









Comparative Studies of some Chemical and Micronutrient Contents in three Sprouted Samples of Bambaranut (*Vigna subterranean* [I] verdc.) Landraces

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Abstract: The research was carried out to evaluate the effect of sprouting on chemical and micronutrient contents of Bambaranut (*Vigna subterranea* [I] verdc.) grown in Kano, Nigeria. Three landraces of Bambaranut (cream, black and zebra) were used for the study. The proximate and mineral contents were analyzed in accordance with the standard methods of analysis. The result of the proximate analysis showed that the moisture and carbohydrate contents reduced significantly ($P < 0.05$) after sprouting while ash, crude protein, crude fat, and crude fiber significantly increased, there was no significant ($P < 0.05$) difference in moisture, crude fiber, and carbohydrate contents between the landraces. The landraces differ in crude protein and fat contents. The results of minerals analysis shows that the landraces differ significantly in Na, Fe, Ca, K, Mg, and Se. However, the amount of Zn, Mn did not significantly differ between the landraces irrespective of sprouting or not. All the three landraces did not differ ($P < 0.05$) in Cu contents. Sprouting leads to decrease in Na, Fe, K, Zn, Mg and Se and increase in Ca and Mn. This study showed that sprouting improves the nutritional quality of Bambaranut irrespective of the landrace.

Keywords: Sprouting, Chemical, Micronutrients, Bambaranut

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1. INTRODUCTION

Bambaranut (*Vigna subterranea* (L.) Verdc) is the third most important grain legume in semi-arid Africa. Resistant to high temperature, drought, insect and weevil attack, bambaranut is suitable for poor soils and has high nutritive value (1). Bambaranut (*Vigna subterranea* (L.) Verdc) has a large number of landraces throughout Africa where small-scale farmers have preserved its genetic diversity on-farm (2). Bambara groundnut is an excellent source macro and micronutrients and has

nutritional potentials that can mitigate malnutrition when properly explored. Therefore, Bambaranut is very important food for poor people especially in Africa who cannot have access to the expensive animal protein. The seed of Bambaranut (Figure 1) is rich in essential amino acids including isoleucine, leucine, lysine, methionine, phenylalanine, threonine, and valine but has not been considered as stable food as cereals such as wheat, maize, and rice (3). Bambara groundnut oil could have medicinal and nutritional benefits, it contains saturated fatty acids that help in lowering the

levels of LDL cholesterol in while increasing HDL cholesterol in blood, this can in turn lower the risk of developing cardiovascular diseases, while providing nutrients, such as vitamin E.

Sprouting is a period characterized by intense metabolic activity in the plant and represents a re-emerging trend in healthy foods and has positive impact on the enhancement of the nutritional properties (4) and taste (5). Germination was reported to increase the nutritional value of cereals and legumes through activation of endogenous enzymes (6). According to the European Sprouted Seeds Association (ESSA) (7) sprouts are the product that results from the germination of seeds and their growth in water or another medium, harvested prior to the development of true leaves and which is set to be eaten whole, including the seed. Consumption of sprouted grains is said to be useful for human health. Positive consumer perceptions about sprouted cereals have resulted in new food and beverage product launches (8). Under ambient conditions, when grains and seeds are soaked and sprouted, endogenous and newly synthesized enzymes begin to alter the seed constituents. Thus, complex macromolecules are broken down into lower molecular weight molecules which are more digestible and more readily absorbed by the body (9). In this work, we determine the effect of sprouting on proximate and mineral composition of three landraces of bambaranut grown in Kano, Nigeria.



Figure 1: Bambaranut seeds.

2. MATERIALS AND METHODS

2.1. Sample Collection and Preparation

Three Bambaranut landraces samples (cream, black, and zebra) were obtained from four areas of Kano state. The sample was sorted, cleaned, washed, and soaked in water overnight. The water was changed at intervals of 3 hours to avoid fermentation (10). The cream and zebra colored samples were soaked for 15 hours overnight whereas the black colored was soaked for 24 hours. The soaked samples were placed in petri dishes for germination period of 5 days. The dishes were covered with cotton to create enabling environment for sprouting to occur. After a period of 5 days, the samples developed shoots (sprouts)

(see Figure 3A, B and C). After sprouting, the sprouted samples were dried and milled. The resulting powdered samples were used for proximate and mineral analysis. The graphical sketch of sprouting are presented in Figure 2.

