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Chemical composition of bambara groundnut (*V. subterranea* L. Verdc) seed parts

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Abstract

The levels of proximate composition, minerals, antinutrients, fibre components and calculated parameters for mineral bioavailability were determined in the testa, dehulled and whole seeds of Bambara groundnut on dry weight basis. Proximate levels were (g/100 g): ash (2.46-4.36); crude fat (2.47-6.99); crude protein (15.2-22.2); crude fibre (1.03-22.9) and carbohydrate (51.6-61.9). The non-starch polysaccharide (NSP) components were (%): ADF, 7.13-29.0 (or 16.1 %-65.5 %); NDF, 1.77-23.6 (or 6.28 %-83.7 %); ADL, 6.15-28.0 (or 14.9 %-67.8 %); cellulose, 1.36-23.3 (or 5.02 %-86.0 %) and hemicellulose, 0.84-26.5 (or 2.86 %-90.1 %). In minerals (mg/100 g): Mn, Co and Cu were not detected; Na, K, Ca, Mg, Fe and P were low in values whereas Zn was high at 11.2-40.2. These parameters were also good for human health: Na/K (0.47-0.51) and Ca/Mg (2.58-4.36). Antinutrient values showed that Phy was high (14.4-29.2 mg/g); oxalate was high (5.02-8.59 mg/g) and unavailable phosphorus as Pp % of P (10.2-49.3 %). The mineral bioavailability showed Ca/Phy to be good at 0.20-0.89 and [Ca] [Phy]/[Zn] to be good at 0.09-0.23 thereby making Zn bioavailable in all the samples.

Keywords: Bambara groundnut seeds; Proximate; Minerals; Antinutrients; Fibre composition

Introduction

Bambara groundnut (*Vigna subterranea* L. Verdc) is a seed crop of African origin. It is cultivated principally by farmers as a famine culture crop because of its agronomic values and the ability to produce in soils considered insufficiently fertile for cultivation of other more favoured species such as common beans and groundnuts (*Arachis hypogaea*) (Anchirinah *et al.*, 2001). It is very adaptable to hot temperatures but it is also tolerant to rainfall (Wrigley, 1981). Bambara seeds may be consumed in various forms for food. Fresh seeds may be consumed raw, boiled, grilled or dry seeds made into a powdery form to make cakes (Adebowale and Lawal, 2002).

The nutritional potentials of bambara groundnut were documented. The seed is regarded as a balanced food because when compared to most food legumes, it is rich in iron and the protein contains high lysine and methionine (Adu-Dapaah and Sangwan, 2004). In addition, it is known to contain 63 % carbohydrates, 18 % oil and the fatty acid content is predominmatly linoleic, palmitic and linolenic acids (Minka and Bruneteau, 2000). It was reported also that it is richer than groundnut in essential amino acids such as leucine, isoleucine, lysine, methionine, phenylalanine, threonine and valine (Ihekoronye and Ngoddy, 1985).

Soils of medium or low fertility, with a pH of 5.0 - 6.5 will produce satisfactory crops. Yields of bambara groundnut on low - fertility soils are generally higher than those of groundnut grown on similar soils. Bambara groundnut will often yield well in environments that may be too hostile for more favoured legumes (Collinson *et al.*, 1996).

Recently, Pasquet *et al.* (1999), using isozyme analysis, found high genetic identities between wild and domesticated bambara groundnut accessions and concluded that the wild bambara is the progenitor of the domesticated form, both being characterized by low total genetic diversity.

An evenly distributed rainfall in the range 600-1000 mm encourages optimum growth but satisfactory yield can be obtained in areas with pronounced dry season since the crop is relatively drought resistant. It is tolerant to periods of heavy rainfall except during the flowering period.

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Water level is an important factor affecting the emergence and development of seedlings (Pollock, 1972). Studies have strongly established the central role of duration of flooding and temperature and their interaction in determining the effect of flooding stress on germination and establishment of seedlings. Massawe *et al.* (1999) reported that pre-swing hydration reduced final germination percentages significantly as the duration of soaking increases from 2 to 8 days and a complete loss in germination occurs when seeds are soaked for 6 days at 35^{o}C and 8 days at all temperatures.

It has been scientifically declared that bambara bean is high in protein quotient, particularly in methionine which makes its protein more complete than any other bean. The proximate composition of the bambara groundnut was reported to be 9.7 % moisture, 16.6 % protein, 5.9 % fat, 2.9 % ash, 4.9 % crude fibre and 64.9 % carbohydrate (Enwere and Hung, 1996).

