

## Characterization of Starch Extracted from Pretreated Whole-Grain Bambara Groundnut

Adamu Hamsatu Sani<sup>1</sup>, Akubor P. I<sup>2</sup>, Abubakar Ummulkhairu<sup>3</sup>

<sup>1</sup>Taraba State University, Nigeria; <sup>2,3</sup>Federal University Wukari Taraba State, Nigeria  
hamsatu.adamu@tsuniversity.edu.ng; akuborpeter@gmail.com

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### Abstract

This study characterized starch extracted from whole-grain and dehulled Bambara groundnut (*Vigna subterranea* L.) pretreated with *Saccharomyces cerevisiae* to determine the effects of dehulling and fermentation on starch yield, composition, functionality, pasting behavior, digestibility, and morphology. Whole-grain Bambara groundnut seeds were soaked for 24 h, manually dehulled where applicable, oven-dried at 40°C to constant weight, and fermented prior to starch extraction. The extracted starches were analyzed using standard methods, and statistical significance was determined at  $p < 0.05$ . The results showed that dehulling followed by fermentation significantly increased starch yield from 34.88% to 39.66%. Proximate analysis indicated moisture content of 5.48–6.01%, crude protein of 0.23–0.77%, ash of 0.50–0.61%, crude fat of 0.52–0.96%, fiber of 0.02–0.90%, and carbohydrate content of 90.92–93.20%, suggesting improved starch purity after pretreatment. Functional properties were also enhanced, with water absorption capacity ranging from 2.43 to 1.50 g/100 g, oil absorption capacity from 2.00 to 1.75 g/100 g, loose density from 0.45 to 0.68 g/ml, bulk density from 0.75 to 0.95 g/ml, swelling capacity from 88.68% to 67.45%, dispersibility from 79.00% to 59.00%, and wettability from 30.00 to 7.50 s. Pasting properties

improved, with peak viscosity of 174.37–215.65 cP, final viscosity of 223.37–226.42 cP, setback of 67.60–74.48 cP, trough of 155.73–188.29 cP, pasting temperature of 68.30–72.12°C, and pasting time of 3.17–7.17 min. Amylose content increased to 20.50–26.16%, whereas amylopectin decreased to 73.84–79.50% following fermentation. In addition, total starch (61.46–66.75%) and rapidly digestible starch (50.05–63.28%) increased, while resistant starch (13.91–21.80%) and slowly digestible starch (23.54–28.44%) decreased after pretreatment. Scanning electron microscopy further revealed oval, spherical, ring-like, elliptical, and irregular granules with slight surface modification. Overall, the study demonstrates that dehulling and fermentation improve the yield, purity, functional performance, pasting characteristics, and digestibility profile of Bambara groundnut starch, highlighting its potential for food and industrial applications.

**Keywords:** Bambara Groundnut Starch; *Vigna subterranea* L.; *Saccharomyces cerevisiae*; Functional Properties; Digestibility Profile

## INTRODUCTION

Starch is a complex carbohydrate composed of numerous glucose units joined by glycosidic bonds [1]. Its chemical structure consists primarily of amylose and amylopectin [2]. Amylose is a linear polymer of glucose molecules linked by  $\alpha$ -1,4 glycosidic bonds and contributes about 20–30% of total starch structure [3]. In contrast, amylopectin is a highly branched polymer containing  $\alpha$ -1,4 linkages in the linear chains and  $\alpha$ -1,6 linkages at the branch points, accounting for approximately 70–80% of starch composition [4]. This structural arrangement imparts distinct physicochemical properties such as gelatinization, viscosity, swelling, and retrogradation behavior [5]. Owing to these characteristics, starch is widely used in food systems as a thickening, gelling, and stabilizing agent [6], and serves as a key ingredient in gluten-free formulations [7].

Conventionally, starch is derived from cereals, roots, and tubers [2]. However, increasing interest in sustainable and alternative raw materials has led to exploration of non-conventional sources such as legumes [2]. Legume seeds reportedly contain 35–46% starch and are considered promising substitutes for conventional starch sources [8]. Legumes are naturally gluten-free and environmentally resilient crops [2]. Compared to cereal starches, legume starches often exhibit higher amylose content, higher gelatinization temperatures, greater degree of polymerization, and more viscous pasting characteristics

[9]. These properties suggest potential technological and nutritional advantages. Starch extraction and characterization from legumes such as Bambara groundnut, faba bean, and common vetch have been documented [8], [2], [10].

Bambara groundnut (*Vigna subterranea* L.) is an extremely hardy leguminous crop widely cultivated in sub-Saharan Africa and serves as a major source of dietary energy and protein [11]. The seeds occur in various colours, sizes, and shapes and are used in the preparation of diverse food products [8]. The amylose content of Bambara groundnut starch ranges between 18–27%, while amylopectin constitutes approximately 73–82% [2]. Morphologically, Bambara groundnut starch granules are typically oval or round with semi-crystalline structures comprising crystalline and amorphous regions, which influence functionality and granular behavior [2], [12]. Despite its potential, utilization of Bambara groundnut starch in industrial applications remains limited.

One major challenge associated with legume starch extraction is achieving high purity, as native starches often contain residual proteins, lipids, fibers, and anti-nutritional compounds [13]. These impurities can affect gelatinization, viscosity, swelling, and overall functional behavior [14]. Bambara groundnut, like many legumes, contains phytates, tannins, trypsin inhibitors, and oxalates [15], which may reduce mineral bioavailability and interfere with starch functionality. Furthermore, legume starches tend to have relatively higher resistant starch content compared to some cereal starches [16], influencing digestibility and structural behavior. Therefore, pretreatment prior to starch extraction is considered essential for improving yield and modifying physicochemical characteristics.