3. METHODS

3.1. Determination of the Proximate Composition

The proximate composition (moisture, ash, crude protein, crude fat, fiber, and carbohydrate) of Bambaranut was determined in accordance with standard methods of AOAC (11) as follows.

3.2. Determination of Moisture Content

A clean petri dish was weighed (W_1) and 5 g of the powdered sample was placed in the dish and then weighed (W_2). The content was placed in an oven at 120 °C for 3 hours. The dish was removed and cooled in desiccators for 30 minutes and finally weighed (W_3) (14). The moisture content was calculated using the equation below.

$$\%Moisture = \frac{W_2 - W_3}{W_2 - W_1} \times 100$$

3.3. Determination of Ash Content

A clean crucible was weighed (W_1) and 5 g of dried powdered sample was measured in to the crucible and weighed (W_2). The crucible was transferred in to muffle furnace at 550 °C for ashing. After ashing, the furnace was turned off and allowed to cool. The samples were carefully removed from the furnace to avoid losing ash that may be fluffy. The crucibles were transferred to desiccator and allowed to cool for 30 minutes and then weighed (W_3) (13). The ash content was calculated using the following equation

$$\%Ash = \frac{W_3 - W_1}{W_2 - W_1} \times 100$$

3.4. Determination of Crude Protein Content

The crude protein was determined by Kjeldahl distillation as described by (13). Based on the Kjeldahl procedure, proteins and other organic food constituents in a sample are digested using sulfuric acid and with catalysts. The total organic nitrogen is transformed to ammonium sulfate and the digest is neutralized using alkali and distilled into a boric acid solution. The resulting borate anions are titrated using standardized acid, which is converted to nitrogen in the sample. The result of the analysis represents the crude protein content of the food because nitrogen also comes from non-protein components.

3.5. General Procedures and Reactions

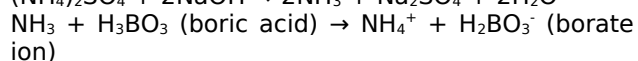
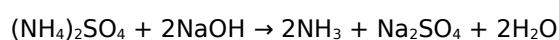
3.5.1. Digestion

The samples were placed in Kjeldahl flask. Acid and catalyst were added to the flask, the mixtures were

allowed to digest formation of clear mixture indicates a complete breakdown of all organic matter in the samples. Non volatile ammonium sulfate was formed in the reaction of nitrogen and sulfuric acid. in the course of digestion, protein nitrogen was liberated leading to the formation of ammonium ions, sulfuric acid oxidizes organic matter and reacts with ammonium that was formed, carbon and hydrogen are converted to carbon dioxide and water.

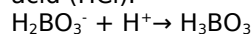
3.5.2. Neutralization and distillation

The digest was diluted using water. Alkali containing sodium thiosulfate was added so that the sulfuric acid is neutralized. The resulting ammonia was distilled into a boric acid solution containing the indicators methylene blue and methyl red.



3.5.3. Titration

Borate ion (proportional to the amount of nitrogen) is titrated with standardized 0.02 M Hydrochloric acid (HCl).



The amount of nitrogen was calculated using the following formula

Moles of HCl = Moles of NH_3 = Moles of N in the sample

A reagent blank is run and reagent nitrogen was subtracted from the sample nitrogen.

$$\%N = \frac{NH_4Cl \times \text{corrected acid volume}}{\text{weight of sample in g}} \times \frac{14 \text{ g of N}}{1 \text{ mole of N}} \times 100$$

Where NH_4Cl = Normality of HCl in moles/1000 mL
Corrected acid volume = (mL of std. acid for sample) - (mL of std. acid for blank)

14 = atomic weight of nitrogen

A factor is used to convert percent N to percent crude protein. Most proteins contain 16 percent N, so the conversion factor is 6.25 (100/16 = 6.25).

