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Proximate analysis of *Sphenostylis stenocarpa* and *Voadzeia subterranean* consumed in South - Eastern Nigeria

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Proximate analyses of two traditional grain legumes consumed in Eastern Nigeria- Bambara groundnut (*Voadzeia subterranean*) and African yam beans (*Sphenostylis stenocarpa*) were carried out. Bambara groundnut was found to contain $2.86 \pm 0.02\%$ moisture, $32.40 \pm 0.02\%$ protein, $7.35 \pm 0.02\%$ fat, $5.78 \pm 0.02\%$ ash, $2.68 \pm 0.02\%$ crude fiber and $51.78 \pm 0.02\%$ total carbohydrates. African yam beans contained following: $1.96 \pm 0.02\%$ moisture, $37.21 \pm 0.02\%$ proteins, $9.49 \pm 0.02\%$ fat, 5.35 ± 0.02 Ash, 3.55 ± 0.02 crude fiber and $44.4 \pm 0.02\%$ total carbohydrates. The result indicated that the two underutilized food legumes have high protein content with African yam beans having slightly higher value of the protein nutrient. They also contained varying levels of ash content, crude fiber and carbohydrates. This revealed that bambara groundnut and African yam beans are important food sources that can be exploited particularly in the developing countries where there is shortage in animal protein and undernutrition facing many families in developing countries.

Key words: Bambara groundnut, African yam beans, proximate analysis, undernutrition, protein.

INTRODUCTION

Currently the need for sustainable development and economic recovery in the developing regions of the world is crucial particularly in the area of access to quality proteinous food and food security. Conversely, the commonly relied source of quality protein which is animal protein is grossly expensive and scarce (Adebowale and Adebowale 2007; Lawal and Adebowale, 2006, Adebowale et al., 2005a, b; Lawal et al., 2005). Thus, legumes are rich sources of plant proteins present as a substitute for the scarce animal proteins. Of the thousand known legumes species, less than 20 are used extensively today as food. Those commonly used include: peanuts, soybeans, peas, lentils, pigeon pea, bambara groundnut and African yam beans. Oils, proteins, gums, resins and medicinal compounds could be derived from this extremely large family. Some leguminous seeds or pulses contain 20 to

40% protein on dry matter basis dry weight, which is nearly twice the value found in cereals and 20 times that of cassava. Only a few grain legumes contain up to 50% proteins. Pulses also contain up to 40 to 60% carbohydrates mainly in the form of starch and some reasonable quantities of niacin, thiamine, riboflavin and ascorbic acid. Pulses are rich in calcium and iron (Maneepun, 2000; Emenalom and Udedibie, 1998).

Bambara groundnuts (*Voadzeia subterranean*) and African yam beans (*Sphenostylis stenocarpa*) are leguminous crops which are cultivated in tropical and subtropical regions. Bambara groundnuts (*V. subterranean*) belong to the family *Fabaceae* which are conspicuously West African crops. Bambara groundnuts are underutilized or neglected due to storage induced defects and antinutritional factors (toxins and biological active substances) such as trypsin inhibitor, haemoagglutinins, cyanogenic glycosides, anticoagulant, toxic histones (Fadahunsi and Sanni, 2010; Agbede and Aletor, 2003; Vasconcelos et al., 1997). However, most of these toxins are reduced to tolerable levels by simple preparative

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procedures such as fermentation, germination, roasting, soaking in warm water and thorough cooking which usually requires high fuel input and prolonged cooking (Oluwole and Taiwo, 2009). Groundnuts present good agronomic advantages such as their ability to thrive in drought weather, low nutrient soils and high nutrition potential (Okpuzor et al., 2010; Anchirinah et al., 2001; Azam –Ali et al., 2001). Cultivars are recognized in Africa differing in shape of leaves, sizes, hardness and color of the seed. Bambara groundnuts are known as jugo beans in Swahili. In Zambia, Bambara groundnuts are known as ntoyoy, *katoyo* or *mbwiila*. In Nigeria, Hausas refer to it as "Gurjiya" or "Kwaruru". Amongst, the Goemai and Kanuri groups in Nigeria, it is referred to as "Kwam" and "Ngamgala" respectively. Bambara groundnut (*V. subterranean*), is a nutritionally balanced seed containing essential amino acids such as isoleucine, leucine, lysine, methionine, phenylalanine, threonine and valine (Ihekoronye and Ngoddy, 1985). According to findings by Adu-Dapaah and Sangwan (2004), the seed of the crop is rich in iron and essential amino acids. Bambara groundnuts are grown for their edible seeds which are used as a nutritional pulse and not as an oil seed due to its low percentage of oil (Minka and Bruneteau, 2000). The fresh seeds are eaten in an unripe state or the pulse, after soaking and boiling as the dried seeds are sometimes hard. The dried seeds are sometimes roasted and ground into flour for preparation of different delicacies (Poulter and Caygill, 2006; Brough et al., 1993).

