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Comparative Studies of Minerals and Proximate Composition in the Shells of Endemic Periwinkle Species of Nembe Community: A Potential Tool for Soil Amendment

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Abstract: This study examines the mineral and proximate composition of three periwinkle species shells endemic to Nembe mangrove swamp for their potential use in soil amendment. 150 periwinkle shells of three different varieties were harvested and their proximate and mineral compositions were determined (following standard procedures) in Biochemistry Lab. Federal University Otuoke. The results reveal morphological diversity and cultural relevance of the periwinkle shell varieties - Tympanotonus radula (Imoron), T. fuscatus (Emoru), and *Pachymelania aurita* (Buhari). Significant variations ($p \le 0.05$) in their fiber, ether extract, crude-protein, and mineral content were observed. T. fuscatys exhibited the lowest moisture content (35.51 ± 1.31) and the highest dry matter (561.87 ± 0.63) , whereas T. radula showed the opposite trend. T. fuscatus also recorded the highest crude fiber (4.68 \pm 0.41%) and crude protein (25.00 \pm 1.09%), while P. aurita had significantly lower values in both categories. In terms of mineral composition, T. fuscatus contained significantly higher (p<0.05) level of calcium, potassium, and magnesium than T. radula and P. aurita, making it particularly suitable for soil amendment and crop biofortification. T. radula had the highest phosphorus content (3.86 ± 0.18) , whereas iodine concentration was highest in T. fuscatus (6.90 \pm 1.05). The study concludes that periwinkle shells, especially those of T. fuscatus, hold significant potential for enhancing soil fertility and improving crop yields as natural soil amendments. This approach will not only support sustainable agricultural practices but also enriches the nutritional quality of crops.

Keywords: Periwinkle Shells, Minerals, Proximate Composition, Soil-Amendment, Nembe.

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INTRODUCTION

Periwinkles are ecologically significant mollusca endemic to coastal ecosystems, primarily inhabiting brackish marine environments such as mangrove swamps, creeks, and estuaries (Okpeku *et al.*, 2013). These prosobranch gastropods, commonly known as swamp snails, belong to the phylum Mollusca and the class Gastropoda (Jamabo and Alfred-Ockiya, 2005). They are conspicuous in the detritus-rich mud of coastal swamps and play an essential role in nutrient cycling. Beyond their ecological importance, periwinkles are a valuable dietary component, particularly in Nigeria's Niger Delta region (Ariahu and Ilori, 1992; Nwaka and Udoh, 2022; Otitoju and Otitoju, 2013; Jamabo and Alfred-Ockiya, 2005), where the shells are often discarded into the environment as waste after their meat is harvested (Oyawoye *et al.*, 2019).

In Bayelsa State, particularly among the Nembe people, periwinkle shells have long been repurposed as a cost-effective alternative to gravel in construction, serving as a conglomerate in concrete reinforcement for paving slabs, soakaways, and road building (Agbede and Manasseh, 2009). Periwinkle species of the genera *Tympanotonus* and *Pachymelania* are commonly found in the Nembe forest swamp (Ogamba *et al.*, 2016; Ekop *et al.*, 2021) and exhibit morphological variations on both micro- and macro-geographic scales (Okpeku *et al.*, 2013). Different varieties are endemic to the Nembe ecotone, with species distributed along the coastal regions of West Africa (Otitoju and Otitoju, 2013; Ogamba et al., 2016). Periwinkle shells possess a complex mineral composition that has garnered increasing interest for applications in engineering, agriculture, and nutrition (Dahunsi, 2003; Omisande and Onugba, 2020; Olawunmi and Imoobe, 2021). Biofortification, a strategy to combat micronutrient deficiencies-especially in developing countries like Nigeria-typically involves enhancing the nutritional content of food crops, soil (via organic amendments), and livestock feed (Bouis and Saltzman, 2017; Bouis, 2018). Soil amendments with mollusk shells have been shown to improve soil conditions and enhance micronutrient bioavailability for plant uptake (Dhaliwal et al., 2022). Additionally, periwinkle shells have been identified as a viable mineral source in animal feeds (Jamabo and Alfred-Ockiya, 2005). Anizoba et al., (2022) observed that periwinkle shell supplementation in poultry layers' diets provided superior calcium benefits compared to limestone, largely due to its high mineral content and affordability (Aimikhe and Lekia, 2021).

