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Baseline serum biochemical profile of *Pachymelania fusca* (Gastropoda: Melanidae) from two tropical lagoon ecosystems

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ABSTRACT

Serum biochemical profile is often applied as an index of physiological condition of invertebrates and thus provide information about the health status of local populations. The study aimed to establish normal ranges for chosen serum biochemical parameters of gastropod species, *Pachymelania fusca* (Gmelin, 1791) from Ologe and Badagry Lagoons using standard methods. In terms of water quality, Badagry Lagoon showed significant (P < 0.05) higher values in total dissolved solid (88.45 mg/L), chemical oxygen demand (41.88 mg/L), biological oxygen demand (35.73 mg/L), total suspended solid (100.44 mg/L), total hardness (102.06 mg/L), sulphate (34.31 mg/L) and nitrate (1.43 mg/L). In dry season, *P. fusca* from Ologe Lagoon had higher activities of aspartate aminotransferase (AST) (29.77±1.78 u/l), alanine aminotransferase (ALT) (11.69±1.68 u/l) and alkaline phosphatase (ALP) (140.58±49.64 u/l) than the activities of that from Badagry Lagoon. In wet season, the activity of ALP in *P. fusca* from Badagry Lagoon ranged from 63.48 to 118.68, and significantly higher than the range of 39.68 to 104.88 in *P. fusca* from Ologe Lagoon. Total protein was also higher in *P. fusca* from Badagry Lagoon, ranging from 10.77 to 42.46 g/L. The haemolymph activity of *P. fusca* from both studied locations varied across season.

Keywords: Biochemical, lagoon, mollusc, serum metabolites.

INTRODUCTON

Molluscs are the largest invertebrate animals and are important resources that contribute considerable economic value to the world's fisheries (Moruf *et al.*, 2020). The global production of marine mollusc for human consumption is more than 17 million tonnes in the year 2018, with China as the major producer with relatively highest percentage of production (FAO, 2020). The phylum, Mollusca is known to radiate successfully into a variety of habitats, the great majority

of which are aquatic while some are found mostly in shallow waters and sometimes in intertidal zones, where they burrow into the mud in the beds of the river, which serves as their habitat (Appleton *et al.*, 2009; Moruf *et al.*, 2018). Accordingly, bivalves and gastropods comprise the most researched molluscs in relation to natural products isolation, and account for roughly 98% of the total molluscan species (Avila, 2006; Moruf *et al.*, 2021). These include animals such as snails, clams, mussels, and oysters that are

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widely used as bio-indicators of ecosystem health due to their significant response to changes in aquatic environment.

The gastropod, Pachymelania fusca (Gmelin, 1791) is a representative of family Melanidae, Super family Cerithiacea and they are found in lagoons, estuaries and mangrove swamps of West Africa. Genus Pachymelania adapts to fresh water but prefers brackish water of higher salinity and is often extremely abundant in the mangrove swamps and on the mud flats within reach of the tide, in the lagoons and river estuaries. Gastropods generally play a key role in connecting primary producers, herbivores, and dentritivores, where the abundance, composition and diversity of gastropods present in sediments can act as a strong for both ecological indicator and hydrological changes (Ajani et al., 2020). The structure, shapes and the chemical composition of the shells can give indications of the past conditions of a water body (Kattel et al., 2018).

Studies of blood chemistry involve measurement of the chemical components in serum or plasma and this includes a wide range of electrolytes, enzymes and hormones (Petri et al., 2006). Serum and plasma from aquatic organisms are characterized by a number of proteins (trypsin, lysozyme, antibodies, C-protein, complement factors and other lytic factors) which play critical roles as antimicrobial agents, and are the first line of defense, primary barrier against invasion and contain the proliferation of pathogens, including parasites (Adeogun et al., 2015). The study of Tunholi-Alves et al. (2012) also validated the use of aminotransferases activity as excellent biomarkers of cell injury in gastropods, especially in the digestive gland.

The shell characteristics, classification and geographical distribution of the genus have been reported (Egonmwam, 2007). The ecology and natural food components of P. aurita has been documented (Uwadiae et al., 2009). The production and population dynamics of P. aurita in the brackish water Lagos Lagoon have been studied by Ajao and Fagade (1990). Other studies conducted on several mollusc species in Lagos lagoon complex had focused on the contaminants and antioxidant stress responses (Moruf and Akinjogunla, 2018; Usese et al., 2019; Afolayan et al., 2020; Moruf and Durojaiye, 2020), but until now, little or no known published information is available on the serum chemistry of brackish gastropod in Nigeria. This study to our knowledge is the first study aimed at examining the serum biochemical profiles P. fusca from two tropical lagoon ecosystems in Nigeria.

