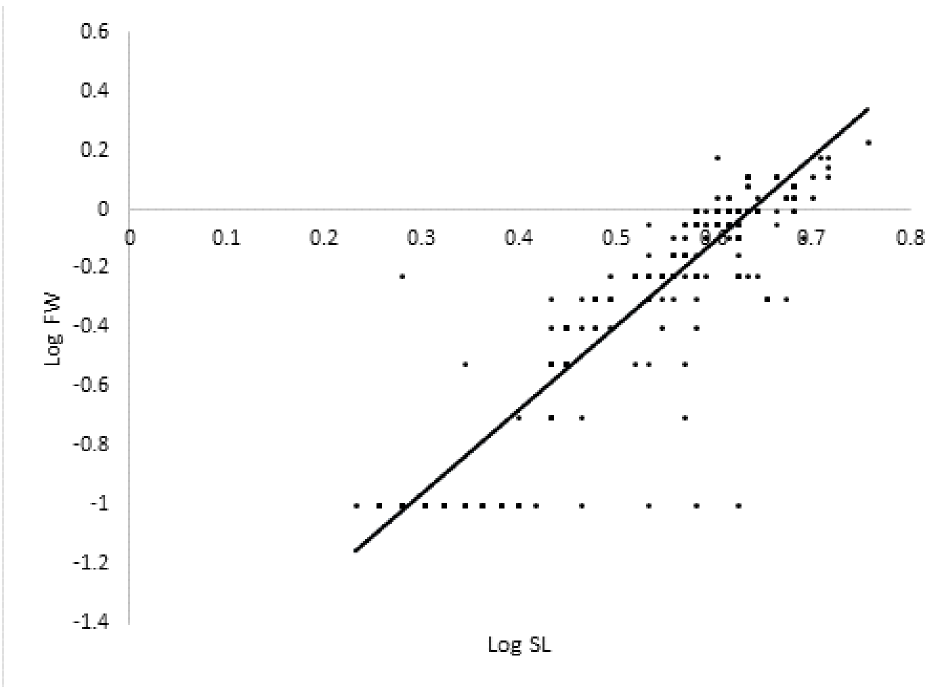


Figure 4: Shell length-foot weight relationship of *Pachymelina aurita* from riparian areas of Lagos lagoon.

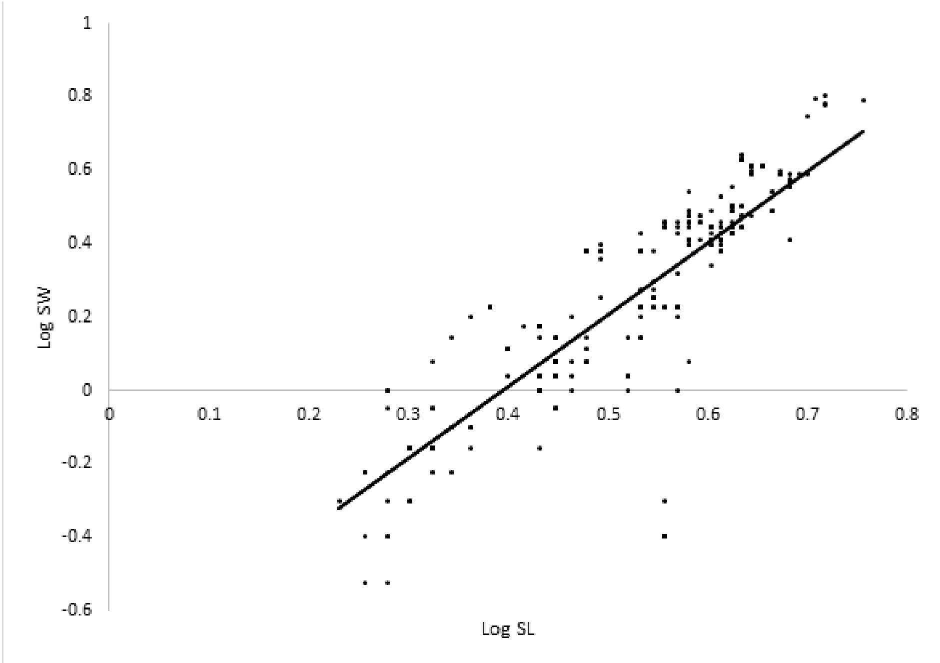


Figure 5: Shell length-shell weight relationship of *P. aurita* from riparian areas of Lagos lagoon.