While biofortification efforts have predominantly focused on staple crops such as cassava, rice, and maize, there is growing interest in exploring local, natural mineral sources for soil amendments and livestock nutrition (Nestel *et al.*, 2006; Bouis *et al.*, 2011; Onuegbu *et al.*, 2017). Recent studies suggest that mollusk shells contain bioactive components and essential minerals that could be harnessed for improving

agricultural and nutritional outcomes (Nkansah, 2021). Thus, considering the mineral-rich environments of Nembe, periwinkle shells may represent an untapped resource for sustainable biofortification. Although, most existing studies have focus on the nutritional value of periwinkle meat rather than their shells. Considering the abundance of periwinkle species in the Nembe region and the limited research on the mineral composition of their shells, this study aims to compare and determine the proximate and mineral composition of periwinkle shells to assess their potential applications.

MATERIALS AND METHODS

Study Area

This study was carried out in Nembe Ecotone between the months of February and May 2023 in three local communities: Sabatoro, Basambiri and Obiama in Nembe Local Government Area of Bayelsa State, Nigeria. The area lies within coordinates 4^o 32' 22" N: 6^o 24' 01"E. The natives engage mainly in fishing at both subsistence and commercial scales. Other major occupations in the area are farming and trading. The Nembe boasts the most extensive mangrove zone in Nigeria. The area is ecologically significant due to its rich biodiversity and the presence of various aquatic species vital to the local economy and ecosystem. Nembe zone is influenced by the tides of the Atlantic Ocean; annual rainfall is between 2000 and 4500 mm.



Figure 1: Showing the study area in Nembe, Bayelsa State

Sample Collection and Identification

A total of 150 periwinkles from each variety were handpicked from the intertidal flats of the mangrove swamp environment in three communities: Sabatoro, Basambiri, and Obiama located in Nembe LGA. Initial identification of the different varieties was conducted by a team of ten native fishermen, who also provided the local indigenous names based on morphological characteristics. Proper scientific identification was later carried out in the Department of Biology, Federal University Otuoke. The collected samples were then transported to the laboratory for further analysis.

Sample Preparation

Three species of periwinkle, known to the indigenous people of Nembe as *Emoru*, *Imoron*, and *Bebefangala (Buhari)*, correspond to *Tympanotonus fuscatus var. fuscatus*, *Tympanotonus fuscatus var. radula*, and *Pachymelania aurita*, respectively. These species were assessed and processed separately. The flesh of each sample was extracted using a local method, which involved washing the shells under running tap water before placing them in hot water to facilitate flesh removal. The shells were then thoroughly washed to eliminate external contaminants such as soil and organic material, dried in an oven at 105°C, and ground into a fine powder using a mechanical grinder.

Proximate and Mineral Composition Analysis of Periwinkle Shells

Determination of the Proximate Composition

The proximate composition of the periwinkle shells, including moisture content, ash content, crude protein, crude fat (ether extract), dry matter, and crude fiber, was determined using standard laboratory techniques as prescribed by the Association of Official Analytical Chemists (AOAC, 2016; 2019). Specific methods employed include: Oven drying method for moisture content, Muffle furnace method for ash content, Kjeldahl method for crude protein, Soxhlet extraction method for crude fat, Gravity analysis for dry matter and Weende method (conventional crude fiber analysis) for crude fiber. Additionally, the procedures outlined by James (1995) and Maynarda and Looshi (2007) were used for dry matter and crude fiber determination, respectively.