$\% N \times 6.25 = \% \text{ protein}$

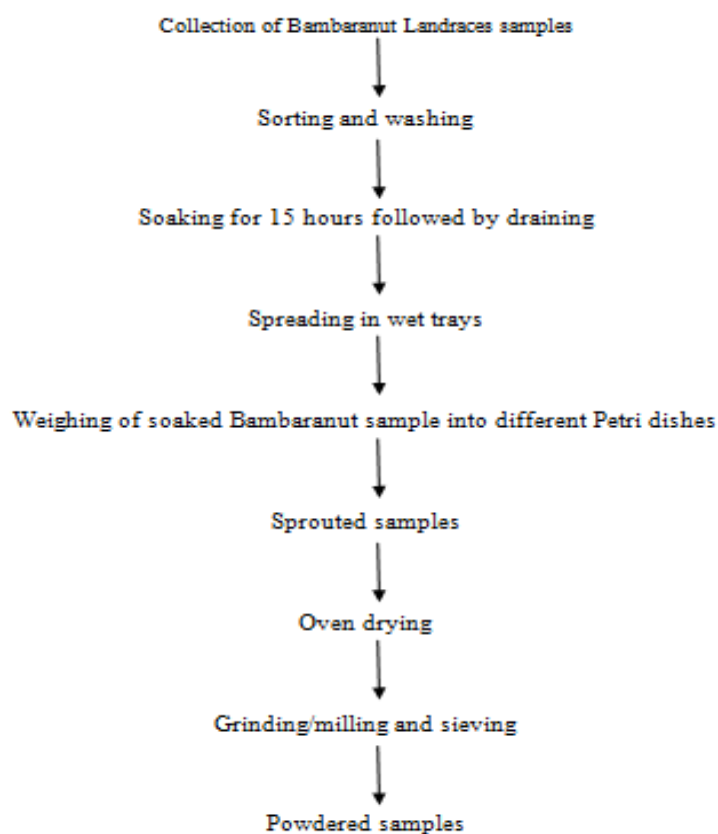


Figure 2: Schematic presentation of sprouting.

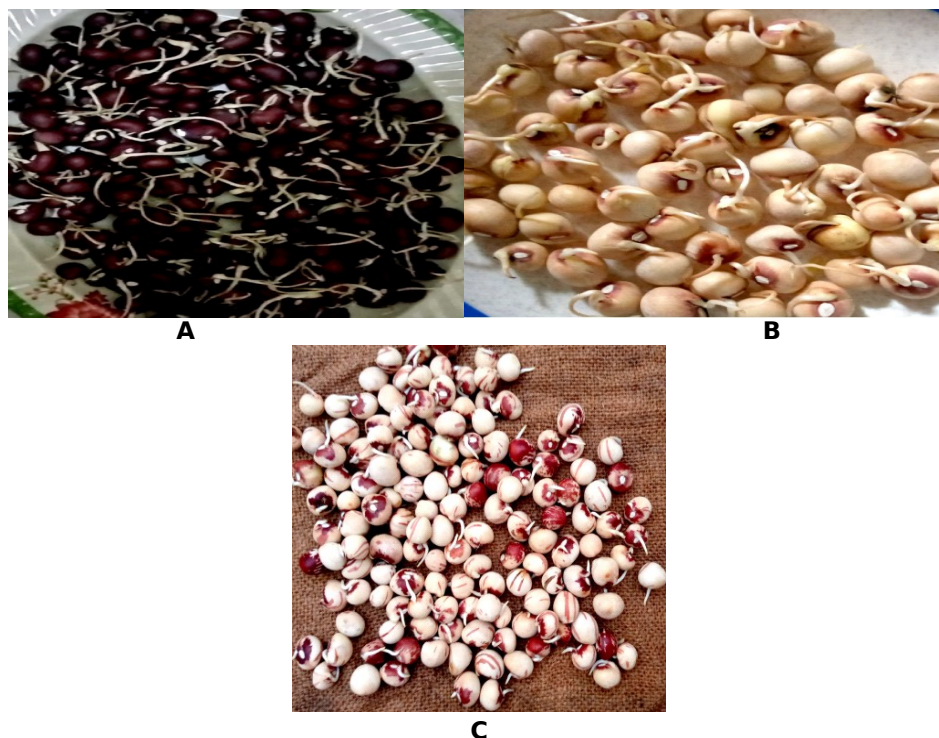


Figure 3: Sprouted Bambaranut (A) Black landrace, (B) Cream landrace (C) Zebra landrace.

3.6. Determination of Crude Fat

3.6.1. Principle

Organic solvents, such as petroleum ether, have the ability to dissolve fat in a given sample. It has a low boiling point (40-80 °C). Therefore when in Soxhlet assembly connected to a reflux condenser, it dissolves the fat in the sample and washes it down in to the flask, and the ether can easily be evaporated and recollected afterwards. The difference in weight between the empty flask before extraction and the weight of flask after extraction gives the weight of lipid being extracted (13).