African yam beans (*S. stenocarpa*) are cultivated both for the seeds and tubers, its valuable and prominent source of plant protein (amino acids –lysine and methionine) in the diets of consumers (Evans and Boulter, 1974). The tuber which is found beneath the ground has resemblance with sweet potatoes or Irish potatoes. Above ground it produces good yields of edible seeds. It could be found in forests, open wooded grasslands, rocky fields, and marshy grounds as weed and cultivated crops (Porter 1992). It grows on a wide range of soils including acid and highly leached sandy soils at altitudes from sea level to 1950 m (Amoae et al., 2000). In the area of cultivation, it serves as a security crop prior to the new season harvest of main staples such as maize, sorghum, yam and cassava in the tropics. Different studies on nutritional, chemical composition and extraction condition for optimum solubilization of the pulse protein have been studied (Aletor and Aladetimi 1989; Edem et al., 1990; Kine et al., 1991; Akinmutini et al., 2007; Eromosele et al., 2008). African yam beans (*S. stenocarpa*) however have antinutritional factors such as phytin and lectin. The large scale breeding and research on these leguminous crops (Bambara groundnut (*V. subterranean*) and African yam beans (*S. stenocarpa*) has been rather slow. However, evidence and fragmentary research results suggested that it is a crop with much promise. The authors seek to ascertain the proximate composition of these leguminous crops found in the South eastern part of Nigeria with the view of

documenting and exploiting them as alternative sources of protein for the emerging population in this zone.

MATERIALS AND METHODS

Crop physiology

Bambara groundnut (*V. subterranean*) is an annual herb with short creeping, much branched stems rooting at nodes and with very short internodes thus, giving a bunchy appearance to the plant. Leaves are pinnately trifoliate, petiole long erect, grooved and thickened at base. Seeds are round, smooth, and hard varying in size up to 1.5 cm broad, whitish red, black or variously mottled or blotched hilum white, sometimes surrounded by black eye. The flowers are self pollinated and are cleistogamous. It is grown from seeds either in mixed cultivation or in pure stands with spacing about 12 × 8 inches and matures in 4 months. It is harvested by pulling up the entire plants to which the pods are adhered by tough wiry stalks. The normal yield is about 500 to 1000 seeds per acre.

African yam beans

S. stenocarpa is described as a prostrate or a climbing vine reaching up to 3 m or more in height (Duke et al., 1977). The stems are reddish glabrous or sparsely pulverous. The plant produces spindle shaped starchy tubes smaller in size than those of sweet potato. It takes between 5 to 8 months to mature.

Collection and preparation

Bambara groundnut (*V. subterranean*) and African yam bean (*S. stenocarpa*) seeds were obtained from a local market (new market) in Enugu, South East of Nigeria. The varieties selected were white black eye typed for the Bambara groundnuts and the greenish type for the yam beans. The seeds were identified and authenticated by a crop scientist and deposited at the department of biochemistry Enugu State University, Enugu. Five hundred and fifty grams (550 g) each of the purchased seeds were washed with distilled water to remove impurities and air dried on aluminum trays to achieve total dryness before pulverization.

Bambara groundnuts (*V. subterranean*) and yam bean (*S. stenocarpa*) seeds were pulverized into powdered flour using a mechanized crown grinding machine Model PHT 94, manufactured in China. This was sieved and used for further work as test samples.

Chemicals and reagents

All chemicals and reagents used were of analytical grade. Petroleum ether spectrophotometric grade was purchased from Merck. Sodium hydroxide, tetraoxosulphate (VI) acid, boric acid and sodium sulphate were sourced from Fisher scientific, US.

