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Macronutrients and Micronutrients Profile of Some Underutilized Beans in South Western Nigeria

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Authors' contributions

This work was carried out in collaboration between all authors. Authors GOLA, AOD, AOAO and LNO designed the study, author GOLA wrote the protocol and supervised the work. Authors GOLA and OTE carried out all laboratories work and performed the statistical analysis. Author OTE managed the analyses of the study. Author OTE wrote the first draft of the manuscript. Authors GOLA and OTE managed the literature searches and edited the manuscript. All authors read and approved the final manuscript.

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ABSTRACT

Beans are a rich source of nutrients in human diet. However a number of edible bean varieties are largely underutilized in developing countries due to little or no information on their nutritional composition. The present study investigated the nutritional content of mung beans, African yam beans, soybeans, black eyed peas and pigeon peas from parts of South Western Nigeria. Samples were collected randomly in duplicates across the six South-western states of Nigeria. Common beans was included as a reference for comparison. Standard methods were used to determine the proximate composition of all bean samples. Mineral nutrients and phaseolin protein fractions

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(albumin, globulin and prolamine) of bean samples (excluding pigeon pea varieties) was also determined. The proximate, mineral and phaseolin protein contents differed significantly (P<.05) between bean types. Highest protein, fat, carbohydrate, crude fibre and ash content was in African yam bean (24.96%), mung beans (6.60%), soybean (62.81%), mung beans (15.24%) and African yam beans (4.30%), respectively. The beans compared fairly with common beans in proximate composition. Mineral nutrients differed significantly (P<.05) between bean types. Calcium, magnesium, phosphorus and potassium (particularly in Mung beans) were in high amounts. Black eyed peas had the lowest total of mineral content while mung beans had the highest. The phaseolin protein fractions were significantly different (P<.05) among bean types. African yam bean had the highest albumin and globulin content (%/mg protein) of 41.89 and 35.70 respectively, while prolamine was highest in soybeans. These results indicated that these underutilized beans compares favourably in terms of nutritional composition with widely consumed common beans in Nigeria. In addition, African yam beans and Mung beans are equally suitable alternatives from a protein-rich standpoint.

Keywords: Underutilized beans; legumes; nutrients; phaseolin protein; Nigeria.

1. INTRODUCTION

Beans are a rich source of essential nutrients in human diet, providing proteins, complex carbohydrates, vitamins, mineral elements, dietary fibres and complex carbohydrates, while being low in fats, sodium and cholesterol-free [1-6]. On a global scale, beansis the most important legume staple for human consumption [4,7]. Their high protein content makes them relatively cheap alternative protein source compared to the highly priced animal or meat-protein sources for households in developing countries [8]. In Nigeria, a number of beans and beans varieties are cultivated. These include varieties of the common beans (*Phaseolus vulgaris*); African yam bean (*Sphenostylis stenocarpa*); mung beans (*Vigna radiata*); soybeans (*Glycine max*); pigeon pea (*Cajanus cajan*); African oil bean (*Pentaclethra macrophylla*) and the black eyed pea (cowpea) (*Vigna unguiculata*) [2,8-12]. With the exception of common beans and cowpea, a number of these bean types are largely underutilized, especially for human consumption. At present a number of these bean seeds are processed into flours and utilized as food or feed [8,13]. Their uses include, the fortification of indigenous starchy meals as composites [14-18]; production of flavouring fermented food condiments used in the preparation of traditional dishes [19-21]; beverage (soy-milk) and cake (soy-cake, tofu) [22,23].

As a specific example, the utilization of Mung bean for human consumption in Nigeria is still very low when compared to its uses and wide adoption rate in other parts of the world, including India and Asia [24]. This is due to the fact that it was recently introduced into the agro-ecological sphere of Southern Nigeria [25-27]. Nevertheless, the mung bean is an important legume cultivated for human consumption (as either fresh sprouts or dry beans) [24]. However, awareness of its nutritive potentials are required in order to increase its current low adoption rate and consumption in Nigeria [27]. Similarly, the African yam bean, Pigeon pea and black eyed pea are all under exploited despite their rich nutritional resource [28-32]. Diversification in the use of the beans can stimulate an increased cultivation and supply of these bean types. Furthermore, the increased utilization of these beans in human nutrition may contribute to achieving the targeted daily dietary protein intake recommended by the World Health Organization [33], especially in poor developing nations. This is essential to attaining sustainable food security in terms of nutritional quality.