MATERIALS AND METHODS Study locations

This study was carried out along the coastal waters of Ologe Lagoon and Badagry Lagoon (Figure 1). These lagoons meet several socioeconomic needs (aquaculture, fishing, sand dredging and drainage) of the various towns and villages bordering it. Twelve (12) stations were selected across the two lagoons (Table 1).

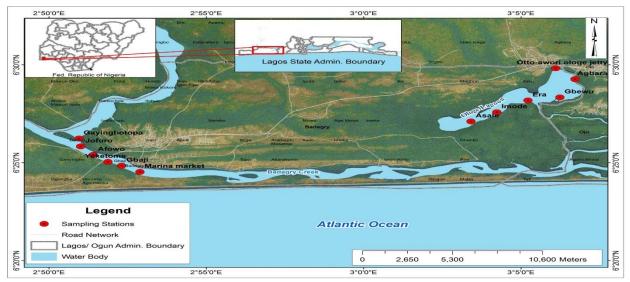


Fig. 1. Map indicating sampling stations (Ologe and Badagry Lagoons)

Table 1. GPS coordinates of sampling sites across two tropical lagoon ecosystems

		1 0	1 0	
S/N	Ologe Lagoon		Badagry Lagoon	
	Sampling Stations	GPS	Stations	GPS
1	Otto-awori ologe jetty	6°29′ 495″ N	Gayingbotopa	6°25′ 415″ N
		3°60′ 618″ E		2°51′ 161″ E
2	Gbewu	6°29′ 231″ N	Joforo	6°29′ 231″ N
		3°60′ 621″ E		2°60′ 621″ E
3	Era	6°28′ 972″ N	Afowo	6°25′ 672″ N
		3°60′ 824″ E		2°50′ 809″ E
4	Imode	6°28′ 838″ N	Yeketome	6°25′ 179″ N
		3°60′ 907″ E	i eketoine	2°51′ 61″ E
5	Asale	6°28′ 679″ N	Gbaji	6°25′ 075″ N
		3°60′ 930″ E		2°51′ 819″ E
6	Agbara	6°29′ 646″ N	Marina market	6°42′ 646″ N
		3°60′ 435″ E		2°60′ 435″ E

Sample collections and preparation

Parameters for water chemistry were measured between Aug. 2016 and Jul. 2018 with a Consort CT-C933T Electrochemisrty meter (TOPAC Instruments). Freshly harvested gastropod *P. fusca* samples used in the study were collected from these 12 stations. The samples were of a healthy appearance and good health condition. In the preparation, sample were dissected and the hemolymph was collected by cardiac puncture, stored in Eppendorf tubes and

maintained at 10^{0} C for the biochemical analyses. The volume of haemolymph (0.3 - 0.5 ml) used for biochemical analyses were obtained from thirty snails each from the two lagoons analyzed individually.

Laboratory procedure and data collection

The collected haemolymph was centrifuged (1000 rpm, 1 minute) at room temperature and the obtained material was analysed using colorimetry (Mindray BS-130) as described by Coles (1986) for the following

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parameters: aspartate aminotransferase (AST), alanine aminotransferase (ALT) and the alkaline phosphatase (ALP) activities. Cholesterol profile was determined using the method of Allain *et al.* (1974) for the determination of $\Delta 4$ – cholestenone after enzymatic cleavage of the cholesterol ester by cholesterol esterase. Total protein assay was performed according to the biuret technique as described by Tunholi-Alves *et al.* (2012).

Data analysis

The obtained results were analysed statistically by calculating the arithmetic mean and standard deviation, which then served for establishing the proposed normal ranges of the studied parameters. Using Microsoft excel 2010, the data were then subjected to one way analysis of variance (ANOVA) at P < 0.05.

RESULTS

Water chemistry of the tropical lagoons

The variation in water quality indices at Ologe and Badagry lagoons between Aug. 2016 and Jul. 2018 is shown in Figure 2. Badagry Lagoon showed significant (P < 0.05) higher values in total dissolved solid, (88.45 mg/L),chemical oxygen TDS demand, COD (41.88 mg/L), biological oxygen demand, BOD (35.73 mg/L), total suspended solid, TSS (100.44 mg/L), total hardness, (102.06 mg/L), sulphate (34.31 mg/L) and nitrate (1.43 mg/L). Dissolved oxygen (DO) was slightly higher in Ologe Lagoon (4.86 mg/L) compared to Badagry Lagoon (4.77 mg/L) with no significant difference (P > 0.05). Furthermore, values for phosphate ranged between 1.97 (Ologe Lagoon) and 2.23 mg/L (Badagry Lagoon)