3.6.2. Procedure

A quantity (3 g) of the sample was carefully weighed (W_1) in to a folded filter paper and small cotton wool placed on top. The content was accurately tied by a thread at both ends of the filter paper and weighed. The content was positioned in the extraction thimble and little cotton wool positioned on top. The whole apparatus was subsequently connected following the addition of 300 milliliters of petroleum ether in to the extraction flask and weighed (W_2). The extraction was maintained for 3 hours by means of the heating mantle and making sure of the incessant flow of water in the condenser. The sample was then removed, air-dried and then positioned in an oven at 80 °C until a constant weight was determined. The extraction flask was transferred to an oven so that the solvent (petroleum ether) can easily be evaporated leaving behind the extract. After evaporation the flask was weighed (W_3). The crude fat in the sample was

calculated using the following equation;

$$\% \text{ crude fat in the sample} = \frac{W_3 - W_2}{W_1} \times 100$$

3.7. Determination of Crude Fiber

Crude fiber is the insoluble and combustible organic residue which remains after the sample has been treated under prescribed conditions of consecutive treatments with light petroleum ether, boiling in dilute sulfuric acid and sodium hydroxide, and washing with boiling water, alcohol and petroleum ether (13).

3.7.1. Procedure

In to the extraction apparatus, 3 g of the sample was weighed and extracted three times using light petroleum ether by stirring, settling followed by decantation. The extracted air dried sample was placed in to a dried 100-milliliter conical flask, 200 mL of 0.127 sulfuric acid was added in to the flask, the sample was dispersed by some of the solution. The content was boiled for 30 minutes, at the same time a stable volume was maintained. The flask was made to rotate every few minutes so as to mix up the contents and get rid of particles from the side. The Buchner funnel was prepared while waiting for the mixture to boil. The Buchner funnel was preset to a perforated plate and linked with the funnel, the filter paper was as well preset to envelop the openings in the plate. The boiled water was added in to the funnel and was allowed to wait until the funnel was hot and afterwards drained by suction. At the end of 30 minutes of the boiling

period, the mixture was allowed to stand for a minute and then poured immediately into a shallow layer of hot water under gentle suction into the funnel. The insoluble matter was washed with boiled water for several times until the washing was free from acid, then it was washed back into the original flask by means of a wash bottle containing 200 mL of 0.313 M sodium hydroxide solution. The mixture was boiled for 30 minutes with the same precautions as those used in the earlier boiling with acid. After 30 minutes of boiling, it was made to stand for a minute and subsequently filtered without delay. The insoluble material was placed in the filter paper using boiling water, after that it was washed using 1% hydrochloric acid, it was finally washed with boiling water making it free from acid. This was subsequently washed two times using alcohol and three times using petroleum ether. The insoluble material was then placed in a dried weighed crucible and dried at 100 °C to constant weight. The crucible with its contents was transferred to a heating mantle in a fume cupboard to remove the organic matter. Subsequently, the crucible was transferred to a muffle furnace at 550°C for 3 hours. The ash content was then obtained by weighing. The crude fiber was calculated using the following equation;

$$\%Crude\ Fiber = \frac{weight\ of\ ash}{weight\ of\ sample} \times 100$$

3.8. Determination of Carbohydrate by Difference

Carbohydrate content was determined by difference as follows;

$$\%Carbohydrate = 100 - (Moisture + Ash) \\ (+ Protein + Lipid + Crude Fiber)$$

3.9. Determination of Mineral Elements Atomic Absorption Spectroscopy (AAS)

Principle

Atomic Absorption Spectrophotometer is an analytical instrument that is based on the principle of atomic absorption spectroscopy. This involves the aspiration of sample solution into the flame and the sample element is changed to atomic vapor. The flame therefore contains atoms of that

element. Some are thermally energized by the flame, but the majority stayed in the ground state. These ground state atoms subsequently absorb radiation of particular wavelengths that is formed by a special source prepared from that element. The absorption follows Beer's Law, that the absorbance is directly proportional to the concentration of the atomic vapor in the flame. That is the concentration of atomic vapor is directly proportional to the concentration of the element in solution being aspirated (16).