Fermentation using microorganisms such as *Saccharomyces cerevisiae* has been reported to degrade phytates, reduce tannins, and inactivate trypsin inhibitors, thereby improving nutritional quality [17]. Microbial enzymatic activities can modify starch structure, influencing gelatinization, solubility, and viscosity while potentially enhancing extraction efficiency [18], [19]. *Saccharomyces cerevisiae* produces enzymes such as amylases capable of hydrolyzing polysaccharides and facilitating starch release [20]. Dehulling, which involves removal of the seed coat, reduces non-starchy components such as fiber, pigments, and phenolic compounds that may interfere with starch purity and yield [21]. Studies have also reported improved physicochemical and structural properties of starch extracted from whole-grain cereals relative to dehulled grains [22], [23].

Although previous investigations have examined starch extraction from Bambara groundnut and other legumes, comprehensive information on the combined effect of dehulling and fermentation with *Saccharomyces cerevisiae* on starch yield, proximate composition, amylose–amylopectin distribution, digestibility fractions, functional properties, and morphological characteristics remains limited. Information on starch extracted from modified whole-grain legumes is particularly scarce. Therefore, this study aimed to characterize starch extracted from whole-grain and dehulled Bambara groundnut (*Vigna subterranea* L.) pretreated with *Saccharomyces cerevisiae*, with emphasis on its physicochemical, functional, digestibility, and structural properties.

## MATERIALS AND METHODS

### Materials

Whole-grain Bambara groundnuts (*Vigna subterranea* (L.) Verdc) and *Saccharomyces cerevisiae* (baker’s yeast) were purchased from Wukari New Market, Taraba State, Nigeria. Sodium hypochlorite solution, distilled water, Yeast Extract Peptone Dextrose broth (YEPD), citric acid, NaOH, and HCl were obtained from the laboratories in the Department of Food Science and Technology, Federal University Wukari. The chemicals used were of analytical grade. The samples were prepared and analyses were carried out in the laboratories of the Department of Food Science and Technology.

### Methods

#### Experimental Design

The experimental design of the work is shown in Table 1. The study consisted of four samples with two treatments. Whole-grain BGN and dehulled BGN were pretreated with *Saccharomyces cerevisiae*, while whole-grain BGN and dehulled BGN without pretreatment served as controls.

**Table 1. Experimental Design for BGN pretreatments**

Microorganisms	Bambara groundnut
Untreatment (control)	Dehulled BGN Whole-Grain BGN
<i>Saccharomyces cerevisiae</i>	Dehulled BGN Whole-Grain BGN

### Preparation of Dehulled Bambara Groundnut without Pretreatment

The dehulled Bambara groundnuts were prepared in the laboratory of the Department of Food Science and Technology, Federal University Wukari, Taraba State, Nigeria. It was done according to the method described by Yahaya *et al.* [24] (Figure 1). The clean matured Bambara groundnut were sorted manually by hand-picking and cleaned properly to separate unwanted materials. The cleaned matured BGN grains (1kg) were soaked in 4 L of water for 24 h to loosen the seed coat and to make dehulling easier. Dehulling was done manually with hand, and the seed coats were removed. The dehulled BGN grains were oven dried at 40 °C to constant moisture. The dried dehulled grains were kept in high density polyethylene bags prior to use.

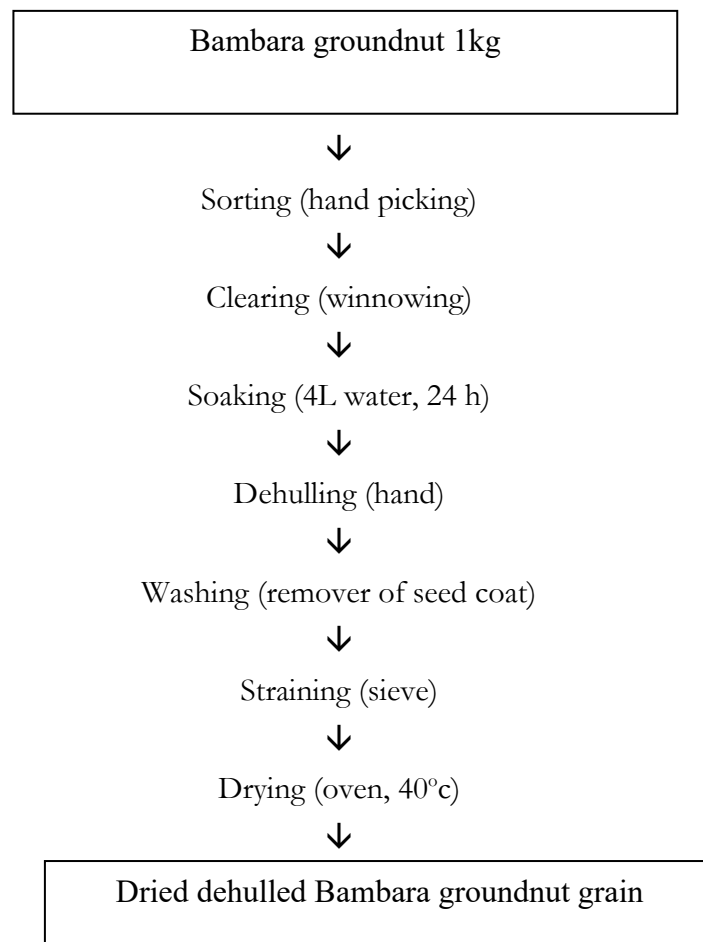
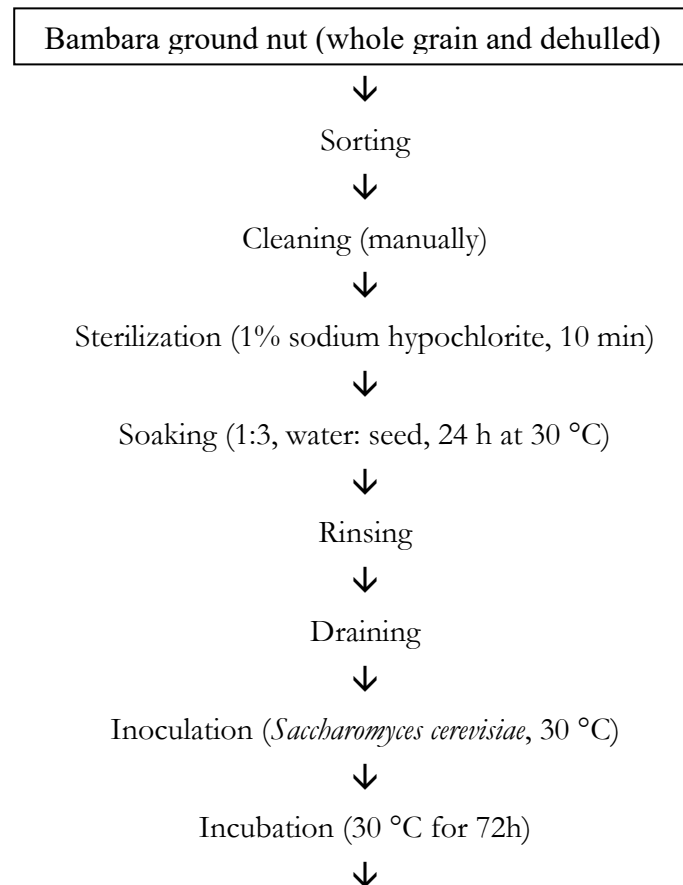


