Determination of Nutrient Composition and Antibacterial Activity 0f Snail (*Archachatina marginata*) Slime

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Abstract
Snail slime has been perceived to make snail flesh unattractive and is usually removed from the snail meat to be discarded as waste during home processing. This study evaluated the slime of Achatina marginata snail for its proximate and mineral compositions and antibacterial properties following established techniques. The standard procedures of AOAC (1990) were followed to analyze the proximate composition and mineral analysis while agar well diffusion was done to determine the snail slime's antibacterial properties against Bacillus subtilis and Salmonella typhi.The analysis's findings showed that the slime's proximate composition (%) was moisture 88.15 ± 0.07 %, ash 1.385 ± 0.04%, crude fat 0.12 ± 0.01%, crude protein 1.305 ± 0.02% and carbohydrate contents 6.29 ± 0.08%. According to the results of the mineral analysis, the sodium content of the slime was highest (78.3 mg/100 g) followed by calcium (66.25 mg/100 g) and then phosphorus (32.61 mg/100 g) and potassium (27.18 mg/100g). Magnesium was moderately present in the slime (13.72mg/100g) while iron and zinc were present at low amounts (1.83 mg/100 g and 1.07 mg/100 g respectively).The result of the antibacterial activity revealed that the slime exhibited a higher inhibitory power against Bacillus subtilis and Salmonella typhi with zones of inhibition of 24.83 mm and 19.00 mm respectively when compared to levofloxacin with zones of inhibition of 23.17 mm and 16.33 mm against Bacillus subtilis and Salmonella typhi respectively. This study therefore encourages the use of slime from Archachatina marginata for nutrition and medicinal preparations.

Keywords; *Archachatina Marginata*, slime, Proximate analysis, Mineral content, Antibacterial activity

1. INTRODUCTION

For many generations, humans have eaten snails as food, and it is currently believed that they have a very high level of proteins (Hayes & Mora, 2021; Fatai, 2018). One of the factors contributing to protein-energy malnutrition in Africa is the rising expense of other animal protein sources including cattle, hog, poultry, and fish (de Vries-Ten Have et al., 2020). Snails are therefore being projected as an alternative source of proteins since they are cheaper to rear (Hayes & Mora, 2021).

Snails are invertebrate organisms that belong to the phylum Mollusca, where they constitute the biggest group (Djikeng et al., 2022). They can be found all around the world, but are most prevalent in fresh water, seas, and other wet environments (Olkeba et al., 2020). The shell and body of snails are fundamentally separated into two sections, with the shell serving as a protective case and the body being utilized to make meat (Olusi et al., 2021). Additionally, a snail produces enormous amount of slippery material referred to as snail slime, which it disperses along its line of travel. It uses this slime to repair its shell and skin when they become injured, keep moisture from evaporating, aid in fluid movement, and shield the body from mechanical harm (Danladi et al., 2020). However, this slime has been perceived to make snail flesh unattractive and so is usually removed from the snail during processing (Djikeng et al., 2022). New researches surfacing online has revealed that snail's slime is now being explored for use in the cosmetic industry and for drug production in pharmaceutical companies (Cristiano and Guagni, 2022; Gugliandolo et al., 2021; McDermott et al., 2021; Murgia, Meuli, Mannu and Casu1, 2021).The most prevalent specie of snails on land is *Archachatina marginata* which is thought to be a member of the family of African Giant Land Snails (Ejidike & Adewuyi, 2018).