The poor utilization of these beans can be attributable to a lack of adequate characterization and documentation of the nutritional profile of these bean seeds, as well as a lack of documentation of the genotypic accessions available in the Nigerian agricultural sphere [11, 34,35]. Hence, we set out to characterize some of the underutilized beans types, including mung beans, pigeon pea, African yam bean and soybean in parts of South Western Nigeria in terms of nutritional, chemical and genotypic content. However, the first two aims are the focus of the current article. Here, we determined the macronutrients, micro nutrients or minerals and the phaseolin protein fractions (major seed storage protein) content of the bean types.

2. MATERIALS AND METHODS

2.1 Sample Collection

Six bean varieties including the African yam bean (*Sphenostylis stenocarpa*), mung bean (*Vigna radiata*), red and grey colour varieties of the pigeon pea (*Cajanus cajan*), black eye pea, pigeon pea (greyish colour variety) and soybean (*Glycine max*) were collected from parts of South Western Nigeria. Samples were collected in clean polythene bags and stored at 4°C prior to analysis.

2.2 Determination of Proximate Composition

The proximate compositions: ash, moisture, fat, carbohydrate, crude protein (Kjeldahl method) and were determined by standard methods of the Association of Official Analytical Chemists (A.O.A.C) [36]. Carbohydrate was obtained by subtracting the sum of other proximate parameters from 100.

2.2.1 Determination of reducing sugar

Reducing sugar was determined according to the method of Somogi-Nelson [37]. To 1 ml of bean sample powder solution in a test tube, 4 volumes of reagent A [12 g $NaCO_3$ (≥ 99.5% purity), 6g NaKtartarate (≥ 99% purity), 8 g $NaHCO_3$ (≥ 99.7% purity), 72 g $NaSO_4$ (≥ 98% purity), and dH_2O 400 ml] and 1 volume of alkaline copper reagent B [2 g $CuSO_4$ (≥ 99% purity), 18 g $NaSO_4$ (≥ 98% purity) and dH_2O 100 ml] were added. Mixture was then boiled for 20mins and allowed to cool on ice. 1 ml of arseno-molybdate (≥ 97% purity) (see appendix for preparation) was added and mixed by gentle agitation. Seven millilitres (7 ml) of distilled water was added to each tube and mixed thoroughly. The absorbance of the solution mixture was measured at 540 nm using a spectrophotometer and subtracted from a blank reading. A standard curve of glucose was used to determine amount of reducing sugar in the samples. All reagents and salts used for analysis were purchased from Sigma-Aldrich, Germany.

2.3 Determination of Micronutrients

Bean samples (with exception of pigeon peas varieties) used in this study were analysed for nutrients including potassium, sodium, calcium, magnesium, iron, zinc, copper and cobalt by following the methods of the AOAC [36] method. Samples were first acid-digested and analysed using the atomic absorption spectrophotometer as described below. To 5.0 g of powdered samples was added 30ml of concentrated Nitric acid (HNO_3) ≥ 95.1% purity. The flask were placed in the dark overnight. Afterwards, 40ml of perchloric acid ($HClO_4$) ≥ 98% purity was added. The mixture was then initially heated on a hot plate at 50ºC for 15 min. Afterwards the temperature of heating was gradually increased to 200ºC. Heating was continued until the white dense fumes of perchloric acid disappeared. After digestion, the contents were cooled, filtered through Whatman filter paper (No. 2), filtrate transferred to a100 ml volumetric flask and then made up to the 100 ml mark with deionized water.

An Atomic Absorption Spectrophotometer (model 1233, Beckmann, US) equipped with a hollow cathode lamp and a fuel rich flame (air acetylene) with 20 Pa pressure and air 45 Pa pressure was used for the mineral content analyses. Parameters were adjusted according to manufacturer's instruction. The hollow cathode lamps for selected minerals with their wavelengths were used as a light source for each analyte. Lamp current was also set. The instrument was calibrated with standard solutions and the sample introduced to it by means of a capillary tube or aspiration. The mean signal responses were recorded at the elements respective wavelength (Potassium, 766 nm; Calcium, 422 nm; Magnesium, 205 nm; Sodium, 509 nm, Copper, 324 nm; Iron, 243 nm; Manganese, 279 nm; Zinc, 213 nm; Cobalt, 240 nm).

The concentration of each element was calculated as follows:

[Standard Concentration x Sample Absorbance x 100 x df] / [Standard Absorbance x Weight of sample]

Where df= dilution factor.