Figure 1. Dehulled Bambara groundnut [24]

## **Pretreatment of Dehulled and Whole-Grain Bambara Groundnuts with *Saccharomyces cerevisiae***

Pretreatment was conducted according to Olaniran et al. [25] with slight modifications. Bambara groundnut grains (1 kg) were sorted and sterilized in 1% sodium hypochlorite for 10 min, then rinsed thoroughly with sterile distilled water. The grains were soaked in distilled water at a 1:3 (w/v) ratio for 24 h at 30°C.

*Saccharomyces cerevisiae* (30 g) was cultured in YEPD broth at 30°C for 24 h to achieve approximately  $1 \times 10^7$  CFU/mL. Optical density was measured at 600 nm to standardize yeast concentration. Hydrated grains were inoculated with yeast to achieve 8% (v/v) concentration and pH was adjusted to 4.5 using citric acid. Fermentation was conducted at 30°C for 72 h under semi-aerobic conditions with intermittent stirring. After fermentation, the mixture was heated at 80°C for 10 min to inactivate yeast. The grains were cooled, separated by centrifugation (5000 rpm, 10 min), dried at 40°C to constant weight, milled using an attrition mill, sieved through 0.25 mm mesh, and stored in airtight containers.



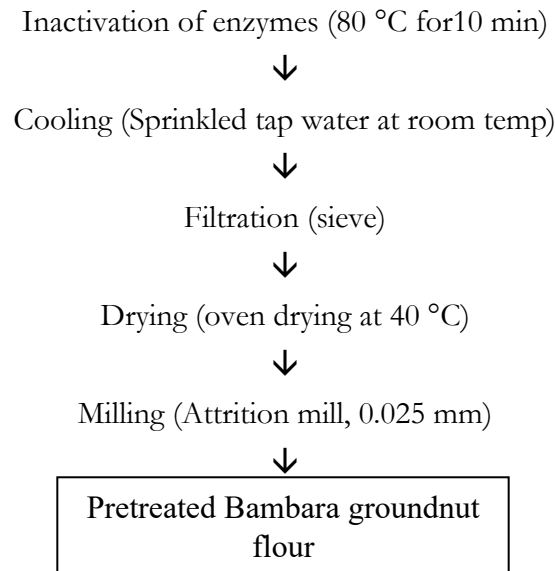


Figure 2. Pretreatment of dehulled and whole-grain Bambara groundnuts with *Saccharomyces cerevisiae* (Olaniran *et al.*, 2020)

### **Traction of Starch from Pretreated and Control Samples**

Following a modified version of the procedure described by Oyeyinka *et al.* (2015), starch was extracted from both the control and yeast pretreated BGN samples. A suspension of 500 g of pre-treated BGN flour was agitated on an orbital shaker at room temperature for 4 h after being mixed with 5 L of a 0.3% (w/v) NaOH solution in a ratio of 1:10. After 4 h of settling, the suspension was strained and the liquid above was poured off. The residue was then mixed with distilled water and the resulting slurry was passed through a 500-mesh sieve, which has a pore size of 0.025 mm. After letting the mixture sit for 12 h to settle, the remaining starch was centrifuged at 10,000×g for 20 min following several washes with distilled water. Following a neutralization step with 0.1 N HCl, the starch was dried at 40 °C for 24 h. Prior to usage, the samples were kept in a cool, dry area, wrapped in plastic bags, and mixed with food.