Determination of Mineral Composition

The mineral composition of calcium, potassium, iron, and magnesium in the periwinkle shells was analysed using Flame Atomic Absorption Spectroscopy (AAS) as described by Skoog *et al.*, (2013). Shell samples were ground into a fine powder using a mechanical grinder to enhance acid digestion efficiency. A combination of nitric acid (HNO₃) and hydrochloric acid (HCl) was used for digestion, converting solid minerals into liquid form. The AAS instrument was calibrated using standard solutions of calcium, potassium, iron, and magnesium at specific

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wavelengths (Ca: 422.7 nm, Mg: 285.2 nm, K: 766.5 nm, Fe: 248.3 nm).

One gram of the powdered sample was weighed, treated with concentrated nitric acid (10 mL), and heated gradually to 150°C. Perchloric acid was added to ensure complete digestion. After cooling, the solution was filtered and diluted with deionized water before being analysed with AAS. The absorbance values were measured and compared with calibration curves to determine mineral concentrations, expressed in mg/kg shell sample.

Determination of Phosphorus, Iodine, and Selenium

The powdered shell samples were passed through strong acid digestion using nitric acid (HNO₃), perchloric acid (HClO₄), and hydrochloric acid (HCl), followed by instrumental analysis for phosphorus, iodine, and selenium.

Iodine Determination (UV-Visible Spectrophotometry)

Following Fuge and Johnson (1986), the sample was digested in nitric acid (HNO₄) and perchloric acid (HClO₄) and diluted to 100 mL. Potassium iodide (KI) was added to react with iodine, forming a triiodide ion (I₃⁻), which was stabilized using sulfuric acid (H₂SO₄). The absorbance was measured at 350 nm using a UV-visible spectrophotometer, with iodine concentration determined by comparing absorbance values with a standard calibration curve.

Determination of Selenium (ICP-MS)

Inductively Coupled Plasma Mass Spectrometry (ICP-MS) was used, as per Hatfield *et al.*, (2014), for sensitive detection of selenium. The digested sample was ionized, and selenium concentration was measured based on the mass-to-charge ratio of the selenium ions.

Phosphorus Determination (Molybdenum Blue Spectrophotometry)

Phosphorus was analysed using the Molybdenum Blue Method (AOA C, 2019). A reaction between ammonium molybdate and ascorbic acid in acidic conditions formed a molybdenum-phosphate blue complex. The intensity of the colour was measured at 880 nm, and phosphorus concentration was calculated based on standard calibration curves, with results expressed in mg/kg shell sample.

Statistical Analysis

The data obtained were analyzed (with SPSS 2022 version) using descriptive statistics (Mean \pm SD), and ANOVA was used to determine statistical significance (p < 0.05).

RESULTS AND DISCUSSION

Table 1 shows the morphological diversity and local cultural significance of the three periwinkle species (*Tympatonus fuscatus* Var. fuscatus, *Tympatonus*

fuscatus Var. radula, and *Pachymelania aurita*) accessed in Nembe. The morphological features help distinguish the species from one another. The differences are not only visually distinct but also functional, as each species is likely adapted to its specific ecological niche within Nembe's mudflats. The table also reveals how Nembe people identify these species with unique local names, suggesting a depth of traditional ecological knowledge, as local names often reflect indigenous uses or distinguishing characteristics. It was observed that *Pachymelania aurita* (Bebefangala) is typically discarded upon harvesting due to its unpalatable quality, which was related to taste, texture, and the shell toughness. The morphological features of the three species inform biofortification strategies considering their uses, which are due to the mineral compositions and relatedness.