3.9.1. Procedure for AAS

The minerals (Ca, Cu, Fe, K, Mg, Mn, Na, Zn, and Se) were determined by Atomic Absorption Spectrophotometer as described by (15). A hollow cathode lamp was selected for the analysis, the lamp was made to warm up for 15 minutes. For the duration of the 15 minutes period, the instrument was aligned by positioning the monochromator at the right wavelength. The monochromator slit width was chosen. The hollow cathode current was as well adjusted in line with the manufacturer's reference. Afterward, the flame was lit and the flow of fuel and oxidant were regulated. The burner and nebulizer flow rate for maximum percent absorption and stability were adjusted after which the photometer was balanced. The samples were aspirated and the concentrations were obtained using calibration curve. A series of standards of the elements in question were run and a calibration curve was constructed by plotting the concentrations of the standards against the absorbance.

3.10. Statistical analysis

Results were analyzed by two-way analysis of variance (Post hoc) using SPSS software. Difference of mean were determined at P<0.05 level of significance.

4. RESULTS

4.1. Proximate Composition

Table 1 presents the effect of sprouting on the proximate contents of three landraces of Bambaranut grown in Kano. The result indicates significant increase in ash, crude protein, crude fat, and crude fiber and decrease in moisture and carbohydrate contents after sprouting.

Table 1: Effect of sprouting on the proximate contents of three landraces of Bambaranut.

Parameter (%)	Sprouted			Unsprouted		
	Black	Cream	Zebra	Black	Cream	Zebra
Moisture	7.10 ± 0.50 ^a	7.42 ± 0.06 ^a	6.81 ± 0.16 ^a	11.99 ± 1.03 ^b	12.01 ± 0.22 ^b	12.09 ± 0.04 ^b
Ash	4.68 ± 0.29 ^a	4.28 ± 0.05 ^b	4.30 ± 0.02 ^a	2.09 ± 0.05 ^c	3.84 ± 0.25 ^d	2.25 ± 0.08 ^c
Crude Protein	17.45 ± 0.56 ^a	19.93 ± 0.13 ^b	15.65 ± 0.37 ^c	4.63 ± 0.07 ^d	10.05 ± 0.08 ^e	6.23 ± 0.05 ^f
crude Fat	14.25 ± 0.59 ^a	14.12 ± 0.08 ^a	15.15 ± 0.07 ^b	11.93 ± 0.61 ^c	3.15 ± 0.32 ^d	9.99 ± 0.57 ^e
Crude Fiber	5.21 ± 0.32 ^a	7.99 ± 0.11 ^b	4.80 ± 0.28 ^c	4.42 ± 0.16 ^d	4.44 ± 0.11 ^d	4.28 ± 0.26 ^d
Carbohydrate	51.3 ± 1.91 ^a	45.39 ± 1.50 ^b	53.33 ± 0.88 ^c	64.89 ± 0.43 ^d	66.46 ± 0.22 ^d	65.13 ± 0.52 ^d

Results are presented as mean ± standard deviation n = 3

Values with different superscript in the same row are significantly different at

P<0.05

Table 2: Effect of Landrace on the Proximate Contents of Bambaranut.

parameters	Landrace			SE
	Black	Cream	Zebra	
Moisture	9.55 ^a	9.71 ^a	9.45 ^a	0.2
Ash	3.39 ^a	4.06 ^b	3.28 ^a	0.07
Crude Protein	11.04 ^a	11.99 ^b	10.94 ^a	0.12
crude Fat	13.09 ^a	8.64 ^b	12.57 ^c	0.18
Crude Fiber	4.82 ^a	6.22 ^b	4.54 ^c	0.08
Carbohydrate	58.10 ^a	55.93 ^b	59.23 ^a	0.45

4.2. Mineral Elements

4.2.1. Mineral contents

The effect of sprouting on mineral contents of

bambaranut is presented in Table 3. Based on the result, sprouting has effect of increasing some of the minerals while decreasing others.

Table 3: Effect of sprouting on Mineral contents of Bambaranut.