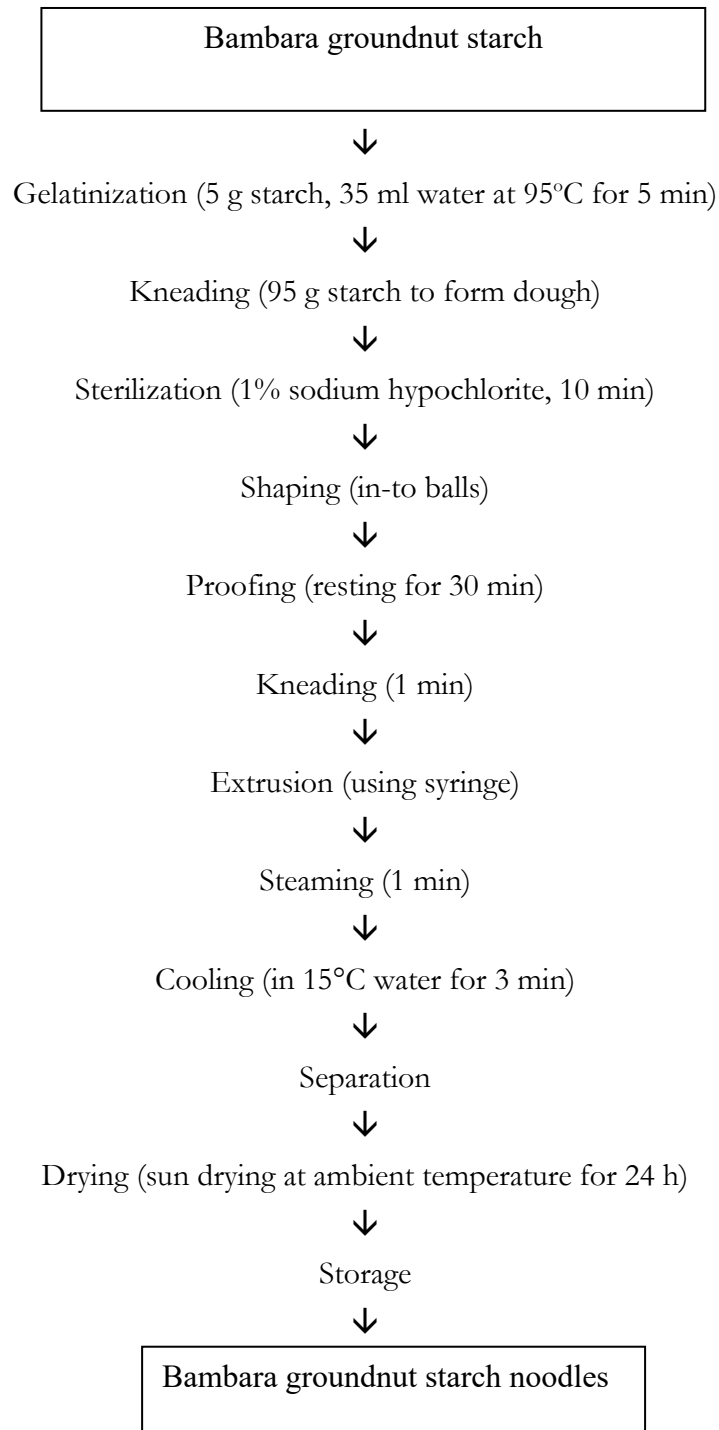


Figure 3. Bambara groundnut starch (*Baljeet et al., 2010*)

## Analytical methods

### Determination of starch yield

The percentage starch yield was determined according to the method described by Chinma *et al.* [26]. using equation 3.1.

$$\% \text{ Yield} = \frac{W_2}{W_1} \times 100 \quad (3.1)$$

Where W1= weight of whole grain BGN

W2 =weight starch after extraction

### Determination of techno-functional properties of Bambara groundnut Starch

#### Water absorption capacity

The ability of the sample to absorb water was measured by adding 10 ml of distilled water to 1 gram of the sample in a beaker, using a procedure adapted from that of Adebawale *et al.* [27]. The mixture was mixed for 5 min using a magnetic stirrer. After that, the mixture was spun in a centrifuge at 3500 rpm for half an hour, and the resulting liquid was transferred to a graduated cylinder with a volume of 10 ml. To determine how much water was absorbed, the volume of the supernatant was subtracted from the amount of water supplied to the sample at the beginning:

$$WAC (\%) = \frac{\text{Amount of water added} - \text{Volume supernated}}{\text{Weight of sample}} \times 100 \quad (3.2)$$

#### Oil absorption capacity

Oil absorption capacity was determined using the method modified by Adebawale *et al.* [28], where 10 ml of oil was added to 1.0 g of the sample in a beaker. For 5mins, the magnetic stirrer was used to mix the mixture. After that, the mixture was spun in a centrifuge at 3500 rpm for half an hour, and the resulting liquid was transferred to a graduated cylinder with a volume of 10 ml. By subtracting the volume of the supernatant from the volume of the oil supplied to the sample initially, the amount of oil absorbed was determined as:

$$OAC (\%) = \frac{\text{Amount of oil added} - \text{Volume supernated oil}}{\text{weight of sample}} \times 100 \quad (3.3)$$

### Swelling capacity

The approach developed by Chinma *et al.* [29] was used to assess the swelling capacity. A cleaned and dried graduated cylinder was used to weigh 20 g of starch. The cylinder was subjected to three taps on the table before being filled with 80 ml of distilled water. After letting the cylinder rest for 1 h, the sample's ultimate volume was recorded. Swelling capacity (SC) on volume basis is the ratio of final volume to the initial volume. After removing the liquid at the top, we measured the starch samples and the cylinder's weight. The swelling capacity was calculated as the ratio of the end sample weight to the initial sample weight, expressed as:

$$SC (\%) = \frac{\text{Volume of soaked sample} - \text{Volume of sample before soaking}}{\text{Grams of sample used}} \times 100 \quad (3.4)$$

### Bulk density (packed and loose)

The bulk density was determined using the method outlined by Onwuka [30]. The 25 mL graduated cylinder was filled with the 10 g test starch sample by gently tapping it on the bench top ten times from a height of 5-8 cm. The packed bulk density was calculated by dividing the volume of the test sample by the volume after tapping, and then taking the final volume measurement. Without tapping, 10 g of starch sample was added to a 25 mL graduated cylinder to obtain the loose bulk density, which was then computed using the packed bulk density method:

$$\text{Bulk density (g/ml)} = \frac{\text{Weight of sample}}{\text{Volume of sample after tapping}} \quad (3.5)$$

### Least gelation concentration

The least gelation concentration was evaluated using the method of Adebowale *et al.* [27]. Flour suspension in 10 mL distilled water was prepared to obtain 2 %, 4 %, 6 %, 8 %, 10% and 12 % (w/v) suspension. The test tubes containing five suspensions were heated in boiling water for 1h and then cooled under running tap water for an h. The least gelation concentrations were determined as the concentration at which the sample from the inverted test tube did not fall or slip.