The high concentration of soluble fibre than any other bean also makes it one step ahead of other beans. This further enhances its quality as nutritious food which reduces the incidence of heart disease and certain types of cancer. Also, bambara beans being nitrogen fixers themselves and along with providing the soil with essential nutrients do not require any artificial fertilizer. The use of artificial flavours or preservatives during the food processing is greatly discarded. (Olaleye, 2013).

Bambara groundnut was reported to have been fairly well supplied with calcium and iron although poor in phosphorus. It contains thiamine, riboflavin, niacin and carotene but very low in ascorbic acid (Oyenuga, 1968). The study of the microstructure of the raw flour and seed showed that they contained differently shaped and sized starch granules and protein materials within the cell wall in the cotyledon. Milling disorganized the arrangement of these components in the cotyledons (Enwere and Hung, 1996). It is a non-oily leguminous seed which contains only about 6% of ether extract. It contains an appreciable amount of lysine (Oyenuga, 1968).

Several other reports have been made on bambara nut. The swelling capacity increases with increase in temperature (Adebowale *et al.*, 2002). Bambara bean is higher in water absorption capacity than that of great Northern bean (Sathe and Salunkhe, 1981).

The digestion and bioavailability of the nutrients in the bambara seeds for animals and human nutrition is limited by antinutrients such as trypsin inhibitors and condensed tannins (Apata and Ologhobo, 1997). Condensed tannins are polyphenolic substances widely distributed in plants, especially in legumes and due to their large structure are known to inhibit protein digestibility by forming irreversible complexes with protein, thereby reducing the bioavailability of amino acids. However, recent research has also indicated that condensed tannins in low concentrations have beneficial effects in animal and human nutrition and health (Akindahunsi and Salawu, 2005).

Trypsin inhibitor is another antinutrient which inhibits the activity of digestive enzymes. Trypsin inhibitor activity in two raw bambara varieties was shown to be 9.4 Tiu/mg protein and 12.2 Tiu/mg protein (Apata and Ologhobo, 1997). Also, trypsin inhibitor activity was reported to range from 6.75 to 15.44 Tiu/mg in 100 g samples from eight bambara genotypes (Linnemann and Azam - Ali, 1993). The difference in activity could be due to different genotypes and environmental conditions such as fertility, climate, seasonality, rainfall and light intensity (Champ, 2002).

Condensed tannin content was found to range from 0.37 to 0.39% in two bambara landraces (Apata and Ologhobo, 1997). The condensed tannin content in bambara landraces is lower than in other legumes such as cowpea (Asante *et al.*, 2004) and pigeon pea (Fasoyiro *et al.*, 2005). However, Apata and Ologhobo (1997) have indicated that different processing methods such as cooking and roasting significantly reduce the tannin in bambara groundnuts.

Removal of the antinutrients would be necessary for effective utilization of proteins, carbohydrates and minerals in human nutrition.

This study reports on the proximate composition, mineral composition, various types of antinutrients, various types of fibre, calculation of various parameters on the quality of minerals and their bioavailability in the testa, dehulled and whole seed samples of Bambara groundnut. This type of work will likely be first of such cutting across the various parts of the seeds; it will likely enrich the food composition tables.

Materials and methods

Sample collection and preparation

The sample (bambara groundnut) was obtained from the Department of Plant Science, Ekiti State University, Ado-Ekiti. The seeds were screened to eliminate the defective ones, washed and rinsed with distilled water. The seeds were divided into two parts. One part was soaked with distilled water overnight while the other part was dried without soaking. The soaked ones were removed after twenty - four hours and were manually dehulled. Both the cotyledon and the testa were dried in an oven at 45^{o}C. All the three samples (whole seed, cotyledon and testa) were dry milled separately to fine powder and stored in a dry, cool place prior to use. The three samples were used for various analyses as described below.

Proximate analysis

The moisture content, ash content and crude fat were determined using the AOAC (2005) methods. The method of Joslyn (1970) was adopted to determine the total crude fibre whilst the method of Pearson (1976) was used to determine the crude protein. Carbohydrate was determined by difference.

Mineral content determination

Ash was dissolved in 10% HCl, heated, cooled, filtered and made up to the mark in 100 mL standard flask with distilled water. The mineral contents of the samples were analyzed for with the aid of atomic absorption spectrophotometer (Buck Scientific Instrument).

Determination of phytic acid and phytin phosphorus

4 g of the sample was soaked in 100 mL 2 % HCl for 3 hours and then filtered. 25 mL of the filtrate was placed in a 100mL conical flask and 5mL of 0.03 % NH_4SCN solution was added as indicator. 50 mL of distilled water was added to give it the proper acidity (pH 4.5). This was titrated with ferric chloride solution which contained 0.005 mg of Fe per mL of $FeCl_3$ used until a brownish yellow colour persisted for 5 min. Phytin phosphorus (Pp) was determined and the phytic acid content was calculated by multiplying the value of Pp by 3.55 (Young and Greaves, 1940). Each milligram of iron is equivalent to 1.19 mg of Pp.