In the medical world,typhoid fever, which is brought on by *Salmonella typhi*, continues to be a serious global public health issue that leads to disabilities and even deaths despite tremendous research and medical improvements (Ashurst, Truong and Woodbury, 2022; Akram et al., 2020). *Bacillus subtilis* is another microorganism asssociated with a lot of medical concerns and this concern is underlined by the fact that *Bacillus spp*. in a number of commercially available probiotic feed supplements for both humans and animals have been shown to be resistant to several antibiotics, such as chloramphenicol, tetracycline, erythromycin, lincomycin, penicillin, and streptomycinm (Adimpong *et al.*, 2021). Recently, *Bacillus subtilis* has been implicated as the causative organism of sepsis; a serious neonatal infections (Lampropoulois et al., 2021). Apart from the problem of microorganisms developing resistance to conventional antibiotics drugs due to misuse and prolonged usage, these drugs have also been reported to be associated with several side effects (Mohsen, Dickinson and Somayaji, 2020; Pancu *et al.*, 2021).There is therefore a continuous need to search for novel antibacterial agents especially of natural origin that will be effective against these multidrug resistant strains of bacteria and be less toxic to human (Terreni, Taccani & Pregnolato, 2021).According to Cilia & Fratini, 2018, very few studies are available on the antimicrobial properties of the mucus of terrestrial molluscs. This study therefore evaluated the proximate and mineral component as well as antibacterial property of *Archachatina marginata*'s slime against common pathogenic organisms (*Bacillus subtilis and Salmonella typhi*) in order to validate its use for nutrition and medicinal preparations.

2. Materials and Method

Archachatina marginata (30 pieces) were collected from a snail farm in Iju Sango-Ota and verified by a zoologist. Analytical grade reagents were used all through the experiment.

Plate 1-*Archachatina marginata*

2.1 Extraction/Preparation of Snail Slime

The shells of the snails were cracked open at the apex after being cleaned off with clean water to remove any dirt or dust. The fleshy body was then separated from the shell by a spirally coiled rod. The bulky body was placed in a bowl with little water while the mucus coating (slime) was gently scraped off the fleshy areas. The snail slime was extracted, filtered, marked as 100% extract, and kept at 4°C for future study (Danladi et al., 2020).

2.2 Proximate analysis

Following standard AOAC methods (1990) as described by Danladi et al., 2020, proximate analysis (determination crude protein, ash, crude fat, carbohydrate and moisture contents) was done on the snail slime.

2.3 Mineral determination

The amount of trace elements (iron and zinc), phosphorus, sodium, calcium, potassium, and magnesium in the snail slime was measured using an atomic absorption spectrophotometer (Varian SpectrAA 220, USA) as described by Lalitha Sree and Vijayalakshmi (2018).

2.4 Assessment of Antibacterial Property

2.4.1 *Test Pathogens*

The test organisms (*Bacillus subtilis* and *Salmonella typhi*) employed in this study were clinical isolates obtained from the Microbiology Laboratory of University of Lagos.

2.2.2 *Antibacterial Assay*

The antibacterial effectiveness of snail slime was assessed using the Agar well diffusion method. Briefly stated, warm agar was seeded with 1 ml of each calibrated test organism

(*Bacillus spp.* and *Salmonella typhi*) and thoroughly mixed using the roll-palm method before pour plating. A cork borer was used to drill wells 10 mm in diameter into solidified nutrient agar plates. Levofloxacin (6.25 µg/ml) and 150 µl of 100 % snail slime were added to the wells and left to stand for several hours to allow for optimal diffusion. The plates were then examined for zones of inhibition after 24 hours of incubation at 37 ºC (Abhay and Rupa, 2016).

3. RESULTS

3.1 Nutrient Composition

The result of the nutrient composition of snail slime is reported in table 1 (Proximate analysis) and table 2 (Mineral content). For the proximate analysis, it was observed that the snail had very high moisture content but low in fat.

Table 1: Proximate analysis of snail slime (*A. marginata*)

Proximate compounds	Percentage composition (%)
Moisture	88.15±0.07
Crude protein	1.305±0.02
Crude fat	0.12±0.01
Ash	1.385±0.04
Carbohydrate	6.29±0.08
Crude fibre	2.75±0.1

Table 2: Mineral content of the snail slime (*A. marginata*)

Minerals	Compositions (mg/100g)
Sodium	78.36
Magnesium	13.72
Phosphorus	32.61
Calcium	66.25
Zinc	1.07
Iron	1.83
Potassium	27.18

The result of the mineral content revealed that the snail slime is high in sodium but low in Zinc and Iron.