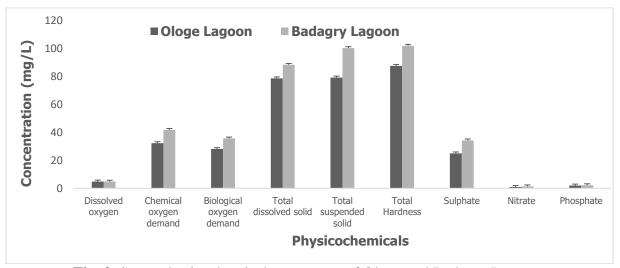


Fig. 2. Some physicochemical parameters of Ologe and Badagry Lagoons

Serum Biochemistry of *Pachymelania* fusca

The result of the seasonal variation in the serum biochemical profile of *P. fusca* from Ologe Lagoon is presented in Figure 3. Relatively, higher range values of total protein (23.04 g/L), AST (31.55 u/l) and cholesterol (57.66 mg/dL) were recorded in the gastropod during wet season. However, relatively higher range values for ALT (38.46-85.1 u/l) and ALP (35.5-59.94 u/l) were obtained in *P. fusca* during dry season.

Meanwhile in Badagry Lagoon, only AST (30.67 u/l) and ALP (97.42 u/l) were higher in the serum of the gastropod during wet season, while total protein (26.72 g/L), ALT (10.33 u/l) and cholesterol (72.11 mg/dL) were higher during dry season (Figure 4).

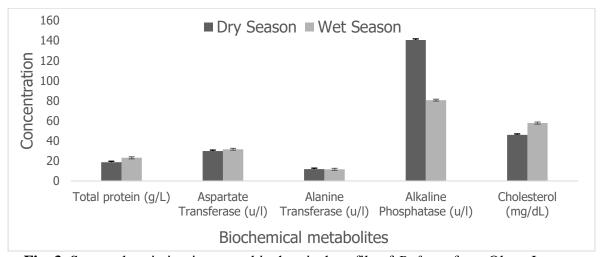


Fig. 3. Seasonal variation in serum biochemical profile of *P. fusca* from Ologe Lagoon

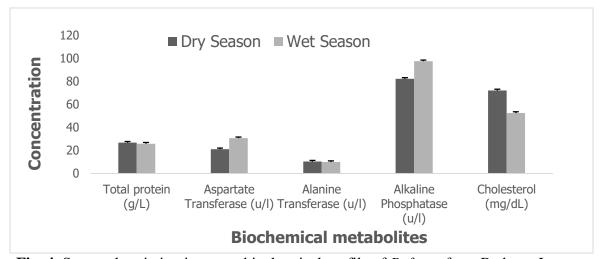


Fig. 4. Seasonal variation in serum biochemical profile of *P. fusca* from Badagry Lagoon

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The comparisons in the serum biochemical metabolites of *P. fusca* between Ologe and Badagry Lagoons during dry and wet seasons are presented in Tables 2 and 3, respectively. In dry season, *P. fusca* from Ologe Lagoon had higher activities of AST (29.77±1.78 u/l), ALT (11.69±1.68 u/l) and ALP (140.58±49.64 u/l) than the activities of that from Badagry Lagoon. In addition, the concentrations of total protein, AST, ALT

and cholesterol in *P. fusca* were significant different across the two tropical lagoon ecosystems (P< 0.05). In wet season, the activity of ALP in *P. fusca* from Badagry Lagoon ranged from 63.48 to 118.68, and significantly higher than the range of 39.68 to 104.88 in *P. fusca* from Ologe Lagoon. Total protein was also higher in *P. fusca* from Badagry Lagoon, ranging from 10.77 to 42.46 g/L.

Table 2: Serum biochemical profile of *P. fusca* from two tropical lagoon ecosystems (Dry season)

Parameters	Pachymela	- P-value		
r arameters	Ologe Lagoon	Badagry Lagoon	i -vaiue	
Total protein (g/L)	(6.15-27.99) 18.54±1.87	(16.61-39.37) 26.72±1.78	0.00*	
Aspartate transferase (u/l)	(16.80-40.63) 29.77±1.78	$(9.67-35.28)\ 21.03\pm1.96$	0.00*	
Alanine transferase (u/l)	(4.64-26.17) 11.69±1.68	$(6.40-15.04)\ 10.33\pm0.51$	0.44	
Alkaline phosphatase (u/l)	$(77.24-983.84) 140.58\pm49.64$	(44.68-102.12) 82.16±4.45	0.25	
Cholesterol (mg/dL)	$(22.00-68.20) 45.84\pm3.29$	$(27.50-147.40)$ 72.11 ± 7.88	0.00*	

Key: (Minimum -Maximum), Mean±Standard error, *= significant difference at P<0.05