Determination of tannin

200 mg of the sample was weighed into a 50 mL sample bottle. 10 mL of 70 % aqueous acetone was added and properly covered. The bottles were put in an orbital shaker and shaken for 2 hours at 30^{o}C. Each solution was then centrifuged and the supernatant stored in ice. 0.2 mL of each solution was pipetted into test tubes and 0.8 mL of distilled water was added. Standard tannic acid solutions were prepared from a 0.5 mg/mL stock and the solution made up to 1 mL with distilled water. 0.5mL folin reagent was added to both sample and standard followed by 2.5 mL of 20 % Na_2CO_3. The solutions were then vortexed and allowed to incubate for 40 minutes at room temperature after which absorbance was taken against a reagent blank concentration of the sample from a standard tannic acid curve (Makkar and Goodchild, 1996).

Determination of oxalate

1g of the sample was weighed into 100 mL conical flask. 75 mL of 1.5 NH_2SO_4 was added and the solution was carefully stirred intermittently with a magnetic stirrer for about 1 hour and then filtered using Whatman filter paper. 25mL of sample filtrate was collected and titrated hot (80-90^{o}C) against 0.1 $MKMnO_4$ solution to the point when a faint pink colour appeared that persisted for at least 30 seconds (Day and Underwood, 1986).

Determination of alkaloid

Alkaloid determination was carried out following the procedure of Harborne (1973). 5.0 g of the sample was weighed into a 250 mL beaker and 200 mL of 10 % acetic acid in ethanol was added and covered and allowed to stand for 4 h. This was filtered and the extract was concentrated on a water bath to one quarter of the original volume. Concentrated ammonium hydroxide was added drop wise to the extract until the precipitation was complete. The whole solution was allowed to settle and the precipitate was collected and washed with dilute ammonium hydroxide and then filtered. The residue is the alkaloid which was dried and weighed.

Determination of saponin

The method used was that of Obadoni and Ochuko (2001). 5 g of the sample was put into a conical flask and 100 mL of 20 % aqueous ethanol were added. The sample was heated over

a hot water bath for 4h with continuous stirring at about 55°C. The mixture was filtered and the residue re-extracted with another 200 mL 20 % ethanol. The combined extracts were reduced to 40mL over water bath at about 90°C. The concentrate was transferred into a 250 mL separating funnel and 20 mL of diethyl ether was added and shaken vigorously. The aqueous layer was recovered while the ether layer was discarded. The purification process was repeated. 60 mL n-butanol was added. The combined n-butanol extracts were washed twice with 10 mL of 5 % aqueous sodium chloride. The remaining solution was heated in a water bath after evaporation; the sample was dried in the oven to a constant weight. The saponin content was calculated as percentage.

Determination of flavonoid

The method of Boham and Kocipai-Abyazan (1974) was followed in the determination of flavonoid. 5 g of the sample was extracted repeatedly with 100 mL of 80 % aqueous methanol at room temperature. The whole solution was filtered through whatman filter paper (125 mL). The filtrate was later transferred into a crucible and evaporated into dryness and weighed to a constant weight

Colorimetric determination of phosphorus

Phosphorus was determined colorimetrically using a Spectronic 20 (Gallenkamp, London, UK) instrument, with KH_2PO_4 as a standard.

Determination of acid detergent fibre (ADF)

0.5 g of the sample was weighed into a Berzelius beaker. 50 ml cold Acid Detergent Solution (ADS) was added and boiled for 5-10 min. It was filtered on a previously weighed Gooch crucible. The residue was washed twice with hot water (90 - 100°C) and washed repeatedly with acetone until it was colourless. The crucible together with the fibre was dried at 105°C overnight and hot weighed and cooled (15°C). 72 % H_2SO_4 was added and stirred with a glass rod to a smooth paste. The crucible was kept at 20-30°C. The crucible was about half filled with tetraoxosulphate (VI) acid and stirred. The glass rod still remaining in the crucible, it was refilled with 72 % H_2SO_4 and stirred at hourly intervals as acid drained away. After 3 h, it was filtered to drain as much acid as possible using a vacuum pump. The content was washed with hot water until it was free from acid (by testing with litmus paper), rinsed and the stirring rod removed. The crucible was dried and hot weighed. The crucible was ignited in a muffle furnace at 550°C for 3 h, cooled to 105°C and hot weighed. ADF - Ash was reported as the difference between last weight and the original weight of the crucible (Van Soest and Robertson, 1980).