3.3 Antibacterial Activity

The antibacterial activity of snail slime and levofloxacin against *Bacillus subtilis* and *Salmonella typhi* is shown in Figure 1 below;

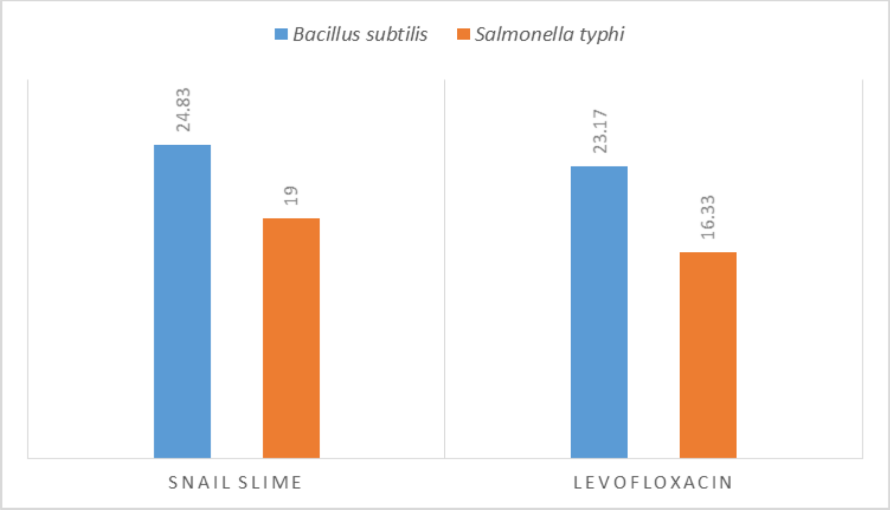

Figure 1: Inhibition Zone Diameter (mm) of Snail Slime and Levofloxacin against *Bacillus subtilis* and *Salmonella typhi*

4. Discussion

This study was done to determine the nutritive value and antibacterial property of *A. marginata's* slime commonly treated as a waste during snail processing. The result of the proximate composition of *A. marginata* slime (table 1) revealed 88.15± 0.07 % moisture content, 1.385 ± 0.04 % ash, 0.12 ± 0.01 % crude fat, 1.305 ± 0.02 % crude protein and 6.29±0.08% carbohydrate contents. The moisture content of 88.15 ± 0.07 % found in the snail slime fits within the range of 73.67-99.20 % that Eneji et al. (2008) reported for snail flesh. An indicator that the slime may be a rich source of protein and minerals that are helpful for humans is the comparatively high level of ash and crude protein content. The low crude fat content of the snail slime suggested that it could be consumed by hypertensive patients (Schwingshackl et al., 2017). However, the snail slime was notably rich in carbohydrates. This is contrary to the low carbohydrate level of snail slime reported by Danladi et al., 2020. The results of the mineral analysis of the slime (table 2) revealed that it has high content of Na (78.3 mg/100g) followed by calcium (66.25 mg/100g) and then phosphorus (32.61 mg/100g) and potassium (27.18 mg/100g). The value of magnesium (13.72 mg/100g) was moderately present in the slime while sodium iron (1.83 mg/100g) and zinc (1.07 mg/100g) were present at low amounts. This finding indicates that *A.marginata*'s slime contains inorganic elements required for living things to function.. However, the result of the mineral composition in this study is higher than the mineral composition of snail (*A.marginata*) slime reported by Danladi et al., 2020 but similar to that of *A.marginata*'s meat fed rumen content inclusion (T1) reported by Raimi, 2019. This could be because mineral content of snails changes by location, feeding habits, seasons and biological cycles (Young & Jackson, 2019; Cagiltay et al., 2011). Also, the result of the antibacterial activity (figure 1), revealed that the slime extracted from *Archachatina marginata* showed a positive antibacterial activity against the test pathogens used. The snail slime showed higher antibacterial activity against *Bacillus subtilis* and *Salmonella typhi* when compared to that of levofloxacin. This result confirms the report of Cilia and Fratini (2018) that snail slime possesses antibacterial properties.

5 Conclusion

The result of this study showed that snail (*A. marginata*) slime contains appreciable level of proximate compounds and minerals that are readily available and necessary for the functioning of living things. The result also revealed that the snail slime possesses antibacterial properties against bacterial pathogens such as *Bacillus subtilis* and *Salmonella typhi* and can be used in the treatment of diseases caused by these microorganisms. The use of snail slime for medicinal preparations and especially for nutrition along side snail meat is therefore encouraged.

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