Determination of proximate analysis

The test sample Bambara groundnut (*V. subterranean*) and African yam beans (*S. stenocarpa*) were subjected to proximate analysis by the methods outlined by AOAC (1984).

Determination of moisture content

This method is based on moisture evaporation. Here, the aluminum

dishes were washed dried in oven and desiccators for cooling. The weight of each dish was taken. 200 g of ground samples of Bambara groundnut (*V. subterranean*) and African yam beans (*S. stenocarpa*) were weighed into a sterile aluminum dish, weight of the dish and weight of undried sample (in duplicate) were taken. This was transferred into an oven set at 80°C for 2 h and at 100°C for 3 h respectively. This was removed and cooled in desiccators. Then the weight was measured using a measuring scale balance. It was transferred back into the oven for another one hour and then reweighed. The process continued until a constant weight was obtained. The difference in weight between the initial weight and the constant weight gained represented the moisture content. The loss in weight multiplied by 100 over the original weight is percentage moisture content. The formula used is presented as:

$$\text{Moisture content (g/100g)} = \frac{\text{Loss in weight (W2-W3) (g)}}{\text{Original weight of sample (W2-W1) (g)}} \times 100$$

Where W1= initial weight of empty crucible, W2 = weight of crucible + food before drying, W3 = final weight of crucible + food after drying.

$$\% \text{ Total solid (Dry matter) (\%)} = 100 - \text{moisture (\%)}$$

Ash content

The ash represents the inorganic component (minerals) of the sample after all moisture has been removed as well as the organic material. The method is a destructive approach based on the decomposition of all organic matter such that the mineral elements may be lost in the process. Twenty grams (20 g) of each of the samples of Bambara groundnut (*V. subterranean*) and African yam beans (*S. stenocarpa*) were weighed into a clean dried and cooled platinum crucible. It was put into a furnace set at 550 to 600°C and allowed to blast for 3 h. It was then brought out and allowed to cool in a desiccator and weighed again. Percentage weight is calculated as weight of ash multiplied by 100 over original weight of the samples used. The formula used is presented as:

$$\text{Ash content} = \frac{\text{Weight of ash (g)}}{\text{Weight of original sample used}} \times 100$$

$$\frac{\text{Loss in weight (W3-W1) (g)}}{\text{W2-W1}} \times 100$$

Where W1 = weight of empty crucible, W2 = weight of crucible + food before drying and or ashing, W3 = weight of crucible + ash.

Determination of lipid content

The method employed was the Soxhlet extraction technique described by Shir law, (1967). 15 g of the samples were weighed and carefully placed inside a fat free thimble. This was covered with cotton wool to avoid the loss of sample. Loaded thimble was put in the Soxhlet extractor, about 200 ml of petroleum ether were poured into a weighed fat free soxhlet flask and the flask was attached to the extractor. The flask was placed on a heating mantle Such that the petroleum ether in the flask refluxed. Cooling was achieved by a running tap connected to the extractor for at least 6 h after which the solvent was completely siphoned into the flask. Rotary vacuum

evaporator was used to evaporate the solvent leaving behind the extracted lipids in the soxhlet. The flask was removed from the evaporator and dried to a constant weight in the oven at 60°C. The flask was then cooled in a desiccator and weighed. Each determination was done in triplicate. The amount of fat extracted was calculated by difference. The formula used is presented as:

Ether extracts (100g) dry matter=

$$\frac{\text{Weight of extracted lipids (g)}}{\text{Weight of dry sample (g)}} \times 100$$

Protein determination

Total protein was determined by the kjeldahl method as modified by Williams (1964). The analysis of a compound of its protein content by kjeldahl method is based upon the determination of the amount of reduced nitrogen present. About 20 g of the samples Bambara ground nut (*V. subterranean*) and African yam bean (*S. stenocarpa*) were weighed into a filter paper and put into a kjedahl flask, 10 tablets of Na₂SO₄ were added with 1 g of CuSO₄ respectively. Twenty milliliter (20 ml) of concentrated H₂SO₄ were added and then digested in a fume cupboard until the solution becomes colorless. It was cooled overnight and transferred into a 500 ml flat bottom flask with 200 ml of water. This was then cooled with the aid of packs of ice block. About 60 to 70 ml of 40% of NaOH were poured into the conical flask which was used as the receiver with 50 ml of 4% boric acid using 3 days of screened methyl red indicator. The ammonia gas was then distilled into the receiver until the whole gas evaporates. Titration was done in the receiver with 0.1 M NH₂SO₄ until the solution became colorless. The formula used is presented as:

The percentage protein =

$$\frac{V_s - V_b \times 0.01401 \times N \text{ acid (6.25)}}{\text{Original wt of sample used}} \times 100$$

Where V_s = Volume (ml) of acid required to titrate sample; V_b = Volume (ml) of acid required to titrate blank; N acid = normality of acid

Crude fiber

The bulk of roughages in food is referred to as fiber and is estimated as crude fiber. Twenty grams (20 g) of *V. subterranean* and (*S. stenocarpa*) of the different samples were defatted with diethyl ether for 8 h and boiled under reflux for exactly 30 min with 200 ml of 1.25% H₂SO₄. It was then filtered through cheese cloth on a fluted funnel. This was later washed with boiling water to completely remove the acid. The residue was then boiled in a round bottomed flask with 200 ml of 1.25% sodium hydroxide (NaOH) for another 30 min and filtered through previously weighed couch crucible. The crucible was then dried with samples in an oven at 100°C, left to cool in a desiccator and later weighed. This was later incinerated in a muffle furnace at 600°C for 2 to 3 h and later allowed to cool in a desiccator and weighed. The formula used is presented as:

Calculation = Weight of fiber = (C2-C3) y

$$\% \text{ fiber} = \frac{\text{C2-C3}}{\text{Wt. of original sample}} \times 100$$

Table 1. Proximate compositions of Bambara groundnut and African yam bean.

Nutrient	(<i>Voandzeia subterranean</i>)	(<i>Sphenostylis stenocarpa</i>)
Moisture	2.86 ± 0.02	1.96 ± 0.02
Protein	32.40 ± 0.02	37.21 ± 0.02
Fat	7.35 ± 0.02	9.49 ± 0.02
ash	5.78 ± 0.02	5.35 ± 0.02
Crude fiber	2.68 ± 0.02	3.55 ± 0.02
Carbohydrates	51.79 ± 0.02	44.4 ± 0.02

Table 2. Percentage total of proximate compositions of Bambara groundnut and yam bean.

Nutrient	(<i>Voandzeia subterranean</i>)	(<i>Sphenostylis stenocarpa</i>)
Moisture	2.86 ±0.02	1.96±0.02
Protein	32.40±0.02	37.21 ±0.02
Fat	7.35±0.02	9.49 ±0.02
Carbohydrates	51.79±0.02	44.4 ±0.02
Total (%)	94.40±0.02	93.06±0.02

Carbohydrate determination

Available carbohydrate (%), = 100 - (protein (%) + Moisture (%) + Ash (%) + Fibre (%) + Fat (%). Here, the different samples of Bambara ground nut (*V. subterranean*) and African yam bean (*S. stenocarpa*) were digested with perchloric acid and hydrolyzed starch. The soluble sugar was determined colorimetrically by anthrone method. Duplicate standards were pipetted out using 1 ml dilute glucose standard solutions and 5 ml of freshly prepared anthrone reagents pipetted rapidly to all the tubes and mixed thoroughly. These covered tubes were placed in boiling water bath for exactly 12 min to allow change in color following which sample was allowed to cool at room temperature. The readings of the two cuvettes containing anthrone reagents and deionized distilled water were measured, and difference in readings noted for correction in the calculations. The prepared solutions were transferred to one glass cuvette and their absorbance read at 630 nm against a blank containing 1ml of deionized distilled water and anthrone reagents. Concentrations of the test samples were obtained via interpolation of the absorbance readings. The mass of the glucose was obtained by calculation of the concentration and dilution made. The mass of starch was obtained by calculating the mass of glucose and a factor of 0.9