Table 2: The linear regression models morphometric relationships of *T. fuscatus* and *P. aurita*

Species	parameters	Equation	R (regression coefficient)	R^2 (correlation coefficient)
T. fuscatus	FW on SL	y = 2.7926x - 1.8003	0.930	0.865
	SW on SL	y = 2.1116x - 0.8552	0.898	0.806
P. aurata	FW on SL	y = 2.8388x - 1.8065	0.911	0.829
	SW on SL	y = 1.9562x - 0.772	0.806	0.650

FW: Foot weight, SW: Shell weight, SL: Shell weight

Variation in morphometric parameters of *T. fuscatus* and *P. aurita*

There were no statistically significant differences between the shell lengths, foot weights and shell weights of *T. fuscatus* and *P. aurita* Shell lengths foot weights and shell weights as shown in Table 3.

Table 3: Variation in morphometric parameters of *T. fuscatus* and *P. aurita*

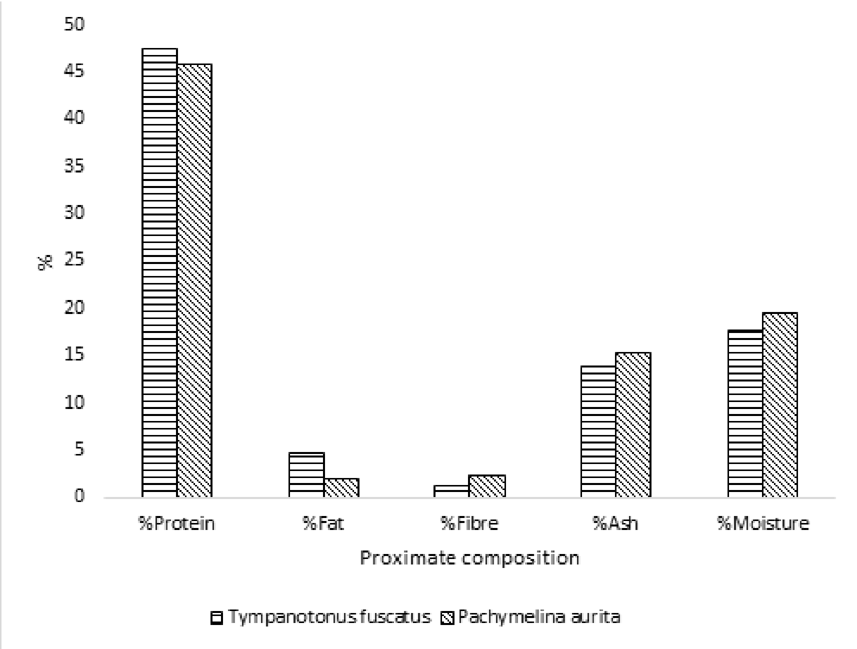
Parameters	*T. fuscatus*	*P. aurata*	t-test	f-ratio	$P \leq 0.05$
Shell length	3.48±0.98	3.44±0.85	0.641	0.522	0.05
Foot weight	0.63±0.42	0.62±0.37	0.360	0.719	0.05
Shell weight	2.21±1.37	2.13±1.17	0.942	0.942	0.05

Proximate Analysis of *T. fuscatus* and *P. aurita* from Riparian areas of Lagos Lagoon.

The proximate composition of *Tympanotonus fuscatus* and *Pachymelina aurita* are presented in Table 4 and Figure 6. Student *t* test revealed that there was no significant difference in the proximate composition of the two species. *T. fuscatus* has a protein content of 47.53±20.79% while *P. aurita* has a protein content of 45.83±19.74%. *T. fuscatus* has a fat content of 4.75±4.24% while *P. aurita* has a fat content of 2.00±0.00%. *T. fuscatus* has a fibre content of 1.13±0.88% while *P. aurita* has a fibre content of 2.38±0.18%. *T. fuscatus* has an ash content of 13.88±3.01% while *P. aurita* has an ash content of 15.25±1.77%. *T. fuscatus* has a moisture content of 17.75±17.32% while *P. aurita* has a moisture content of 19.50±14.14%.