2.3.1 Determination of phosphorus

The A.O.A.C [36] method was used for the determination of phosphorus. Four millilitres of concentrated sulphuric acid (H_2SO_4) was added to 2 g of sample in a beaker. The mixture was left for 1 h to allow digestion of the sample. The sample digest was neutralized with 10 ml of 5 N NaOH solution, allowed to sit for a further 20 min

and thereafter filtered with Whatman filter paper (Whatman No. 1). The filtrate was then made up to 50 ml with distilled water in a standard volumetric flask. To 5.0 ml aliquot of the diluted filtrate was added 35 ml of distilled water and 2.5 ml of vanadomolybdate reagent (1.25 g common vanadate dissolved in 500 ml water with 10 ml HNO3, 0.5 ml of molybdate; 13 g of sodium molybdate in 200 ml of water) to produce a yellow-orange complex The solution was mixed and the absorbance measured at 520 nm wavelength with a spectrophotometer (PTUV/Vis spectrophotometer). The phosphorus content of the sample was calculated using absorbance of 1.0 mg potassium dihydrate phosphate (KH_2PO_4) as follows:

Phosphorus = [Sample Absorbance x Standard Concentration x100 x df(10)]/ [Standard Absorbance x Weight of sample]

2.4 Determination of Phaseolin Content

The solubility fractionation of seed protein was carried out according to the method of Gheyasuddin et al. [38]. The protein phaseolin was extracted by a sequence of successive extractions at room temperature for 1 h, at milled sample to solvent ratio of (1:10 w/v) on a magnetic stirrer. Firstly, distilled water was used to extract albumin at 1:10 w/v; then, 40 g/Liter Sodium Chloride solution was used to extract globulins at 1:10 w/v. Thereafter, 600 g/l Isopropanol was used to extract prolamine at 1:10 w/v. A 4 g/Liter sodium hydroxide solution was then added to extracted sample. After extraction, the slurry was centrifuged at 1500 x g for 10 min. The supernatant was collected and the extraction steps repeated three times successively on the residue. Supernatants from each extraction batch were pooled together. The nitrogen content in each extract (i.e. supernatant) was determined by a micro-kjeldahl method.

%Nitrogen = [0.014 x (sample titre - blank titre) x 0.1N x 100]/ [Sample weight]

2.5 Statistical Analysis

Data obtained were subjected to one-way analysis of variance (ANOVA) to determine significance ($P \leq .05$). Were significant, means were separated using Duncan multiple range test. Statistical package for the Social Sciences (SPSS) (IBM, Version21) was used for statistical analyses.

3. RESULTS AND DISCUSSION

Beans are important legumes in human nutrition [1]. They have been reported to contain essential nutrients and minerals required for human growth and body functioning [1,4,6]. In the present study, the chemical and nutritional composition of six “underutilized” beans were determined. The proximate composition of beans are shown in Table 1. Overall, there were significant differences ($P \leq .05$) between the proximate compositions of bean types. The proximate compositions are also comparable to values reported in literature elsewhere [32, 39-42]. Proteins which are essential building blocks of body tissues was highest (24.96%) in African yam bean (AYB) (*Sphenostylis stenocarpa*). Mung beans (*Vigna radiata*) had the second highest crude protein content of 22.07%. AYB and mung beans were the only bean types which had higher protein content than the “control’ (common beans, *Phaseolus vulgaris*, 21.07%). Soybean (*Glycine max*) had the lowest crude protein content of all bean types. The high crude protein content of these beans make them qualify as suitable alternatives and relatively affordable protein source [8,19] in comparison to the rather expensive price of animal-meat protein in developing countries. Some of these beans are currently being used as protein-rich condiments to enhance the flavour of traditional soups in Nigeria [8,19,20]. Ash content is an indication of the mineral nutrients present in a food material. The ash contents of the beans were significantly different ($P < .05$) among bean types. The highest mineral composition was in the African yam bean (4.3%), followed by Soybean (4.01%). The ash content of both pigeon pea varieties and black eyed peas did not differ significantly ($P > .05$). All the beans had higher ash content than common beans (3.26%). These observation shows that these underutilized beans are quite richer in minerals than common beans. Beans have been reported to be a good source of essential minerals and vitamins in human diet [1,6]. It is important to note that mineral content data does not translate to absolute bioavailability of these minerals when beans are consumed; the presence of anti-nutritional factors following bean processing are part of factors to be taken into consideration in determining absolute nutritive value of bean seeds [6,39,43].

The mean carbohydrate (CHO) content of black eyed pea and soybean were similar (62.81%) and were the highest values obtained. Mung beans had the lowest CHO content (47.47%).