### **Wettability**

The wettability of the sample was determined according to the method of Onwuka [30]. A 25 ml test tube with a 1cm diameter was used for the starch (1 g). To prevent spilling the sample by turning it upside down, a finger was put on the specimen's aperture. The specimen was held with its fingertip 10 cm above a 600 ml beaker filled with 500 ml of pure water. After removing the finger, the sample was carefully transferred to the beaker. How long it takes for a sample to get totally moist is known as its wettability.

### **Dispersibility**

The dispersibility of starch was determined according to the method described by Chinma *et al.* [26]. A fixed sample weight of 10 g was placed in 100 ml graduated cylinder and distilled water was added to make a volume of 100 ml. The mixture was vigorously stirred and allowed to stand for 3 h. The volume of settled particles was recorded and subtracted from 100 and the difference was reported as percentage dispersibility:

$$\text{Dispersibility (\%)} = 100 - \text{Volume of settled particles} \quad (3.6)$$

### **Determination of Pasting Properties of the Starch:**

Pasting properties was determined with the Rapid Visco Analyzer (RVA), RVA-Super3 (Newport Scientific Pty, Ltd., Australia). The samples, weighing 3.0 g, were carefully added to the test canister. A volume of 25.0 ml of distilled water was added to the container. Once turned on, the Visco Analyzer instantly recorded the flour's pasting performance on its graded sheet.

### **Determination of Morphological Properties of Starch**

The dehulled and whole grain BGN starch samples were analyzed by scanning electron microscopy (SEM) according to the protocol described by Chinma *et al.* [31]. To start, a carbon coater was sprayed onto the samples to eliminate charge effect. Coated samples were put on an aluminum stub and examined using a scanning electron microscope after being exposed to an electron beam in a vacuum. Images were acquired at a magnification of 1000X using an acceleration voltage of 10 kV, while the microscope was operating.

## **Determination of Chemical Composition of the Starch**

### **Amylose and amylopectin contents**

The AOAC-described technique was used to evaluate amylose and amylopectin AOAC [32]. A Buck Scientific BLC10/11-model HPLC with a UV 205 nm detector was fed 20  $\mu$ l of the meticulously prepared sample. At room temperature, a mobile phase consisting of 95:5 (methanol: water) was utilized with a C18 column of 4.5 x 150 mm and 5  $\mu$ m particle size, running at a flow rate of 1.00 mL/min. The same methodology was used to identify 0.1 mg of mixed standards. In order to identify the peaks, we compared the retention durations of the samples with those of the real standards. A calibration curve with four points was used to determine concentrations.

### **Evaluation of starch digestibility**

#### **Determination of Resistant and digestible starch**

Resistant starch were determined by the method described by (AOAC, 2002) using Megazyme Resistant Starch Assay. Boiled and homogenised samples were incubated with 10 mL of HCl-KCl buffer (pH 1.5) and 20 mg pepsin for 1 h at 37 °C. After that, the samples were subjected to continuous shaking at 37 °C for 16 h in order to hydrolyze starch using a pancreatic  $\alpha$ -amylase (10 mg/mL) solution that included amyloglucosidase (AMG). Three washes with ethanol (99% v/v and 50% ethanol) were performed on the samples after hydrolysis. Following additional digestion with 2 M KOH, the pellet and supernatant were incubated with AMG independently. Megazyme, a glucose oxidase-peroxidase kit, was used to quantify the glucose released. A spectrophotometer (Jenway 6405, UK) was used to detect the absorbance at 510 nm in comparison to the reagent blank. The digestible starch (DS) and resistant starch (RS) were determined using the glucose content of the supernatant and digested pellet, respectively, by using a factor of 0.9.

**Total starch (TS) was then derived as the sum of DS and RS.**

#### **Rapidly digestible starch (RDS) and slowly digestible starch (SDS)**

The process of Goni *et al.* [33], was used as a basis for the modified *in vitro* approach that was used. In a 1-h incubation at 37 °C with continuous shaking, the homogenized and boiled samples were mixed with 10 mL of HCl-KCl buffer (pH 1.5) and 20 mg of pepsin. An incubation period of 45 min at 40°C followed the addition of 200L of

pancreatic  $\alpha$ -amylase solution (1.5 mg/10 mL phosphate buffer) to increase the pH. The tris-maleate buffer (pH 6.9) was used to dilute the samples to 25 mL after 70  $\mu$ L of  $\text{Na}_2\text{CO}_3$  solution was added to halt the enzyme reaction. The sample was then incubated at 37 °C with steady shaking after adding 5 mL of pancreatic  $\alpha$ -amylase solution. To stop the enzyme process, 1 mL aliquots were collected from the samples at 30 and 120 min and mixed with hot water while being vigorously shaken for 5 minutes. Separate portions were mixed with 3 ml of a 0.4M sodium acetate buffer (pH 4.75) and 60  $\mu$ l of AMG (3300 U/mL), and thereafter left to incubate at 60 °C for 45 minutes while being constantly shaken. A glucose oxidase-peroxidase (GOPOD) kit was used to quantify the glucose that was released. In comparison to the reagent blank, absorbance was measured at a wavelength of 510 nm. A factor of 0.9 was used to convert glucose into starch. Hydrolysis times of 30 min were used to represent quickly digested starch (RDS) and 120 minutes for slowly digestible starch (SDS).