RESULTS AND DISCUSSION

The proximate analysis for the legumes is shown in (Tables 1 and 2) and (Figures 1 and 2). In developing countries, there is an increasing mismatch between the rate of population growth and the rate of food production. However, disastrous consequences such as under nutrition often accompanied by diseases such as kwashiorkor or death result, when there is insufficient supply of protein foods. Furthermore, it is known that the brain reaches 80 to 90% of its full weight by age of four; protein malnutrition during this critical period when neurons and neural connections are being formed may cause irreversible brain damage leading to low IQ. This

may persist throughout life and may not be remedied by later corrective measures. Women and children in developing countries are more prone to this challenge. The issue of malnutrition alleviation can simply be dealt with by proper understanding and awareness of the food we eat, thus, this research work was aimed at evaluating the nutritive value of the neglected and under-utilized crops in sub Saharan Africa (Bambara groundnut (*V. subterranean*) and African yam beans (*S. stenocarpa*) and to assess their potentials in dealing with protein under- nutrition prevalent in the Eastern Nigeria and other parts of the world. From research studies, legumes are plant foods which are nutritionally rich in high protein content which is twice the percentage found in cereals and thrice of such as found in cassava. However, plant proteins are generally nutritional inferior to those from animal sources due to the protein rich portions of the grains being surrounded by protective husks cellulose and other polysaccharides that are not hydrolyzed by intestinal enzymes. Conversely, high costs of animal protein have made it unavailable in some parts of the world. The combinations of plant proteins can provide a completely adequate and balance mixture of essential amino acids. The content of the essential amino acids is therefore very essential in nutrition, where it provides precursors for protein biosynthesis needed for synthesizing the proteins needed for the maintenance, repair and growth of tissues. From the results in Table 1 the composition of the two legumes showed that they are good sources of protein with the protein content accounting for 32.40 ± 0.02% and 37.21±% for Bambara groundnut (*V. subterranean*) and Africa yam beans respectively. This result compares well with the reports of Okpuzor et al. (2010) who identified a total of ten and twelve different proteins in the malted and dry seeds of

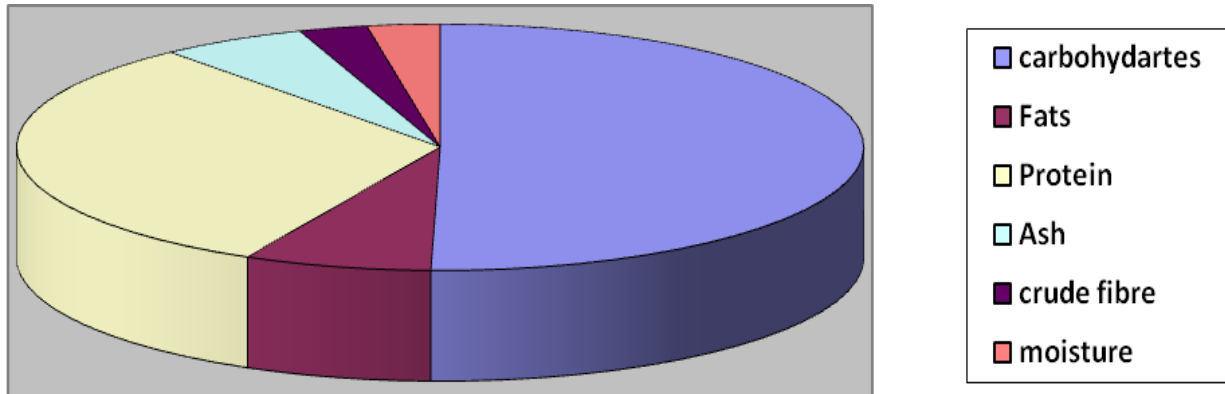


Figure 1. Pie chart represents proximate analysis for Bambara groundnut (*Voadnzeia subterranean*).