Elements (mg/kg)	sprouted			Unsprouted		
	Black	Cream	Zebra	Black	Cream	Zebra
Na	17.2 ± 0.05 ^a	16.89 ± 0.64 ^a	12.88 ± 0.31 ^b	26.17 ± 0.46 ^c	110.93 ± 0.37 ^d	13.92 ± 0.04 ^e
Fe	2.40 ± 0.31 ^a	2.20 ± 0.18 ^a	4.78 ± 0.25 ^b	7.10 ± 0.009 _c	4.06 ± 0.05 ^d	1.79 ± 0.04 ^e
Ca	45.04 ± 0.03 ^a	44.16 ± 0.02 ^b	37.37 ± 0.05 ^c	39.49 ± 0.025 _d	38.31 ± 0.55 ^e	43.2 ± 0.63 ^f
K	519.97 ± 0.41 ^a	551.30 ± 0.60 ^b	518.98 ± 0.22 ^a	551.9 4 ± 0.21 ^c	464.8 ± 0.40 ^d	615.57 ± 0.05 ^e
Zn	1.17 ± 0.11 ^a	1.09 ± 0.04 ^b	1.12 ± 0.001 ^a	1.08 ± 0.004 _b	1.61 ± 0.02 ^c	1.07 ± 0.005 ^b
Mg	102.77 ± 0.30 ^a	89.86 ± 0.31 ^b	106.9 ± 0.20 ^c	106.0 4 ± 0.54 ^c	87.66 ± 0.09 ^d	114.97 ± 0.35 ^e
Mn	0.51 ± 0.007 ^a	0.75 ± 0.05 ^b	0.51 ± 0.03 ^a	0.50 ± 0.04 ^a	0.58 ± 0.02 ^c	0.51 ± 0.02 ^a
Se	38.93 ± 0.35 ^a	30.31 ± 0.51 ^b	24.62 ± 0.01 ^c	32.32 ± 0.55 ^d	29.00 ± 0.20 ^e	36.51 ± 0.03 ^f
Cu	0.40 ± 0.04 ^a	0.31 ± 0.04 ^b	0.30 ± 0.04 ^b	0.32 ± 0.22 ^b	0.29 ± 0.03 ^b	0.33 ± 0.04 ^b

Results are presented as mean ± standard deviation n = 3

Values with different superscript in the same row are significantly different at P<0.05

Table 4: Effect of Landrace on Mineral contents of Bambaranut.

Elements	Sprouted	Unsprouted	SE
Na	15.66 ^a	50.34 ^b	0.13
Fe	3.12 ^a	4.32 ^b	0.04
Ca	42.19 ^a	40.33 ^b	0.12
K	530.08 ^a	544.10 ^b	0.12
Zn	1.13 ^a	1.25 ^b	0.02
Mg	99.84 ^a	102.89 ^b	0.11
Mn	0.59 ^a	0.53 ^b	0.01
Se	31.29 ^a	32.61 ^b	0.12
Cu	0.34 ^a	0.31 ^a	0.13

5. DISCUSSION

5.1. Proximate Contents

The effects of sprouting on the proximate contents of three landraces of Bambaranut are presented in Table 1. Generally, irrespective of sprouting or not, *Vigna subterranea* landraces did not significantly ($P<0.05$) differ in moisture contents but differ in other proximate parameters (Table 2). Similarly, sprouting leads to significant changes in all the proximate parameters irrespective of the landrace. The moisture and carbohydrate contents tend to decrease significantly after sprouting whereas crude protein, ash, crude fiber and crude fat increased significantly. The decrease in moisture content is in disagreement with (19) that sprouting leads to increase in moisture content. The result of this study also disagrees with Shah *et al.* (2011) that sprouting has no effect on moisture content. The decrease in moisture content is an indication that sprouted Bambaranut will have a better shelf life.

Increase in ash contents observed in this study agrees with (17) when chick pea and soybean were sprouted for 2 - 4 days and (18) when mung bean, pea, and lentil were sprouted. The increased ash contents observed indicates that the sprouted bambaranut could have more mineral contents. The decrease in carbohydrate contents during sprouting may have led to the apparent increase observed in ash (18). The apparent increase in protein observed is attributed to increased synthesis of enzyme proteins such as proteases by sprouted seeds or a compositional change due to degradation of other constituents (19). The decrease in fiber contents observed reverberates the findings of (16), the increase could be attributed to the release of nutrients. Previous studies reported a decrease in crude fat contents following sprouting (16). However, the current study has observed significant increase. The significant reduction in carbohydrate contents is in agreement with (16), sprouting cause biochemical changes in carbohydrate that may lead to change in energy value. Hydrolytic enzymes are activated during sprouting leading to the breakdown of starch and non-starch polysaccharides (20). The actions of α - and β -amylases and α -glucosidase result in starch breakdown. These hydrolytic processes produce products (sugars and short-chain carbohydrates) that are perfect for weaning foods as viscosity and digestibility raise along with nutrient absorption.