Table 4: Proximate composition of *Tympanotonus fuscatus* and *Pachymelina aurita*

Nutrient	**Periwinkle**	**Range (%)**	**Mean (%)**	***t* (798)**	**Sig.**
%Protein	*T. fuscatus*	32.83-62.23	47.53±20.79		
	P. aurita	31.87-59.78	45.83±19.74	0.084	0.941
%Fat	*T. fuscatus*	1.75-7.75	4.75±4.24		
	P. aurita	2.00-2.00	2.00±0.00	0.917	0.528
%Fibre	*T. fuscatus*	0.50-1.75	1.13±0.88		
	P. aurita	2.25-2.50	2.38±0.18	-1.961	0.286
%Ash	*T. fuscatus*	11.75-16.00	13.88±3.01		
	P. aurita	14.00-16.00	15.25±1.77	-0.558	0.644
%Moisture	*T. fuscatus*	5.50-30.00	17.75±17.32		
	P. aurita	9.50-29.50	19.50±14.14	-0.111	0.922



Heavy metals Levels in *Tympanotonus fuscatus* and *Pachymelina aurita*.

The levels of heavy metals in *Tympanotonus fuscatus* and *Pachymelina aurita* are presented in Table 5. Cadmium (Cd) level in *Tympanotonus fuscatus* and *Pachymelina aurita* are 0.1122 ppm and 0.1542 ppm respectively, Cobalt (Co) level in *Tympanotonus fuscatus* and *Pachymelina aurita* are 0.0544 ppm and 0.0565 ppm respectively, Cromium (Cr) level in *Tympanotonus fuscatus* and *Pachymelina aurita* are 0.4024 ppm and 1.2896 ppm respectively, Cupper (Cu) in *Tympanotonus fuscatus* and *Pachymelina aurita* are 1.0444 ppm and 0.9376 ppm respectively.Iron (Fe) level in *Tympanotonus fuscatus* and *Pachymelina aurita* are 7.3169 ppm and 27.3465 ppm respectively, Lead (Pb) level in *Tympanotonus fuscatus* and *Pachymelina aurita* are 0.0363 ppm and 0.0585 ppm respectively and Zinc (Zn) level in *Tympanotonus fuscatus* and *Pachymelina aurita* are 0.9011 ppm and 0.0184 ppm respectively.

Table 5: Levels of heavy metals *Tympanotonus fuscatus* and *Pachymelina aurita*

Heavy metals	**Species**		**FAO/WHO limit**
	Tympanotonus fuscatus	*Pachymelina aurita*	
Cd (ppm)	0.1122	0.1542	2.00
Co (ppm)	0.0544	0.0565	
Cr (ppm)	0.4024	1.2896	1.00
Cu (ppm)	1.0444	0.9376	0.40
Fe (ppm)	7.3169	27.3465	2.50
Pb (ppm)	0.0363	0.0585	0.30
Zn (ppm)	0.9011	0.9184	1.00

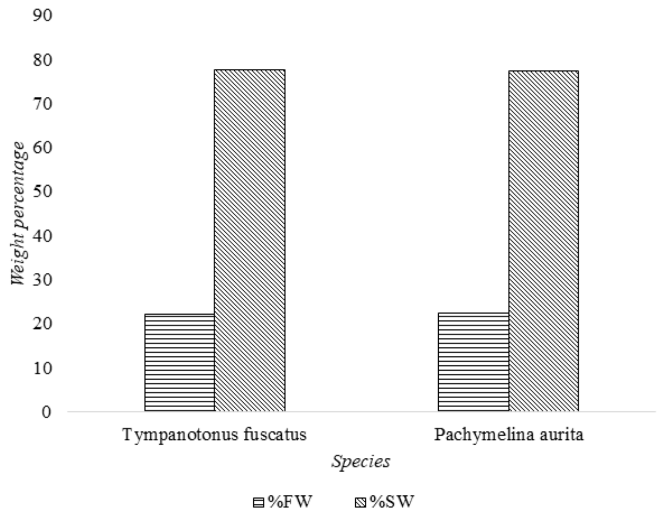


Figure 7: Percentage foot weight ratio percentage shell weight of *Tympanotonus fuscatus* and *Pachymelina aurita* from riparian areas of Lagos lagoon.