Reducing sugars were highest in common beans and lowest in pigeon pea (grey variety). Crude fibre content was highest in Mung beans (15.24%) and lowest (excluding the control, common beans) in AYB (6.26%). Beans have been known to be a source of complex carbohydrates, including dietary fibre and resistant starch [1,44]. Fat content of the beans were generally low. Beans and legumes in general are low in fat and cholesterol [6]. Mung beans had the highest fat content (6.60%), while the lowest was pigeon pea (grey variety) (0.79%).

The macro and micro mineral elements of bean types/varieties are shown in Table 2. Nutrient composition of the bean was significantly different among bean sample types. All elements determined in this study was present in all bean types. These results reaffirm the mineral richness of beans as stated earlier in the introduction. Overall, calcium, magnesium, phosphorus and potassium (in mung beans) were present in high concentrations in the beans. While trace amounts of micro nutrients, coper and manganese were present. The total minerals composition was highest in mung beans and lowest in black eyed peas (Fig. 1). The importance of these mineral elements in humans have been published elsewhere [46-49]. Going by the assumption that ash content indicates the amount of mineral nutrients in beans, then it will be expected that the bean type with highest mineral elements in Fig. 1 ought to have the highest ash content in Table 1. However, the data obtained in this study does not seem to suggest that assumption. The discrepancy may be attributable to the presence of other minerals which were not assayed for in the present study (at least one of these minerals would be expected to be higher in African yam bean than the mung beans)

Presented in Table 3 are the phaseolin protein fractions of the beans. Phaseolin is a major storage protein in bean seeds [7]. The different fractions of phaseolin protein differed significantly (P<0.05) in between seeds. Overall, highest phaseolin fractions were present in African yam beans (AYB) and lowest in black eyed peas (BEP). The albumin fraction was highest in AYB and lowest in soybean, while the globulin fraction was highest in AYB and lowest in BEP. On the other hand, prolamine was highest (not taking into account common beans which served as control) in soybean and lowest in mung beans. Similar observation was reported on velvet beans. Among the seed proteins studied, globulins (9% to 62%) were higher, followed by albumin (4% to 21%), glutelin (1.3% to 2.9%), and prolamin (0.8% to 2%) [50-52]. Seeds of *Pachyrhizus erosus* (Yam beans) have been analyzed and reported glutelins constituting highest protein fraction followed by globulins. Compared to the globulins, albumins and protamins were low and the ratios were 28.8%, 16.3%, and 7%, respectively [53].

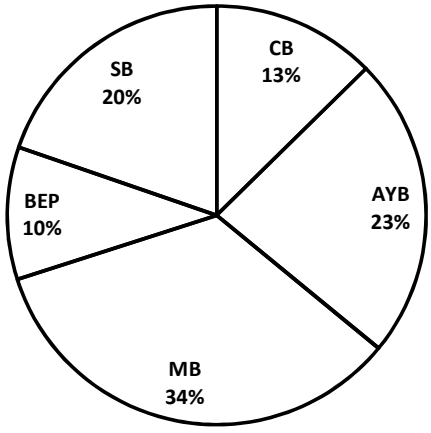

Fig. 1. Relative mineral composition of beans

CB, common beans; AYB, African yam beans; MB, Mung beans; BEP, Black eyed peas; SB, soybean

Table 1. Proximate composition of bean samples

Parameters (%)	*Common beans	African yam beans	Mung beans	Pigeon pea (red)	Pigeon pea (grey)	Black eyed pea	Soybean
Moisture	7.09±0.01^{b}	1.93±0.01^{f}	4.74±0.01^{d}	5.81±0.01^{c}	1.24±0.03^{g}	7.48±0.03^{a}	3.58±0.01^{e}
Ash	3.26±0.03^{e}	4.30±0.14^{a}	3.82±0.03^{c}	3.55±0.01^{d}	3.48±0.01^{d}	3.41±0.01^{d}	4.01±0.03^{b}
Fat	2.52±0.14^{d}	2.70±0.01^{c}	6.60±0.01^{a}	2.57±0.01^{d}	0.79±0.01^{f}	1.89±0.01^{e}	5.89±0.01^{b}
Crude protein	21.07±0.01^{c}	24.96±0.01^{a}	22.07±0.03^{b}	18.67±0.01^{d}	17.98±0.01^{e}	16.59±0.03^{f}	15.45±0.03^{g}
CHO	60.36±0.31^{c}	59.85±0.04^{d}	47.47±0.01^{e}	60.12±0.03^{cd}	61.52±0.01^{b}	62.81±0.03^{a}	62.81±0.01^{a}
Crude fibre	5.71±0.20^{g}	6.26±0.14^{f}	15.24±0.04^{a}	9.28±0.06^{c}	15.00±0.06^{b}	7.82±0.06^{e}	8.26±0.04^{d}
Reducing sugar	37.90±0.04^{b}	38.37±0.01^{a}	27.53±0.01^{d}	28.46±0.03^{c}	18.58±0.01^{g}	21.92±0.01^{f}	23.36±0.01^{e}
$^{+}$Calories (Kcal/100 g)	348.38±0.06^{c}	363.54±0.01^{b}	338.1±0.18^{d}	338.29±0.30^{d}	325.07±0.05^{f}	334.61±0.35^{e}	366.05±0.30^{a}