Determination of neutral detergent fibre (NDF)

0.5 g of the sample was weighed into a 600 mL Berzelius beaker. 50 mL cold neutral detergent solution (NDS) was added and boiled on a refluxing unit. The heat was adjusted in the process to even boiling and avoiding foaming, keeping the sample particles suspended. It was refluxed for 60 minutes from onset of boiling. It was filtered to a previously weighed crucible using light sunction. The residue was washed twice with hot water, twice with acetone and dried using sunction. Acetone washing was continued until a clear solution was obtained. The lumps were broken so that the solvent could come in contact with all particles of fibre.

The crucible was dried in air for 10-15 minutes (to drain part of the acetone) and oven dried for 8 hours at 105°C. It was hot weighed to obtain yield of cell wall. The crucible was ashed at 500°C for 8 hours, removed from furnace, put in oven (set at 100°C) and hot weighed. The loss in weight was the ash free cell wall (Van Soest and Robertson, 1980).

Lignin (%) = weight after oven drying-weight after furnace ashing x 100/weight of sample

Determination of cellulose

2.0 g of the sample was weighed into a 250 ml conical flask and 2 drops of paraffin oil, 25 mL of 8 % (v/v) acetic acid, 1 Lof concentrated nitric acid and 4 glass beads were added. It was refluxed for 20 min on a refluxing apparatus. The digest was washed into 500 mL centrifuge tube with hot 95 % ethanol and centrifuged at 1800 r. p. m. for 5 minutes. The liquid was decanted and 95 % ethanol was added, stirred and centrifuged for another 5 minutes. Liquid was decanted and sample washed twice with 95 % ethanol, hot benzene and once with petroleum ether into 25 mL crucible. The crucible was placed in the oven at 100°C for 1 hour and later placed in the desicator to cool and it was then weighed. The crucible was later placed in the furnace at a temperature of 600°C for 4 hours, cooled in a desicator and weighed for ash content.

The base weight less ash weight is the weight of cellulose.(Usoro *et al.,*1982)

Determination of hemicellulose

2 g of sample was weighed into two different conical flasks (A and B). 5 % (v/v) KOH was added to the sample in flask A while 24 % (v/v) KOH was added to the sample in flask B and both samples were allowed to stand for 2 hours.

The suspension was filtered using filter paper, washed with additional KOH solution and the filtrate was received into 2 different flasks (A and B) containing excess of glacial acetic acid. The hemicellulose was then quantitatively precipitated by the addition of ethanol. The precipitated hemicellulose was isolated by centrifuging for 10 minutes and was washed with ethanol and filtered. The samples were oven dried for 2 hours at 80^{o}C and transferred into a desicator and allowed to cool. Percentage hemicellulose was then calculated (Usoro *et al.*,1982).

Results and discussion

Proximate composition

The proximate compositions of the *Vigna subterranea* testa, dehulled and whole seeds are shown in Table I. The moisture content was generally low (5.23-9.23 g/100 g) in the three samples. The low moisture content would afford a long shelf life of the samples. The current report revealed the ash content as 2.46 g/100 g for testa, 2.97 g/100 g for dehulled and 4.36 g/100 g for whole seed. The ash value is in the order of testa< dehulled < whole seeds. Ash is a rough estimate of the mineral content of a sample. The present report for total ash is comparable to the value reported by Aremu *et al*. (2006) for *Prosopis africana*. The crude protein was generally high; this being typical of all legumes. However, the values obtained in this report (15.2-22.2 g/100 g) were lower than the value (29.0 g/100 g) reported for raw groundnut seeds (Adeyeye, 2011) and that of Prosopis africana with a value of 23.6 g/100 g (Aremu *et al.*, 2006). High level of protein in bambara groundnut would make it useful in supplementing the nutrients derived from tubers and cereals in places where other legumes are not easily available. However, the protein contents of the three samples: testa, dehulled and whole seeds (Table I) generally fell below the recommended 23-56 g/100 g human daily protein requirement (NRC, 1989). Thecarbohydrate contents (51.6 - 61.9 g/100 g) in the three samples were lower than the value (65.7 g/100 g) reported by Adeyeye and Aye (2005) for *Triticum durum* whole meal flour. The levels of lipids, protein, carbohydrate and energy were highest in the dehulled samples. Moisture content (9.23 g/100 g) and ash (4.36 g/100 g) were highest in the whole seed sample whereas crude fibre (22.9 g/100 g) was highest in the testa. The relatively high value of crude fibre in the testa is a nutritional advantage since fibre is important in facilitating faecal elimination and in dealing with cancer of the small intestine. Generally, the results of moisture, ash, protein and carbohydrate were close when compared on pair wise basis as it was evident in the levels of the coefficient of variation percent (CV %) with the range between 9.70 - 34.3. However, the CV % of the crude fibre (142) revealed a wide variation of results especially the testa as compared to other samples. Enwere and Hung (1996) had reported the proximate compositions of bambara groundnut samples as (g/100 g): 9.7 moisture, 2.9 ash, 5.9 fat, 16.6 protein, 4.9 crude fibre and 64.9 carbohydrate. Most of these results are close to the results of the present study except for crude fibre which was almost twice the value of the present study in the whole seed. Also, the report of Mune *et al.* (2007) was generally close