Table 3: Serum biochemical profile of *P. fusca* from two tropical lagoon ecosystems (Wet season)

Parameters	Pachymelo	- P-value		
Farameters	Ologe Lagoon	Badagry Lagoon	- r-vaiue	
Total protein (g/L)	(8.61-42.14) 23.04±2.91	(10.77-42.46) 25.86±2.67	0.48	
Aspartate transferase (u/l)	$(10.15-49.74) 31.55\pm3.34$	$(10.00-67.00)\ 30.67\pm4.29$	0.87	
Alanine transferase (u/l)	(5.28-15.68) 11.54±0.92	$(2.56-18.19)\ 10.00\pm1.17$	0.31	
Alkaline phosphatase (u/l)	(39.68-104.88) 80.50±5.29	$(63.48-118.68) 97.42\pm4.10$	0.02*	
Cholesterol (mg/dL)	(27.60-87.40) 57.66±5.11	(27.50-100.00) 52.51±4.75	0.47	

Key: (Minimum -Maximum), Mean±Standard error, *= significant difference at P<0.05

DISCUSSION

The few physicochemical parameters estimated and characterized by this study reflect typical tropical lagoon water quality characteristics. For instance, DO, BOD and COD were within previously recorded limits (Ndimele and Kumolu-Johnson, Ogundele and Olarinde, 2018). DO level was slightly higher in Ologe Laggon than Badagry Lagoon. It is possible that the breaking of high energy plunging tides on the coastline was responsible for the increased dissolution of atmospheric oxygen in the waters. According to Lawal-Are et al. (2010),

DO decreases with increased BOD, probably due to increased metabolic activities of bacteria and fungi which are common in polluted sites. It is important to note however that BOD and COD can be employed to determine the level of pollution in an aquatic environment. BOD values less than 2.0 mg/L indicate clean water, 2.0 to 4.0 mg/L indicate moderate pollution while above 8.0 mg/L indicate severe stress (Nwankwo et al., 2013; Lawal-Are et al., 2019). In this regard, the physicochemical parameters of both studied locations fell within **FAO** (1994)recormended limits: DO (>4.0 mg/L), BOD

(8.0 mg/L), TDS (2000 mg/L), Sulphate (400 mg/L), Nitrate (50 mg/L) and Phosphate (2 mg/L).

Nutrients concentrations recorded in this study were high especially sulphates. The high levels of nitrate-nitrogen and sulphide may be due to the effect of direct discharges of pollutants and other biodegradable wastes into the coastal waters coupled with the enrichment of adjoining wetlands, creek and subsequent run-offs for the coastal water of south-western Nigeria (Lawal-Are et al., 2010). The high levels of nitrate may be due to the effect of direct discharges of pollutants and other biodegradable wastes into the coastal waters coupled with the enrichment of adjoining wetlands, creek and subsequent run-offs for the coastal water of South-Western Nigeria.

Variations in serum parameters have been attributed to responses to a changed physiological and energetic requirements and may be an early warning measure of stress before population declines are observed (Adeogun et al., 2015; Moruf and Lawal-Are, 2018). To better understand the changes observed in the serum biochemistry of P. fusca, from Ologe and Badagry Laggons, we determined the aminotransferases activity across seasons. Aminotransferases are a group of enzymes that catalyze the interconversion of amino acids into aketoacids by transfer of amine groups (Tunholi-Alves et al., 2015). They play an important role in the link between the amino acids and carbohydrate metabolism. Thus, the dry season increase in the activity of these enzymes in P. fusca from Ologe Lagoon is related to the mobilization of amino acids, favoring their depletion in groups from polluted environment. According to Sanni et al. (2020), the changes in AST could be attributed to the interference with the immune system of the organism, resulting to cell damage or a way in which the organisms are reacting to the exposure to pollutants. The mean total protein and cholesterol albumin levels in *P. fusca* from Ologe and Badagry Lagoons were higher than values i.e. 2.2±0.06 g/dL and 2.20±0.12 g/dL reported for *Bulinus globosus* snail under favourable environmental conditions (Akande *et al.*, 2010).

CONCLUSION

The present investigation concludes that most of the studied physico-chemical parameters are within the permissible limit of FAO for fish farming. The results of the research enabled the determination of physiological normal ranges for gastropod species, Pachymelania fusca from Ologe and Badagry Lagoons. Such species-specific baseline values provide a reference point for comparing responses of members of this taxon to differential habitat quality. Thus, if discharges are reduced by proper waste disposal, the water quality of the Ologe and Badagry Lagoons can be significantly improved. The authors recommended that further research be done in different seasons, considering other water quality parameters including heavy metals and trace organic compounds.

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