### **Statistical Analysis**

The study followed a completely randomized design. All experiments were conducted in triplicate. Data were analyzed using ANOVA in IBM SPSS version 23. Mean separation was performed using Duncan's New Multiple Range Test (DNMRT) at  $p < 0.05$  significance level.

## **RESULTS AND DISCUSSION**

### **Yield of Starch from Bambara Groundnut**

The yields of the starches from Bambara groundnut are shown in Table 2. The yields varied from 34.88% for the whole BGN grains to 39.66% for the pretreated dehulled BGN grains. Dehulling and pretreatment of BGN with yeast improved the starch yield. Dehulled BGN (37.04%) had significantly ( $p < 0.05$ ) higher starch yield than the undeulled whole BGN (34.88%). Pretreatment of dehulled BGN with yeast produced the highest starch yield (39.66%), which was significantly ( $p < 0.05$ ) different from those of other treatments. The pretreated whole BGN grains also had higher starch yield than untreated grains but lower than that of the dehulled and pretreated samples. The higher starch yield for the dehulled grains may be attributed to the removal of the seed coat, which contains fibre, tannins, and anti-nutritional compounds that often interfere with starch recovery [34]. Removing the hull therefore, exposed the cotyledon and made starch granules more

accessible during the extraction. Similarly, the increase in yield due to the yeast (*S. cerevisiae*) pretreatment may be ascribed to microbial and enzymatic activities, particularly the partial breakdown of cell wall polysaccharides and proteins, which weakened the matrix surrounding the starch granules and facilitated their release [34]. Fermentation also helped to reduce bound phenolic compounds and soluble sugars that compete with starch during extraction, thereby improving starch recovery efficiency [34]. These results did not corroborate the findings of Chinma *et al.* [8], who reported starch yields for raw and germinated BGN to be within 28.06–32.10%. The variation may be attributed to differences in BGN varieties used, growing conditions, and the pretreatment methods applied.

**Table 2: starch yield from Bambara groundnut**

Samples	Starch yield (%)
A	34.88 <sup>d</sup> ±0.01
B	37.04 <sup>b</sup> ±0.01
C	36.34 <sup>c</sup> ±0.01
D	39.66 <sup>a</sup> ±0.01

Values are means  $\pm$  SD of three replications. Means within a column not followed by the same superscript are significantly different ( $p < 0.05$ ).

A= Starch from whole-grain Bambara groundnut.

B= dehulled Bambara groundnut.

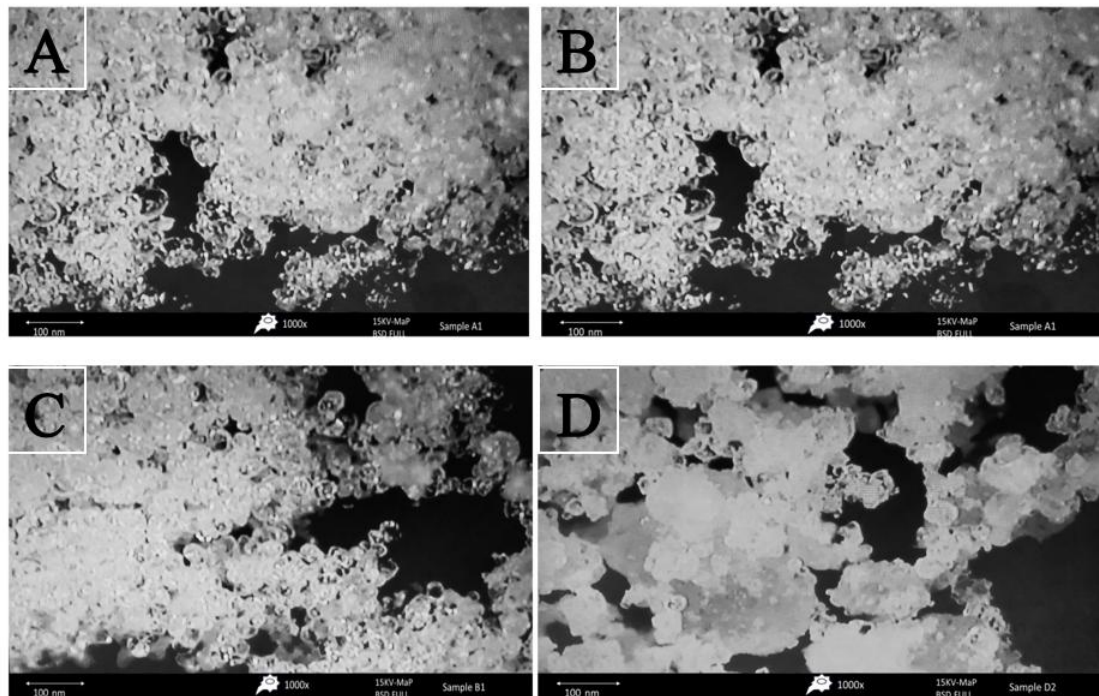
C = pretreated whole-grain Bambara groundnut.

D= pretreated dehulled Bambara groundnut.