Table I. Proximate composition (g/100 g) of bambara groundnut samples

	Samples			Mean	SD	CV%
Parameter	Testa	Dehulled	Whole seed			
Moisture	5.36	5.23	9.23	6.61	2.27	34.3
Ash	2.46	2.97	4.36	3.26	0.98	30.1
Lipid	2.47	6.99	5.17	4.88	2.27	46.5
Protein	15.2	22.2	18.4	18.6	3.50	18.8
Fibre	22.9	1.03	2.05	8.66	12.3	142
Carbohydrate	51.6	61.9	60.8	58.1	5.66	9.70
Energy (kJ/100g)	91.4	259	191	180	84.3	46.8

except for crude protein which was fairly higher than the value obtained in the present report.

Table II shows the non-starch polysaccharide (NSP) components of bambara groundnut testa, dehulled and whole seed. The contents of acid detergent fibre (ADF), neutral detergent fibre (NDF) and acid detergent lignin (ADL) varied from 7.13 - 29.0 %, 1.77-23.6 % and 6.15 - 28 % respectively. The trend of ADF, NDF and ADL was testa > whole seed > dehulled. Among the three samples, testa was most concentrated with percentage concentration of 65.5 % (ADL), 83.7 % (NDF) and 67.8 % (ADL). The ADF and NDF content in the present report were lower than 31.9 - 49.6% (ADF) and 55.6 - 71.7 % (NDF) recorded for wild rice and cultivated rice, whereas ADL was higher (2.68 - 5.71%). Acid detergent fibre is a portion of the plant fibre (Van Soest and Robertson,1980). It includes the cellulose and lignin from cell walls and variable amounts of xylans and other components. Neutral detergent fibre is considered to be the entire fibre fraction of the feed, but it is known to underestimate cell wall concentration because most of the pectic substances in the wall are solubulized (Van Soest and Robertson, 1994). As a result, NDF is a poor estimate of cell wall concentration for the pectin-rich legumes. Acid detergent lignin is the most common lignin method in ruminant nutrition, but increasing evidence indicates, it underestimates lignin due to solubilzation of some lignin at the ADF step in the procedure (Lowry *et al.*, 1994).

Table II. Non starch polysaccharide (NSP) components of bambara groundnut samples (%)

	Samples			Mean	SD	CV%
Parameter	Testa	Dehulled	Whole seed			
ADF	29.0 (65.5%)	7.13 (16.1%)	8.16 (18.4%)	14.8	12.3	81.3
NDF	23.6 (83.7%)	1.77 (6.28%)	2.81 (9.96%)	9.39	12.3	131
ADL	28.0 (67.8%)	6.15 (14.9%)	7.17 (17.4%)	13.8	12.3	89.1
Cellulose	23.3 (86.0%)	1.36 (5.02%)	2.41 (8.89%)	9.02	12.4	138
HMC	26.5 (90.1%)	2.05 (6.97%)	0.84 (2.86%)	9.80	14.5	148

ADF = Acid detergent fibre; NDF = Neutral detergent fibre; ADL = Acid detergent lignin;
HMC = Hemicellulose.

Table III. Mineral content (mg/100g) of testa, dehulled and whole seed samples of bambara groundnut

	Samples			Mean	SD	CV%
Parameter	Testa	Dehulled	Whole seed			
Sodium	12.2	24.9	23.9	20.3	7.06	34.8
Potassium	25.8	49.3	50.7	41.9	13.9	33.2
Calcium	35.2	82.2	77.7	65.0	25.9	39.8
Magnesium	8.08	31.9	20.9	20.3	11.9	58.6
Zinc	11.2	40.2	25.6	25.7	14.5	56.4
Iron	1.91	5.27	4.25	3.81	1.72	45.1
Manganese	ND	ND	ND	-	-	-
Cobalt	ND	ND	ND	-	-	-
Copper	ND	ND	ND	-	-	-
Phosphorus	80.5	10.0	39.6	43.4	35.4	81.6
K/Na	2.11	1.98	2.12	2.07	0.078	3.77
Na/K	0.47	0.51	0.47	0.48	0.023	4.79
Ca/P	0.44	8.22	1.96	3.54	4.12	116
Ca/Mg	4.36	2.58	3.72	3.55	0.902	25.4

SD=Standard deviation, CV %=Coefficient of variation percent, ND= Not detected.