Discussion

The shell length of *Tympanotonus fuscatus* and *Pachymelina aurita* used in this study ranged from 1.80-6.00 cm and 1.70-5.70 cm respectively. The correlation coefficient for length-weight relationship (SL – FL) was high for T. *fuscatus* (r = 0.930) indicates a strong correlation and allows a fair calculation of weight for a given length. This agrees with earlier studies involving the same species from different parts of Niger-Delta (Jamabo *et al.* 2009; Udo 2013; Solomon *et al.* 2017). The value for growth exponent, b, obtained for the species is within the limit or range of 2 and 4 reported for most shell-fish (Bagenal and Tesh, 1978). Shell length-foot weight relationship of *T. fuscatus* revealed a positive allometric growth, indicating that an increase in the shell length also lead to an increase in the weight of the foot. Also, shell length-shell weight relationship of *T. fuscatus* revealed a positive allometric growth which indicates that an increase in the shell length also lead to an increase in the weight of the shell. Shell length-foot weight relationship of *P. aurita* revealed a positive allometric growth, indicating that an increase in the shell length also lead to an increase in the weight of the foot and the shell length-shell weight relationship of *P. aurita* revealed a negaive allometric growth which indicates that an increase in the shell length does not lead to increase in the weight of the shell as noted by Jamabo *et al.* (2009). The shell length doesn't necessarily affect the foot weight of the periwinkles in this study.

Percentage crude protein of *T. fuscatus* observed in this study ranged from 32.83 to 62.23% while Adebayo-Tayo and Ogunjobi (2008) reported that crude protein of *Tympanotonus sp*. ranged from 41.39 to 60.04% while comparing the effects of oven-drying and sundrying on microbiological, proximate nutrient and mineral composition of the periwinkle *Tympanotonus sp*. and Oyster *Grassostrea sp*. from Itu River, in Akwa Ibom State.

The insignificant difference in the nutritional values of *T. fuscatus* and *P. aurita* in this study might be due to the richness in nutrient of the environment in which the two organisms were explored from.

All the heavy metals analyzed were below the limit recommended by FAO/WHO except chromium in the case of *P. aurita* and Iron that was above the safe level in both species. Chromium (Cr) concentration in the *P. aurita* was above the FAO/WHO limit of 1.00 ppm. Organisms accumulate heavy metal like chromium either directly from the surrounding water or by ingestion of food in environment (Said *et al*., 1992). Chromium is a carcinogen, particularly of the lung through inhalation (Pompella, 2003).

Iron (Fe) was found to have the highest concentration in all the samples analyzed. Iron (Fe) concentration in all the periwinkle species samples were extremely above the FAO/WHO limit of 2.5 ppm. It has been reported that iron occurs at high concentration in Nigerian soils (Asaolu and Olaofe, 2004). Fe is involved in the haemoglobin synthesis in the red blood corpuscles of the blood, Fe also help with red blood cell production. It is a necessary element in human diet and plays a significant role in metabolic processes. In this study, the observed mean value of Fe in the samples far exceeded the FAO/WHO recommended limits of 0.50 ppm in fish foods (FAO/WHO, 2015). Though an essential heavy metal, Fe has the tendency to become toxic to living organisms, even when exposure is low.

T. fuscatus and P. aurita may be useful as a potential indicators of metal pollution, but it should be remembered that there is no certainty that the metal concentration in the environment will be accurately reflected in the tissue of the periwinkles used for this study. The levels of heavy metals recorded in this study was low, this may be as result of degree of regulation and elimination of metal in the body of shell fishes (Sayyad *et al.,* (2007).

The ratio of percentage foot weight to percentage shell weight of *T. fuscatus* and *P. aurita* were the same (1:4), which means just 20% of the perinwinkle is consumed and 80% are often discarded as waste. Egonmwan (2008) reported that a positive relationship existed between mollusc shells and calcium concentration. The presence of these vital mineral needed for strong teeth in mollusc shells warranted their inclusion in formula for washing teeth in rural areas of Ondo state where the shell is oven dried and ground into fine powder and kept in safe container (Ademolu, *et al*., 2015). Base on this fact, shells of periwinkle should therefore be washed, oven dried and ground into fine powder and properly packaged for dental purposes in Nigeria.

It should therefore be noted that *T. fuscatus and P. aurita* found around the riparian area of Lagos Lagoon, though being good protein sources of food, pose risk of chromium and iron induced illnesses if consumed in large quantity.

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