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## Comparative Evaluation of the Physicochemical Composition of Chitosan from Commercial and Biowaste Sources

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### Abstract

Chitosan which is a derivative of chitin after the process of deacetylation has multiple commercial and medical uses based on its degree of deacetylation. This research aims to extract chitosan from snail shell, a biowaste using groundnut husk, an agrowaste as agent of deproteination and deacetylation (CGH) and compare the chitosan quality with commercial chitosan (CC) after examination of physicochemical parameters using their standard methods. The results show that moisture, ash, protein, fat contents varied from 8.94 – 9.65 %, 1.60 – 1.73 %, 1.79 – 1.84 %, 0.39 – 0.72 % respectively. Chitosan yield of 61.93 % was obtained for CGH and both chitosan products were soluble in 1 % acetic acid. Water binding capacity (WBC), fat binding capacity (FBC) and degree of deacetylation (DDA) varied from 775 – 820, 113.70, 1.33.70 - 177.73 and 71.56 - 83.51 respectively. Based on these characteristics, CGH can achieve chitosan standard quality for industrial application by performing traditional methods of deproteination, demineralization and deacetylation using groundnut husk as agent of deproteination and deacetylation.

**Keywords:** Chitosan, snail shell, groundnut husk, deproteination, deacetylation

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### 1. Introduction

Chitosan, a natural polysaccharide derived from chitin, has emerged as a promising material for applications in agriculture, nutraceutical, pharmaceutical, engineering, medical, biomedical and even in wastewater treatment due to its unique properties such as biodegradable, non-toxic, renewability, biocompatibility etc. [1 – 7]. Nanochitosan, which is obtained by reducing the particle size of chitosan to the nanometer scale, exhibits enhanced surface area and reactivity, resulting in improved efficiency more than the parent chitosan [8 – 12].

Chitosan synthesis involves some major processes such as deproteination, demineralization and deacetylation [13, 14]. These processes often employ harsh chemicals such as strong acids and alkalis, which may lead to the production of heterogeneous chitosan and as well generate hazardous waste.

Agricultural waste materials contain organic compounds like cellulose, lignin, and hemicellulose that can be hydrolyzed and transformed into alkali solutions [15, 16]. Consequently, alkali solutions extracted from these waste materials may be used as deproteination and deacetylation agents in the synthesis of chitosan from biowastes. Nigeria may be able to achieve some of its sustainable development goals by using alkali from agricultural waste to produce homogenous chitosan, which would boost the country's economy and reduce pollution [17, 18]. Some scholars have worked in this emerging area. For example, Okafor et al. [19] carried out a comparative study of chitosan-silver nanocomposites from commercial and biowaste sources. Mohanasrinivasan et al. [20] studied heavy



metal removal efficiency and antibacterial activity of chitosan prepared from shrimp shell waste. Okafor et al. [21] investigated the potentiality of diethylamine as agent of deproteination and deacetylation in the extraction of chitosan from *Scylla serrata* shell. Okafor et al. [22] synthesized nanochitosan from *Cambarus bartonii* waste which was utilized in the removal of polycyclic aromatic hydrocarbons from surface water. Okafor and Obiefuna [23] compared the physicochemical properties of chitosan synthesized from periwinkle shell using alkali from *Moringa oleifera* petiole as an agent of deproteination and deacetylation and commercial chitosan. However, there is little or no literature on the use of naturally sourced alkali from groundnut (*Arachis hypogea*) husk as agent of deproteination and deacetylation in chitosan extraction from snail shell, and hence, this study.

Therefore, this work aims to extract chitosan from *A. achatina* shell utilizing naturally sourced alkali from *A. hypogea* husk as deproteination and deacetylation agent and compare its physicochemical properties with those of commercial chitosan.

## 2. Materials and Methods

### Sample collection and preparation:

Fresh snail shells were procured from Nnamdi Azikiwe University Zoological Garden while groundnut husk was collected from groundnut refuse dumpsite at Eke Awka, Anambra State. Commercial chitosan was supplied by MarkNature, United State of America (USA). The snail shells were washed thoroughly with distilled water to remove dirt and impurities, dried in an oven to constant weight at a temperature of 40°C and ground to a fine powder and stored in a clean dry plastic bag at room temperature for further use. A 200 g of the dried groundnut husk was placed in open aluminum heating pan and heated until fully ignited and reduced to ash. The finely powdered ashed groundnut husk were subjected to alkaline hydrolysis following protocols used by Okafor et al. [23].

### Extraction of Chitosan and Physicochemical Analysis

Method of extraction described by Okafor et al. [23] was adopted in the extraction of chitin which was followed by the deacetylation of the chitin employing the protocol employed by Novikov et al. [24] with modifications. Physicochemical parameters including moisture, ash, protein, and fat contents of the chitosan particles were conducted following the methods described by Ganogpichayagrai and Suksaard [25]. Percentage yield, solubility in 1 % acetic acid, water and fat binding capacities and degree of deacetylation were determined as described by El-Araby et al. [26]

## 3. Results and Discussion

### Physicochemical Composition

Commercial chitosan typically has a moisture content of less than 10%, according to numerous studies that have confirmed the hygroscopic nature of chitosan over the years [4, 27, 28]. Remarkably, the current investigation produced chitosan with the desired moisture content of CGH (9.65) and CC (8.94). The study conducted by Anand et al. [29] found that the chitosan moisture content from shell wastes of crab, squilla and commercial chitosan was 0.35, 0.41, and 0.52%, respectively. Furthermore, chitosan derived from crab, fish scale, and shrimp shells has a moisture content of 0.0004%, 0.009%, and 0.0004%, respectively, according to Kumari et al. [30]. All these findings were significantly less than those of the current investigation. Therefore, this investigation suggests that the high moisture content of *A. hypogea* husk may be the reason why CGH had a higher moisture content than CC (Table 1). The moisture content of CC and CGH, however, compared favourably.