Scientific name	Family/Genus	Local name	Shell morphological features	Image
Tympatonus	Littorinidae/	Emoru (common	Conical and turreted spiny shell with a	Month -
fuscatus Var.	Tympanotonus	name: Isemi,	pointed apex, and prominent whorls.	and the second
fuscatus		periwinkle)		-ditte-
				THE STATE
Tympatonus	Littorinidae/	Imoron (common	Slightly thicker, elongated, conical	
fuscatus Var.	Tympanotonus	name: Isemi	shape shell. Narrower than T. fuscatus.	WISED BRITEIN
radula		periwinkle)	Have sharp apex and more regular	and the second s
			prominent angular whorls.	
Pachymelania	Pachychilidae/	*Bebefangala	Conical-shaped shells and smoother	4 4 4
aurita	Pachymelania	(Buahari)	surfaces often lack prominent ridges.	
	-	periwinkle	Tougher shell and darker coloration. It	
			is less common on the open mudflats.	
*Not	eaten by the indig	enous people of the s	tudy area, usually discarded when harveste	d

Tuble 11 hot photogreat characteristics of the Terroritine Species Shens freeessed in the study file	Table 1: Morphological Characteristics of the Periwinkle Species Shells Accessed in the stud	y Area
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The results of the proximate composition of the three periwinkle species' shells are presented in Table 2. The shell moisture content did not varied significantly (p ≥ 0.05) among the periwinkle species, though *Tympanotonus radula* (Imoron) exhibits the highest moisture content at 41.32 ± 6.96 , while *Tympanotonus fuscatus* (Emoru) had the lowest at 35.51 ± 1.31 . The dry matter percentage, which inversely correlates with moisture content, was prevalent and lowest in *T. fuscatus* 61.87 ± 0.63 and 35.51 ± 1.31 respectively. The low moisture and high dry matter levels in the species indicate a potential for soil amendments, where the goal is to enhance soil nutrient profile. Previous work of

Drewnowski and Almiron-Roig (2010) asserted that substances with low moisture content may be preferable for biofortification because the nutrients and minerals are more concentrated. The highest crude fiber content in the species was recorded in *T. fuscatus* at 4.68 \pm 0.41, followed closely by *T. radula at* 4.18 \pm 1.10, while *P. aurita* had a significantly low (P \leq 0.05) value of 2.29 \pm 0.11. The high fiber content in *T. fuscatus* and *T. radula* suggests they could contribute positively to soil structure when used as organic amendments, supporting the previous assertion of Brady and Weil (2016) and Bardgett and van der Putten (2014), who opined that fiber aids in improving soil aeration and water retention.

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Parameter	T. fuscatus	T. radula	P. aurita	F-value	p-value
Moisture Content (%)	35.51 ± 1.31	41.32 ± 6.96	35.78 ± 1.24	2.54	0.157
Dry Matter (%)	61.87 ± 0.63	54.08 ± 7.00	57.13 ± 2.63	1.92	0.251
Crude Fiber (%)	4.46 ± 0.41	4.18 ± 1.10	2.29 ± 0.11	9.64	0.015
Ether Extract (%)	0.64 ± 0.04	0.35 ± 0.02	0.97 ± 0.06	104.00	< 0.001
Ash Content (%)	5.44 ± 1.14	3.09 ± 0.31	2.71 ± 0.29	7.52	0.024
Crude Protein (%)	25.00 ± 1.09	21.46 ± 3.26	18.27 ± 0.35	7.74	0.023
Note: the f	igures represen	t mean values ±	SD of triplicate	e values	

Ash content reflects the total mineral constituents in the shells. The highest ash content occurred in *T. fuscatus* (5.44 ± 1.14), suggesting a greater overall mineral reserve when compared (P \leq 0.05) to other species, which ranged from 3.09 ± 0.31 in *T. radula* to 2.71 ± 0.29 in *P. aurita*. The high ash content indicates high essential mineral content, which may be beneficial for biofortification and soil amendment aimed at improving the mineral density of crops (Ravindran and

Blair, 1992; López *et al.*, 2004). Protein content in the species varied significantly (P \leq 0.05), with *T. fuscatus* having the highest crude protein (25.00 ± 1.09), followed by *T. radula* (21.46 ± 3.26), while *P. aurita* contained the least (18.27 ± 0.35). High crude protein indicates that the periwinkle shells can be a valuable source of amino acids, the building blocks of proteins, of which FAO (2004) asserted could be used to develop nutrient-dense

biofortified crops aimed at improving both macro- and micronutrient intake.