The sprouted cream bambaranut landrace has the highest protein content, the value $19.93\pm 0.13\%$ is significantly different ($P<0.05$) from those of unsprouted cream landrace as well as the other two landraces (sprouted and unsprouted) (Table 1). Lowest carbohydrate ($45.39\pm 1.50\%$) content was observed in sprouted cream bambaranut landrace, the value differs significantly ($P<0.05$). The

sprouted cream Bambaranut landrace has the highest crude fiber contents ($7.99\pm 0.11\%$) and is significantly ($P<0.05$) different from other landraces irrespective of sprouting or not. Highest crude fat ($15.15\pm 0.07\%$) content was observed in sprouted zebra landrace, the value differs significantly ($P<0.05$) from other landraces. There is no significant difference ($P<0.05$) between fat contents of sprouted black and sprouted cream landrace. The sprouted Black landrace has the highest ash content.

In view of the aforementioned observations, most especially the apparent decrease in moisture contents, increase in ash contents coupled with increased protein and decreased carbohydrate contents, it could be deduced that the effect of sprouting on the proximate parameters of Bambaranut lead to an increases in the nutritional quality. The decrease in moisture content could translate to an increase in shelf life of bambaranut flour. Similarly, increase in protein and the decreased in carbohydrate implies that sprouted bambaranut could be utilized by diabetic patients.

5.2. Minerals

The results of minerals contents of three sprouted and unsprouted landraces of bambaranut are presented in Table 3. The landraces differs significantly in some of the minerals such as Na, Fe, Ca, K, Zn, Mg, Mn, and Se. However, the amount of some minerals did not significantly differ between some landraces irrespective of sprouting or not. For instance the black and zebra landraces have similar Zn and Mn contents. All the three unsprouted landraces have similar Cu contents. Irrespective of the landrace, significant ($P<0.05$) difference was observed in Na, Fe, Ca, K, Zn, Mg, Mn, and Se after sprouting but there was no significant difference in Cu. Overall, sprouting leads to decrease in Na, Fe, K, Zn, Mg and Se and increase in Ca and Mn (Table 4). There is no significant difference ($P<0.05$) in Na and Fe between sprouted black and cream landraces, Zn and Mn between sprouted black and zebra. However, sprouted landraces differed in Ca, K, Mg, and Se. The results of this study showed that the sprouted cream and black landraces have increased in selenium contents while increased in zinc content is observed in black and zebra landraces.

Regulation of the intracellular-extracellular potassium (K^+) gradient is very important for life, acute changes in K^+ plasma levels may lead to fatal consequences. Potassium is a vasodilator, it increases blood flow and decrease in potassium concentration in blood produces vasoconstriction (21). Dietary supplementation and restriction of potassium influence the resistance to blood flow through vascular beds such as cerebral and renal vascular beds (22).

Calcium (Ca) is needed for normal growth, development and maintenance of skeleton (23). Calcium is required in high amount during all stages of life but the requirement is more during growth period for instance childhood, during pregnancy and breast feeding (24). Appropriate intake of calcium causes reduction in hypertension and hypertension disorders related to pregnancy, lowering cholesterol levels, proper muscular contraction, oocyte activation, aiding blood clotting, regulation of heart beat and fluid balance (25). Deficiency of calcium, especially over a long time, can lead to osteoporosis in which the bone deteriorates and there is an increased risk of fractures. Consumption of a well-balanced diet can provide all the necessary nutrients and help prevent calcium deficiency (26).

Magnesium is required for many metabolic reactions as a cofactor, such reactions include protein synthesis, cellular energy production and storage, reproduction, DNA and RNA synthesis, stabilizing mitochondrial membranes among others (27). Magnesium plays a critical role in nerve transmission, cardiac excitability, activation of the immune system, neuromuscular conduction, muscular contraction, vasomotor tone, blood pressure, and glucose and insulin metabolism. Magnesium has multiple functions within the body, therefore potassium plays a major role in disease prevention and overall health, hence the role of magnesium is essential (27). Deficiency of magnesium is associated with a number of chronic diseases such as migraine headaches (28), Alzheimer's disease (29), stroke and hypertension, cardiovascular disease, and type 2 diabetes mellitus (30). Magnesium is found to influence fetal programming and disease presentation in child and adulthood (27) and it is indispensable for the aggregation of ribosomes into polysomes. To be biologically active and be synthesized from ADP, ATP requires magnesium (31).