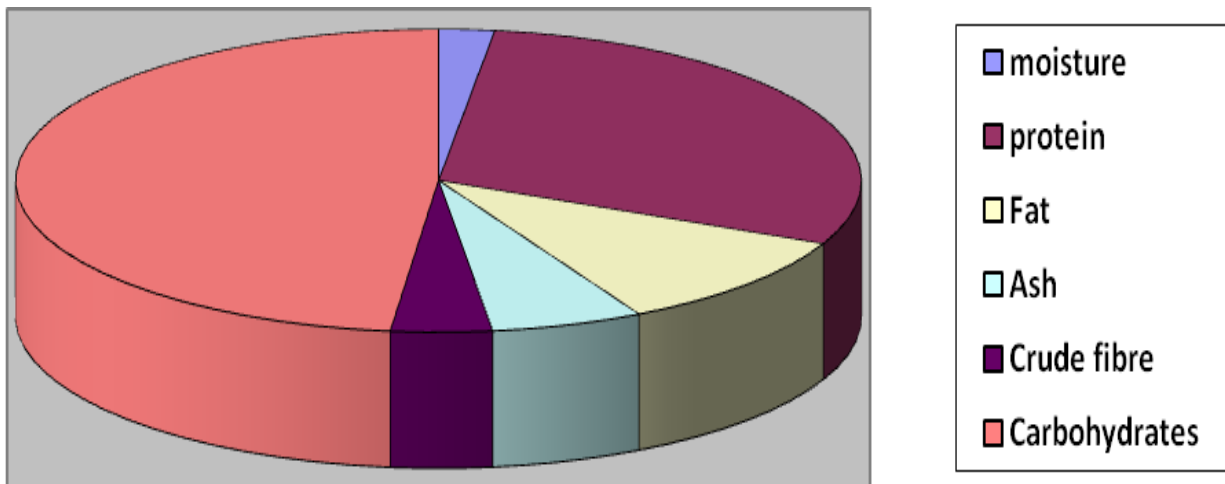


Figure 2. Pie chart represents proximate analysis of African yam bean (*Sphenostylis stenocarpa*).

Bambara groundnut (*V. subterranean*). This shows that the seeds have a great potential in alleviating chronic protein deficiency syndromes that are widespread in developing countries such as Nigeria. According to Adu-Dapaah and Sangwan (2004), the seed of Bambara is regarded as a balanced food because when compared to most food legumes, it is rich in iron with high lysine and methionine content. Minka and Bruneteau (2000) reported that Bambara groundnuts (*V. subterranean*) contain about 63% carbohydrates, 18% oil and the fatty acid composition is predominantly linoleic, palmitic and linolenic acids. From our results in (Table 1 and Figures 1 and 2) on the content of carbohydrates and fat in the Bambara (*V. subterranean*) were 51.79 and 7.35% for African yam bean (*S. stenocarpa*) 44.4 and 9.49%. This showed that there are marginal differences between the two legumes in terms of the content of carbohydrates and fats. On comparing the results of the analysis reported by Minka and Bruneteau (2004) on Bambara seed, the difference in result may be due to the processes involved with the

handling of the seeds during preparation of different foods from the seeds. In line with this fact, Okpuzor et al. (2010) reported about ten different proteins in the malted seed while the dry seeds were twelve different proteins. The deflection in the values may be due to some contributory factors which may include the germination of the seeds; where part of the protein may have been utilized by the growing embryo. It could also be different in mineral content of the soil, climatic factors and the presence of tubercles or nodules of roots that contain nitrogen-fixing bacteria. However, in these observations, the two legume plants studied are relatively very cheap and readily available compared to other legume and protein sources. The nutritional benefits derivable from these legumes can be harnessed by reduction of anti-nutrient contents and formulation of infant foods and diet (Brough et al., 1993; Poulter and Caygill, 2006). According to Adeyeye and Agesin (1997), (*S. stenocarpa*) has a high mineral content which is needful for sustainable health of human life. The mineral composition include:

iron, copper, manganese, phosphorus and calcium. Recent findings reported that seed proteins of bambara (*V. subterranean*) have been implicated in anti-hypertensive activities (Yamada et al., 2008); regulation of food intake and body weight in humans, (Anderson and Moore, 2004) and the improvement in plasma and liver lipid profile of rats fed with high cholesterol diet (Yang et al., 2007). The ash content reveals the degree of purity and mineral content of the grains. The low water content of 2.86 and 1.96% for Bambara groundnut (*V. subterranean*) and African yam beans (*S. stenocarpa*) showed that the grain has a high storage potential. Thus, the rate of deterioration of the seeds will be low in view of the little moisture level. Thus, the consumption of the leguminous foods rich in the essential minerals could be an alternative to the solution of protein- energy malnutrition and preference to animal fats which is believed to predispose people to atherosclerosis and coronary heart diseases due to high content of cholesterol. Bambara groundnuts (*V. subterranean*) and African yam beans (*S. stenocarpa*) still exhibit great potential as cash crops which are grown in mixed association with other root tubers. From this study, Bambara groundnuts (*V. subterranean*) and African yam beans (*S. stenocarpa*) offers nutritional and health benefits to the ever –increasing populations of the developing world.

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