The results of cellulose and hemicelluloses in Table IV.II also showed that testa had the highest concentration with percentage concentration of 86.1% (cellulose) and 90.2% (hemicellulose). Cellulose is a major part of the structural fibre in forages and can be utilized by microorganisms in the rumen. Like cellulose, hemicellulose is a carbohydrate that exists in almost all plant cell wall along with cellulose. Cellulose is composed of only glucose whereas hemicellulose is composed of many different other sugars, (e.g, glucose, xylose, mannose, galactose, arabinose, etc).

Generally, high variation existed among the samples as seen in the values of coefficient of variation percent (CV %).

Mineral composition

Table III depicts the mineral contents (mg/100 g) of the three samples of bambara groundnut: testa, dehulled and whole seeds. Manganese (Mn), cobalt (Co) and copper (Cu) were not detected in any of the samples. Testa showed the highest level of phosphorus and Ca/Mg. Dehulled had the highest levels of Na, Ca, Mg, Zn, Fe, Na/K and Ca/P, whereas whole seed was best in K and K/Na. Both sodium and potassium were low in the samples. Sodium and potassium are required to maintain osmotic balance of the body fluid, the pH of the body, regulate muscles and nerve irritability, control glucose absorption and enhance normal retention of protein during growth (NRC, 1989). The major minerals in the present report were comparably lower than the values reported by Adeyeye (2011) for groundnut seed flour. The iron (Fe) level: 1.91-5.27 mg/100 g in the present study was comparatively lower than the 16.66 mg/100 g reported by Mune *et al.* (2007) for bambara groundnut bean flour, whereas, Ca and Zn were higher. Fe requirement by humans is 10-15 mg for children, 18 mg for women and 12 mg for men (Fleck, 1976). The phosphorus levels of 10-80.5 mg/100 g in this report were much below the recommended daily allowance (RDA) level of 800 mg. Also, the calcium levels of 35.2 - 82.2 mg/100 g were much lower than the RDA level of 800 mg (Adeyeye, 2011a). If Ca is adequately present in the diet, Fe is utilized to better advantage. This is an instance of 'sparing action' (Fleck, 1976). The Zn level (11.2 mg/100 g) in the testa was lower than the Zn allowance of about 15-20 mg per day (Fleck, 1976) whereas it was higher in dehulled (40.2 mg/100 g) and whole seed (25.6 mg/100 g). The Na/K and K/Na levels are also shown in Table III. The Na/K ratios in the samples ranging between 0.47 - 0.51 were good as they were lower than the 0.60 requirement to avoid high blood pressure (Adeyeye and Adamu, 2005). The K/Na ratios in the samples ranged between 1.98 - 2.12 meaning that more sodium is required to balance up to 1:1. K/Na enhances the salt balance of the body fluid (Adeyeye, 2011). The Ca/P weight ratio with values 8.22 and 1.96 in the dehulled and whole seed sample respectively were above the recommended value of 1.0 (NRC, 1989), whereas 0.44 recorded for testa fell far below. The Ca/P ratio was reported to have some effects on Ca in the blood of many animals (NRC, 1989) and also in the absorption of phosphorus. Food is considered 'good' if the Ca/P ratio is above 1.0 and 'poor' if the ratio is less than 0.5 (Aremu *et al.*, 2006). The Ca/Mg weight ratios in the samples fell within the range 2.58 - 4.36. These values

Table IV. Antinutrients in testa, dehulled and whole seed samples of bambara groundnut

	Samples			Mean	SD	CV%
Parameter	Testa	Dehulled	Whole seed			
Tannic acid (mg/100 g)	0.84	0.76	0.09	0.56	0.41	73.3
Phytin phosphorus (mg/100 g)	8.24	4.93	4.06	5.74	2.21	38.5
Phytic acid (mg/ g)	29.2	17.5	14.4	20.4	7.81	38.3
Oxalate (mg/ g)	6.06	8.59	5.02	6.56	1.84	28.0
Saponin (g/100 g)	1.28	1.38	1.01	1.22	0.19	15.6
Alkaloids (g/100 g)	0.39	0.27	0.14	0.27	0.13	48.1
Flavonoid (g/100 g)	`0.79	0.54	0.34	0.56	0.23	41.1
Total phosphorus (mg/100 g)	80.5	10.0	39.6	43.4	35.4	81.6
Pp % of P	10.2	49.3	10.3	23.3	22.5	96.6

Pp = Phytin phosphorus; P = Phosphorus.

were far above the recommended value of 1.0 (NRC, 1989). It means more magnesium would have to be supplied from other sources when the bambara groundnut is a source of food in the diet.