The mineral composition of periwinkle shells is presented in Table 3. T fuscatus exhibited significantly higher concentrations (p < 0.05) of iodine (6.90 ± 1.05 mg/kg), calcium (15.21 ± 0.89 mg/kg), potassium (9.40 ± 0.46 mg/kg), and magnesium (8.45 ± 0.44 mg/kg) compared to T. radula and P. aurita. However, phosphorus concentration in T. radula (3.86 ± 0.18 mg/kg) was significantly different (p < 0.05) from that of T. fuscatus and P. aurita (Table 3). The high mineral content in the periwinkle shells is likely influenced by environmental factors, dietary intake, and biological mechanisms (Richard and Prezant, 2021). These findings align with Frederic *et al.*, (2012), who reported that swamp water contains dissolved calcium ions (Ca²⁺), which Mollusca actively extract for shell formation. Additionally, Magee (1993), Atkinson *et al.*, (2023), and Rygało-Galewska *et al.*, (2023) suggest that swamp snails obtain calcium from detritus, sediments, and algae, further contributing to mineral accumulation in their shells.

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Element	T. fuscatus	T. radula	P. aurita	F-stat.	p-value
Iodine	6.90 ± 1.05	4.21 ± 0.59	4.93 ± 1.12	6.43	0.032
Selenium	0.21 ± 0.02	0.17 ± 0.01	0.22 ± 0.04	3.75	0.085
Phosphorus	3.33 ± 0.04	3.86 ± 0.18	2.11 ± 0.68	14.48	0.005
Calcium	15.21 ± 0.89	9.42 ± 0.37	11.83 ± 0.46	67.48	0.00008
Potassium	9.40 ± 0.46	8.07 ± 0.15	5.20 ± 0.20	152.63	0.00001
Magnesium	8.45 ± 0.44	6.57 ± 0.42	7.85 ± 0.38	16.34	0.0037

Table 3: The Results of the Mineral Composition (in mg/kg) of Periwinkle Species

The mineral composition of periwinkle shells is intrinsically linked to their swamp habitat, characterized by a mineral-rich ecosystem and unique endogenous factors. Davies et al., (2016) emphasized that environmental influences such as diet, water salinity, and surrounding geological features play a crucial role in shell mineralization. Iodine concentrations across the studied species ranged from 4.21 \pm 0.59 to 6.90 \pm 1.05 mg/kg, with T. fuscatus exhibiting the highest levels. This suggests its potential as a natural amendment for iodine-deficient soils. Supporting this, Fuge (2005), Medrano-Macías et al., (2016), and Duborska et al., (2020) noted that iodine-rich biofortified materials can enhance iodine availability in crops and animal feed, addressing dietary deficiencies. Selenium levels were comparable across species (Table 3), whereas phosphorus, calcium, potassium, and magnesium concentrations remained significantly higher ($p \le 0.05$) in T. fuscatus than in T. radula and P. aurita. These findings asserted the potential of periwinkle shells, particularly T. fuscatus, as a valuable source of essential minerals.

IN CONCLUSION

The findings of this study reveals the values of the indigenous periwinkles as a sustainable resource for enhancing crop biofortification strategies Beyond their ecological significance, these shells can be repurposed as a natural fertilizer to enhance soil fertility and boost crop mineral content in nutrient-deficient regions (Fischer *et al.*, 2018). This practice will not only support farmers in improving both crop and livestock nutrition, but also contributes to food security (Ugwumba, 2018; Zhao *et al.*, 2019). Moreover, incorporating shells as organic amendments can strengthen local agricultural systems, increasing resilience to soil depletion and climate variability (Shahzad *et al.*, 2020).

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