Manganese is essential micronutrients in human that is principally obtained from the diet (32). It functions primarily as a coenzyme in many biological processes, such as macronutrient metabolism, bone formation, free radical defense systems, and in the brain ammonia clearance and neurotransmitter synthesis. It is found in all tissues and is a critical component of many proteins and enzymes (33). An association has been reported between dietary Mn and glucose metabolism, insufficient dietary Mn impairs insulin secretion and glucose metabolism. However, supplementation of Mn modifies the enzyme profiles of carbohydrate metabolism and improves high-fat-diet-induced beta cell injury and insulin resistance in animal models of diabetes (34). Manganese is a component of metalloproteins such as arginase, acetyl-CoA carboxylase, phosphoenolpyruvate decarboxylase, and pyruvate carboxylase, Mn superoxide dismutase (a mitochondrial

antioxidant), glutamine synthetase and glycosyltransferases (33). Human milk is generally low in Mn (1.8–27.5 µg/L); but concentrations in infant formulas can vary dramatically (33–300 µg/L) (54).

Adequate provision of Fe is essential for the functioning of many biochemical processes including electron transfer reactions, gene regulation, binding and transport of oxygen, and regulation of cell growth and differentiation, DNA synthesis and healthy function and development of brain (35). In a global scale, Fe deficiency is the most prevalent micro nutrient deficiency and a major public health risk in developing and industrialized countries. The deficiency affects more than a billion people of different age groups around the world, it is the common cause of anemia and a common deficiency among non anemic children, especially among children of developing countries (36). Deficiency of iron causes impaired immune function, impaired mental function, impaired physical performance, complications of pregnancy, increased absorption of lead and cadmium, altered drug metabolism, increased insulin sensitivity and fatigue (35).

The increase in selenium observed in cream and black landrace is of interest, selenium is a micronutrient that is necessary in trace amounts for the proper functioning of organisms (37). It has a key importance for maintaining homeostasis of the human system, especially for the proper functioning of the immune system. As a component of glutathione peroxidase, selenium is essentially involved in the protection of cells against the effect of excess H₂O₂, and also plays role in heavy metal detoxification. Selenium is required for proper functioning of the thyroid gland, it induces the occurrence of the selenoprotein synthesis (a process involved in the antioxidant defense mechanism of the organism) and it possesses anticarcinogenic properties against some cancers as well as anti-diabetic activities and reproductive efficiency (38).

Copper has multiple metabolic functions, severe deficiencies are associated with cardiac, bone, immune, and central nervous system problems and must be acquired through the diet and trafficked to the organs, cells, and proteins requiring copper for health (39). It is an indispensable cofactor used for redox reactions involving copper containing oxidases. Copper enzymes regulate various physiologic pathways such as energy production, iron metabolism, connective tissue maturation, and neurotransmission (40). Dietary copper deficiency or overload leads to dysregulation of lipid metabolism.

Zinc is essentially involved in the regulation of many physiological and biochemical events in the organism such as maintenance of integrity of cell

membrane, metabolism of protein, carbohydrate and lipid, recovery from wound injury and in the regulation of a number of other biological processes associated with normal growth and development (41). Zinc acts as a cofactor of many enzymes, at least one enzyme in every enzyme classification (42). Deficiency of Zn in children causes an increase in infection and diarrhea leading to the death of about 800,000 children worldwide yearly (43). Zinc deficiency was also indicated as a risk factor for immune deficiency and hence susceptibility to infection in the elderly (41). Depletion of zinc stores has been reported to be a risk factor of pneumonia in the elderly. Zinc is being considered as prophylactic or adjunct therapy for COVID-19, with 12 clinical trials underway, highlighting the relevance of this trace element for global pandemics (44). Zinc is a cofactor of at least one enzyme in every enzyme classification (43).

6. CONCLUSION

This study showed that sprouting enhances the nutritional quality of bambaranut irrespective of the landrace and could be used to increase the shelf life of foods. Similarly, sprouting could be used in healthy diet preparations. The increase in protein and decreased in carbohydrate contents is an indication that sprouted bambaranut could be utilized by diabetic patients.

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