Antinutrients

The anti-nutritional factors are depicted in Table IV. The trend in tannin, phytin phosphorus (Pp), phytic acid (Phy), alkaloid and flavonoid was testa > dehulled > whole seed. In oxalate and saponin, it was dehulled > testa > whole seed while in Pp as % of P, it was dehulled > whole seed > testa. Phy had the highest concentration (14.4-29.2 mg/g) in all the three samples. The Phy levels fell within the range of values reported for 13 spices (390 - 6210 mg/100 g) obtained in Nigeria (Adeyeye and Fagbohun, 2005) and seven varieties of Nigerian garden egg fruits (507-2788 mg/100 g) (Adeyeye and Fagbohun, 2006). The present Phy values were higher than the values reported for many Nigerian foods such as legumes (14-344 mg / 100 g) and cereals (112-287 mg / 100 g) (Adeyeye *et al.*, 2000) and far above the range of 5.1-18.5 mg/100 g reported for *Canavaliafor ensiformis,* 6.0 - 15.3 mg /100 g for *Mucuna pruriens* seed flours (Agbede and Aletor, 2005) and 33.4 - 37.8 mg/100 g for groundnut seed samples (Adeyeye, 2011). The tannin contents in the present report (0.09-0.84 mg/100 g) were much lower than 0.35-0.85 mg/100 g reported for groundnut samples (Adeyeye, 2011), 0.3-0.9 g/100 g for *Canavalia ensiformis* and 0.8-7.8 g/100 g for *Mucuna pruriens* (Agbede and Aletor, 2005). The 4.8 - 8.24 mg/100 g recorded for Pp in the present report fell below 9.43 - 10.7 mg/100 g in groundnut seed flour (Adeyeye, 2011) and also fall below 0.06-0.29 g/100 g in 17 wild leguminous crop seeds (Balogun and Fetuga, 1986) but higher than 1.7-4.3 mg/100g in *Mucuna pruriens* and 1.4-5.44 mg/100 g in *Canavalia ensiformis* (Agbede and Aletor, 2005). The Pp as % of P ranging between 10.2-49.3 was higher than 2.28 - 2.67 in groundnut seed flour (Adeyeye, 2011). Pp as % of P shows how much P is linked to Pp and abnormal change in levels will affect the utilization of divalent minerals and also will render some essential amino acids unavailable.

The present report showed that dehulling enhanced the oxalate content of bambara groundnut. The oxalate content (5.02-8.59 mg/100 g) in this report is comparatively higher

Table V. Differences in antinutrients between whole seed and testa, and between whole seed and dehulled

Antinutrients	Whole seed-testa	Whole seed-dehulled	Means	SD	CV%
Tannic acid	- 0.75(-833%)	-0.67(-744%)	0.71	0.06	8.45
Pp	- 4.18(-103%)	-0.87(-21.4%)	2.53	2.34	92.5
Phytic acid	-14.8(103%)	-3.1(-21.5%)	8.95	8.27	92.4
Oxalate	-1.04(-20.7%)	-3.57(-71.1%)	2.31	1.79	77.5
Saponin	-0.27(-26.7%)	-0.37(-36.6%)	0.32	0.07	21.9
Alkaloid	-0.25(-179%)	-0.13(-92.9%)	0.19	0.08	42.1
Flavonoid	-0.45(-132%)	-0.2(-58.8%)	0.33	0.18	54.5
Pp as % of P	+0.1 (0.97%)	-39.0(-379%)	19.6	27.5	140

Table VI. Concentration of Zn, Ca, Phytate and calculated Phy: Zn Ca: Phy and [Ca] [Phy] / [Zn] molar ratios of testa, dehulled and whole seed samples of bambara groundnut

	Samples			Mean	SD	CV%
Parameter	Testa	Dehulled	Whole seed			
Zn (mg/100 g)	11.2	40.2	25.6	25.7	14.5	56.4
Ca (mg/100 g)	35.2	82.2	65.0	65.0	25.9	39.8
Phy (mg/100 g)	2920	1750	2036	2036	781	38.4
Phy / Zn	25.8	4.31	5.57	11.9	12.1	102
Ca / Phy	0.20	0.77	0.89	0.62	0.37	59.7
[Ca] [Phy]/ [Zn].	0.23	0.09	0.11	0.14	0.08	57.1