One important measure of the efficiency of the removal of inorganic materials during the demineralization stage is the amount of ash in chitosan [31]. The demineralization process is less successful when higher ash content is obtained, and vice versa [32]. Since CC's ash content (1.73 %) is somewhat higher than CGH's (1.60%), there is no discernible difference between the two in terms of ash content. This result contrasts with that of Adekanmi et al. [33], who reported ash contents from crayfish shells ranging from 0.12 to 0.86 % and 0.19 to 0.51 %. In another development, ash content of chitosan from fish scale, crab and shrimp shell were reported as 1 %, 2.5 % and 0.03 % respectively [30].



**Table 1:** Physicochemical Composition of the chitosan particles

Parameter	CGH	CC
Moisture (%)	9.65	8.94
Ash (%)	1.60	1.73
Protein (%)	1.84	1.79
Fat (%)	0.39	0.72
Chitosan yield (%)	61.93	-
Solubility in acetic acid	++	++
Water binding capacity (%)	775	820
Fat binding capacity (%)	133.70	177.73
Degree of deacetylation (%)	71.56	*83.51

CGH = chitosan extracted using groundnut husk alkali, CC = commercial chitosan, \*manufacturer's information

From Table 1, the protein content of CGH (1.84%) is higher than that of CC (1.79%). Due to the high degree of deacetylation, the protein content of the chitosan samples was within the same range following the deacetylation of the chitin. This demonstrates that the alkali from the husk of *A. hypogea* is an effective deacetylating and deproteinating agent that may be used in place of commercial caustic alkali. Okafor et al. [19] found that the protein content of chitosan recovered from snail shells employing sodium hydroxide as a deproteination and deacetylation agent was 2.80 % which is higher than the values obtained in the current study.

The extracted chitosan's fat content shows that CGH (0.39 %) < CC (0.72 %). The results were comparatively higher than those of Byun et al. [34], who reported a fat content of 0.03% from crab shell. The current study's overall chitosan yield (61.93 %) was significantly higher than the 52.2 % that was extracted from *Litopenaeus vannamei* [35] and the 17 % that was obtained from *Catharsius molossus* L. residue [36]. Additionally, the chitosan yield in this study was found to be higher than 5.89% obtained from freshwater crab shell waste, *Potamon algeriense* [37].

According to el Knidri et al. [38], chitosan has D-glucosamine units that contain mobile amino groups that acquire positively charged ions. These units are responsible for the biopolymer's important solubility and antibacterial characteristics. In chitosan application, the deprotonation of the amino groups in an acidic solution is vital. In the current study, all the chitosan products were soluble in a solution of 1% acetic acid after 30 minutes under the same circumstances. Chitosan's amino groups are protonated ( $-NH_3^+$ ) in acidic solution and therefore, the electrostatic repulsion between the chitosan chains is reduced, increasing their solubility in acetic acid and forming soluble salts. The amount of chitin that is deacetylated during the chitosan extraction process determines the solubility of the chitosan as solubility increases with the degree of deacetylation [39]. This argument was further supported by Mohan et al. [40], who also noted that high chitosan solubility indicates the existence of free amine groups that are readily deprotonated in the presence of aqueous solutions.

According to Luo et al. [41] and Rasweefali et al. [42], a hydrophilic substance's water binding capacity (WBC) is its ability to associate favourably with water, but its fat binding capacity (FBC) is the amount of absorbed oil per unit weight of the material. Research has indicated that the degree of demineralization, deproteinization, and deacetylation processes among chitosan has a significant impact on its WBC [13, 14]. This suggests that in order to accomplish the desired WBC, the type of acid and alkali used, along with their concentration and ratio of mixing with the solute, must be carefully determined. Commercial chitosan has been reported with WBC of 1078 (Okafor et al., 2024) and 812.67 [43] having compared with 1095.66, 1270, and 1161.67 % recorded by another study [41]. From Table 1, WBC of CGH (775%) is lower than that of CC (820 %) but in good comparison. The FTB of CGH (133.70 %) is also lower than that of CC (177.73 %) but somewhat in unfavourable comparison. In addition, Zaghib, et al. [44], reported chitosan with WBC of 652 which was less than those obtained in the present work.

The degree to which acetyl chains are removed from the chitin molecular chains and the accompanying addition of amino groups is known as the degree of deacetylation (DDA) of chitosan. According to Rasweefali et al. [14], chitosan is only considered beneficial and referred to as such when its DDA is greater than 70 %. It is evidence from Table 1, that CC (83.51 %) has a higher degree of deacetylation than CGH (80.10 %). Kucukgulmez et al. [45] and



Gbenebor et al. [46] opined that the degree of deacetylation is a crucial factor when considering chitosan for application in particular industries because it affects characteristics like solubility, viscosity, ion-exchange capacity, flocculation ability, tensile strength, ability to chelate metal ions, immune-adjuvant activity, and reaction with amino groups.

#### 4. Conclusion

Chitosan was successfully extracted from snail shell, a biowaste, using alkali extracted from groundnut husk. The physicochemical properties of the two chitosan products compared favourably. As a result, chitosan synthesized from snail shell using alkali from groundnut husk can be utilized for a variety of purposes, including wastewater treatment, biomedical applications, and other industrial uses like the commercial chitosan.

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