### Morphological Structure of Bambara Groundnut Starches

The micrograph of the Scanning Electron Microscopy (SEM) of the starches is presented in plate. 4. The size and shape of starch granules are important as they help in determining the functionality and applications of starch in food. The smallness or largeness of the starch granules determines their uses. Generally, cereal starch granules are mostly small and the tuber starch granules are large. In contrast, the legume starch granules seem intermediate between the two, but some legume starch granules are very small [9] The shape of the starch granules studied with scanning electron microscopy (SEM) has oval, although, spherical, round or ring- like, elliptical and irregularly shaped. Most surfaces of

native starch granules are smooth without pinholes and cracks, but there are exceptions. Maraan et al. [35] also reported rough surfaces and some indentations on navy bean, rice bean, lablab bean, and tepary bean starch granules. In another study, the starch granular surfaces of pea, faba bean [36], and seven varieties of field peas were observed to be smooth, although, some of the large sized granules possessed deep indentations and grooves [37]. It seems that surface flaws in legume starch granules are proportional to their size. One probable correlation could be susceptibility to breaking which increases with size. These depressions might be the result of drying and isolation rather than anything to do with the plant's physiology or biology as it grows. Pinholes were not restricted to legume starches, they were also found on surface granules of corn, sorghum, and potato starches [9]. In contrast, pinholes were not indicated on compound granules (rice and oat), tuber and root starches (tapioca, arrowroot, canna, and potato). Some physical modification processes like hydrothermal treatment [37], and high hydrostatic pressure treatment [28] can bring about the manifestation of cracks on the granule surfaces. In one special case, the surfaces of rice bean starch granules extracted with water were smooth and that extracted with ethanol were rough with some indentations [37]. The manifestation of small pores might be during granule growth and under the influence of genetic control [9]. The cause of the individual characteristics and morphologies of various starch granules are not known, but the factors are genetic control, types, and amounts of enzymes in the biosynthesis of the starch molecules, membranous structure of the amyloplast organelles, arrangement, and association of starch molecules [9]. The clustering of single granules to form compound granules is fairly common in the study of legume starches. Compound granules were evident in white cowpea [9], wrinkled peas [39], beach pea, green pea and grass pea starches [9]. This could be due to the presence of residual protein or might be ascribed to the drying conditions that produce slight gelatinization on the granular surfaces and cause the granules to adhere together to form aggregates. The dissimilarities in granule morphology might be ascribed to the biological origin, biochemistry of the amyloplast, and physiology of the plant [9]. Many studies revealed that water has the biggest environmental impact on the granule size, but the granule shape might be minimally affected by rainfall and temperature [26].



**Plate 4. Scanning Electron Microscopy (SEM) of Bambara Groundnut Starch.**

A= whole grain Bambara groundnut.

B= dehulled Bambara groundnut.

C = pretreated whole-grain Bambara groundnut.

D= pretreated dehulled Bambara groundnut.

### **Amylose and Amylopectin of Bambara Groundnut Starches**

The amylose and amylopectin contents of whole-grain Bambara groundnut, dehulled Bambara groundnut, pretreated whole-grain Bambara groundnut and pretreated dehulled Bambara groundnut starches are shown in Table 3. The amylose and amylopectin contents of the Bambara Groundnut starch samples ranged from 20.50% to 26.16% and 73.84% to 79.50%, respectively. There was no significant difference ( $p>0.05$ ) in amylose and amylopectin contents of pretreated dehulled Bambara groundnut and pretreated whole grain Bambara groundnut starches. A similar report on amylose fraction in the yam starches ranged from 20.80 to 33.11 % [40]. Lower amylose contents observed for whole grain Bambara groundnut indicate higher swelling power than the pretreated whole grain Bambara groundnut. The higher the amylose contents, the lower the swelling power [41].

**Table 3: Amylose and Amylopectin of Bambara Groundnut Starch**

Samples	Amylose	Amylopectin
A	20.50 <sup>d</sup> ±0.03	79.50 <sup>a</sup> ±0.03
B	22.33 <sup>c</sup> ±0.03	77.68 <sup>b</sup> ±0.02
C	26.16 <sup>a</sup> ±0.01	73.84 <sup>d</sup> ±0.01
D	24.47 <sup>b</sup> ±0.02	75.54 <sup>c</sup> ±0.02

Values are means  $\pm$  SD of three replications. Means within a column not followed by the same superscript are significantly different ( $p \leq 0.05$ ).

A= whole grain Bambara groundnut.

B= dehulled Bambara groundnut.

C = pretreated whole-grain Bambara groundnut.

D= pretreated dehulled Bambara groundnut.

### Digestibility of Bambara Groundnut Starch

The starch digestibility of whole-grain Bambara groundnut and dehulled Bambara groundnut, pretreated whole grain Bambara groundnut and pretreated dehulled Bambara groundnut subjected to *S. cerevisiae* are shown in Table 4. The Total Starch (TS), Resistant Starch (RS), Slowly Digestible Starch (SDS) and Rapidly Digestible Starch (RDS) contents of the Bambara Groundnut starch samples ranged from 61.46% to 66.75%, 13.91% to 21.80%, 23.54% to 28.44% and 50.05% to 63.28%, respectively. Compared to the whole-grain Bambara groundnut, pretreated whole grain Bambara groundnut had higher TS and RDS contents and lower RS and SDS contents. These may be due the effect of pretreatment with *S. cerevisiae*, which broke down the lignocellulosic components of the Bambara groundnut. Starch digestibility is largely dependent on starch morphological, crystalline, and helical structures. Maraan *et al* [35] reported a similar range of results on microwave treatment, which caused the appearance of hollows on the surface of the starch granules, and facilitated digestion by enabling the invasion of enzymes.

**Table 4: Digestibility of Bambara Groundnut Starch**

Samples	TS (%)	RS (%)	SDS (%)	RDS (%)
A	61.46 <sup>d</sup> ±0.03	21.80 <sup>a</sup> ±0.02	28.44 <sup>a</sup> ±0.02	50.07 <sup>d</sup> ±0.04
B	62.98 <sup>c</sup> ±0.01	18.68 <sup>b</sup> ±0.01	26.28 <sup>b</sup> ±0.01	55.05 <sup>c</sup> ±0.03
C	66.75 <sup>a</sup> ±0.01	13.91 <sup>d</sup> ±0.02	23.54 <sup>d</sup> ±0.02	63.28 <sup>a</sup> ±0.04
D	64.50 <sup>b</sup> ±0.03	15.98 <sup>c</sup> ±0.01	25.65 <sup>c</sup> ±0.01	58.48 <sup>b</sup> ±0.02

Values are means  $\pm$  SD of three replications. Means within a column not followed by the same superscript are significantly different ( $p \leq 0.05$ ).