Table VII. Differences in Phy: Zn, Ca: Phy and [Ca] [Phy] / [Zn] molar ratios of bambara groundnut samples

Antinutrients	Whole seed-testa	Whole seed-dehulled	Means	SD	CV%
[a]Phy: Zn	-20.2(-363%)	1.26(22.6%)	10.7	13.4	125
[b]Ca: Phy	0.69(77.5%)	0.12(13.5%)	0.41	0.40	97.6
[c][Ca] [Phy] / [Zn]	-0.12(-109%)	0.02(18.2%)	0.07	0.07	100

[a]mg of Phy/MW (molecular weight of Phy): mg of Zn/MW of Zn; [b] mg of Ca/MW of Ca: mg of Phy/MW of Phy; [c] [mol/kg Ca] x [mol/kg Phy]/ [mol/kg Zn]

than 4.08-6.42 mg/100 g reported for groundnut seed flour (Adeyeye, 2011). The presence of oxalate negatively affects the absorption and utilization of calcium. Oxalate combines with calcium to form calcium oxalate which passes through the intestine without being absorbed. Calcium oxalate is responsible for most of the kidney stone formation. Formation of these stones frequently reflects chronic alkalinity of bladder and renal pelvic urine caused by infection with bacteria that hydrolyse urea, releasing ammonia (White *et al.*, 1973). Generally, the antinutrients in this report were much concentrated in the testa. This agrees with the observation of Preet and Punia (2000) in cowpea varieties.

Tannic acid was low at 0.09-0.84 mg/100 g; saponin was low at 1.01-1.38 g/100 g; flavonoid was low 0.34-0.79 g/100 g; alkaloid was low at 0.14 -0.39 g/100 g. Tannins have been reported to bring about their antinutritional influences (especially in the monogastric animals) largely by precipitating dietary proteins and digestive enzymes to form complexes which are not readily digestible (Aletor, 1993). Saponins can either be beneficial or deleterious. There are suggestions that saponin consumption be encouraged because of their hypocholesterolemic activity, forage saponins have been reported by Cheeke *et al.* (1978) to cause toxic and anorexia effects in the rats and swine thereby limiting the feeding value of high saponin animal feeds such as alfalfa. Alkaloids have been reported to have analgesic properties.

The parameters in Table V showed that all whole - testa were negative towards whole seed except in Pp as % of P which was positive towards whole. In the case of whole-dehulled, all were positive towards dehulled. This showed that all the antinutrients in this report were more concentrated in testa and dehulled than in whole seed.

Mineral bioavailability

The Phy: Zn, Ca: Phy and [Ca][Phy] / [Zn] molar ratios are depicted in Table VI. It has been shown that foods with a molar ratio Phy: Zn less than 10 showed adequate availability of Zn and when the ratio was greater that 15, there were problems (Oberleas and Harland, 1981). Phy: Zn molar ratios of 15:1 had been associated with reduced Zn bioavailability (Turnlund *et al.*, 1984). Phy: Zn molar ratios in the whole seed and dehulled samples were less than 10 meaning that Zn would be readily available in the two samples. However, Phy: Zn in testa was much above the recommended value. A Ca: Phy molar ratio lower than 6:1 makes Phy precipitation incomplete so that some of the dietary Zn remains in solution. The proportion remaining in solution increases with decreasing Ca: Phy molar ratios (Wise, 1983). In this present report, all the Ca: Phy molar ratios were lower than the critical level of 6:1. Ellis *et al*. (1987) and Davies and Warrington (1986) indicated that the ratio of [Ca] [Phy] / [Zn] is a better predictor of Zn availability and noted that if the value is greater than 0.5 mol/kg, then, there would be interference with the availability of Zn. In the present report, [Ca] [Phy] / [Zn] values in all the samples were lower than 0.5 mol/kg and this would promote Zn bioavailability.

The differences in the Phy: Zn, Ca: Phy and [Ca] [Phy] / [Zn] molar ratios in the samples are shown in Table VIII. In whole - testa, Ca: Phy was positive for whole seed while the other parameters were positive for testa. In whole-dehulled, all the parameters were positive for whole seed sample.

Conclusion

Bambara groundnut is a good source of proteins and carbohydrates with low levels of most of the major minerals but a good Na/K ratio. The results of the antinutrients showed that

most of the antinutritional factors were concentrated in testa and the [Ca][Phy]/[Zn] molar ratios in the samples would promote Zn bioavailability.

It could therefore be concluded that bambara groundnut should be dehulled when it is being used as food supplement especially for infants. This is because most of the nutrients were concentrated in the dehulled sample while high levels of antinutrients and fibre were concentrated in the testa, the alimentary system of infants are not so developed to digest fibre.

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