A= whole grain Bambara groundnut.

B= dehulled Bambara groundnut.

C = pretreated whole-grain Bambara groundnut.

D= pretreated dehulled Bambara groundnut.

### **Proximate Composition of Bambara groundnut Starches**

The proximate composition of Bambara groundnut starches is shown in Table 5. There was significant ( $p > 0.05$ ) difference in the crude fiber contents of the starch flours ( $p < 0.05$ ). The crude fiber contents of the starches from whole-grain Bambara groundnut, dehulled Bambara groundnut, pretreated dehulled Bambara groundnut and pretreated whole-grain Bambara groundnut were 0.02%, 0.40%, 0.55% and 0.90%, respectively.

There was no significant difference ( $p > 0.05$ ) in crude fiber contents of the samples of starch. Crude fiber contents of the starch flour samples ranged from 2.02-8.90%. The pretreated whole grain Bambara groundnut had higher fibre content than whole grain Bambara groundnut with the lowest fibre content. The values obtained in this study corroborated with 0.51% reported by Oppong et al. [42] for refined wheat flour. Chinma et al [26] reported crude fibre contents of 8.19% for pigeon pea, 9.58% for cowpea, 4.61% for mung bean and 6.83% for pea starch, which were significantly higher than those obtained in this study. Similar increases in fibre content after dehulling and germination have been reported in horsegram [39]. The moisture contents of the starches ranged from 5.48% to 6.01%, with pretreated whole grain Bambara groundnut having the highest value. These values were similar to those of Akubor et al. [44] for blends of wheat, unripe banana and cowpea flours. All values were within the acceptable limit of 10% for long-term storage Oppong et al. [42], which as Adebo et al. [45] noted in cereals and legumes, enhanced shelf-life by preventing microbial growth.

The crude protein contents of the starch samples were 0.23%, 0.58%, 0.61% and 0.77% for whole grain, dehulled, pretreated dehulled and pretreated whole grain starch flours, respectively, with the higher values in pretreated samples attributed to microbial hydrolysis and amino acid release. Fermentation has similarly been shown to improve

protein quality in urad bean [46], grey pea [47] and horsegram Akubor et al. [44]. Ash contents ranged from 0.50% to 0.61%, values lower than the 1.00–3.00% reported by Oppong et al. [42]. Nonetheless, pretreated samples showed slightly higher ash content, comparable to findings in yellow eye beans where dehulling enhanced mineral bioavailability [48]. Fat contents varied between 0.52% and 0.96%, with pretreated samples having higher values, trend also noted in faba beans [26], and other fermented legumes [45].

Carbohydrate contents ranged from 90.92% to 93.20%, with significant differences among samples ( $p > 0.05$ ) except for the pretreated whole grain starch. The high carbohydrate concentration suggests potential use in combating protein-energy malnutrition. A similar trend was observed for grey pea, where fermentation increased carbohydrate levels while enhancing phenolic compounds [47]. As Okereke et al. [49] reported, high carbohydrate starches are desirable for energy provision in weaning and breakfast foods.

**Table 5: Proximate Composition (%) Bambara Groundnut Starches**

Samples	Crude fiber	Moisture	Crude protein	Ash	Crude fat	Carbohydrates
A	0.02 <sup>d</sup> ±0.03	5.52 <sup>b</sup> ±0.03	0.23 <sup>d</sup> ±0.04	0.51 <sup>c</sup> ±0.01	0.52 <sup>c</sup> ±0.01	93.20 <sup>a</sup> ±0.29
B	0.40 <sup>c</sup> ±0.14	5.48 <sup>b</sup> ±0.04	0.58 <sup>c</sup> ±0.04	0.50 <sup>c</sup> ±0.01	0.55 <sup>c</sup> ±0.02	92.49 <sup>b</sup> ±0.09
C	0.90 <sup>a</sup> ±0.14	6.01 <sup>a</sup> ±0.01	0.77 <sup>a</sup> ±0.03	0.57 <sup>b</sup> ±0.00	0.83 <sup>b</sup> ±0.02	90.92 <sup>d</sup> ±0.18
D	0.55 <sup>b</sup> ±0.07	5.53 <sup>b</sup> ±0.04	0.61 <sup>b</sup> ±0.03	0.61 <sup>a</sup> ±0.12	0.96 <sup>a</sup> ±0.01	91.74 <sup>c</sup> ±0.25

Values are means  $\pm$  SD of three replications. Means within a column not followed by the same superscript are significantly different ( $p \leq 0.05$ ).

A= Whole grain Bambara groundnut.

B= Dehulled Bambara groundnut.

C = Pretreated whole-grain Bambara groundnut.

D= Pretreated dehulled Bambara groundnut.

## CONCLUSION

This study comprehensively characterized starch extracted from pretreated whole-grain Bambara groundnut. Dehulling and fermentation significantly improved starch yield, purity, and digestibility while moderately modifying molecular composition and

microstructure. The increase in amylose content and reduction in resistant starch indicate enhanced enzymatic accessibility and functional performance. SEM analysis confirmed structural modification without complete granule destruction.

Overall, pretreatment strategies effectively enhanced the physicochemical and nutritional properties of Bambara groundnut starch. These findings support its potential application in food processing, industrial formulations, and value-added legume utilization. Future research should explore pasting properties, thermal transitions, rheological performance, and industrial-scale optimization to further expand its commercial potential.

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