**Common beans (Phaseolus vulgaris) was included in the analysis for the purpose of comparison since it is more widely utilized in Nigeria. +Calculated by summing the product of the crude protein, fat and carbohydrate content with the respective Atwater's general energy conversion factors (4, 9, 4) [45]. Values are mean ± standard deviation (n=2). Values with different superscript on the same row are statistically different (P < .05)*

Table 2. Microelements of bean samples

Mineral nutrients (mg/100 g)	*Common beans	African yam bean	Mung beans	Black eyed pea	Soybeans
Potassium	5.91±0.01^{e}	11.35±0.1^{d}	154.7±0.42^{a}	13.77±0.03^{c}	15.06±0.01^{b}
Calcium	108.55±0.64^{d}	203.96±0.01^{a}	194.27±0.01^{b}	71.56±0.01^{e}	135.48±0.01^{c}
Magnesium	60.5±0.28^{e}	120.60±0.42^{b}	186.05±0.02^{a}	71.28±0.1^{d}	80.19±0.03^{c}
Sodium	12.33±0.18^{e}	30.04±0.01^{b}	18.44±0.01^{c}	40.20±0.14^{a}	14.99±0.01^{d}
Iron	0.91±0.00^{b}	19.91±0.01^{a}	11.01±0.01^{b}	5.86±0.03^{c}	2.54±0.1^{d}
Zinc	6.67±0.03^{c}	10.13±0.01^{b}	18.33±0.01^{a}	4.68±0.01 ^{d}	3.99±0.01^{d}
Phosphorus	151.38±0.03^{d}	244.63±0.01^{c}	357.52±0.03^{a}	121.91±0.03^{e}	286.61±0.01^{b}
Cobalt	0.135±0.00^{a}	0.06±0.04^{b}	0.13±0.01^{a}	0.11±0.00^{a}	0.02±0.00^{b}
Copper	0.90±0.00^{b}	2.31±0.00^{a}	0.33±0.00 ^{e}	0.64±0.03^{c}	0.46±0.00^{d}
Manganese	0.99±0.1^{e}	8.38±0.00^{a}	1.40±0.14^{d}	6.23±0.03^{b}	4.53±0.01^{c}

**Common beans (Phaseolus vulgaris) was included in the analysis for the purpose of comparison since it is more widely utilized in Nigeria. Values are means ± Standard error of mean (n=2). Values with the same superscript on the same row are statistically the same (P< .05)*

Table 3. Phaseolin protein fraction of beans

Protein (%/mg)	*Common beans	African yam beans	Mung beans	Black eyed pea	soybean
Albumin	34.38±0.53^{c}	41.89±1.57^{a}	37.19±0.69^{b}	31.69±0.72^{d}	29.96±1.21^{d}
Globulin	28.59±0.72^{c}	35.70±0.71 ^{a}	31.85±0.49^{b}	26.69±0.43^{d}	27.79±0.55^{cd}
Prolamine	16.10±0.42^{a}	11.45±0.14^{c}	9.13±0.74^{d}	8.92±0.40^{d}	12.90±0.42^{b}
Total protein	77.61±0.30^{b}	83.07±1.49^{a}	72.23±2.16^{c}	66.12±0.00^{d}	67.36±1.75^{d}

**Common beans (Phaseolus vulgaris) was included in the analysis for the purpose of comparison since it is more widely utilized in Nigeria. Values are means ± Standard error of mean (n=2). Values with the same superscript on the same row are statistically the same (P< .05)*

4. CONCLUSION

The underutilized bean seeds are rich in nutrients, especially protein and mineral elements. Their nutritional composition are comparable to widely adopted and consumed beans—common beans—in Nigeria. With the exception of anti-nutritional factors and other undesirable factors which were not the subject of the present study, these underutilized beans, especially African yam bean and mung beans, appear to be suitable alternatives to common beans for human consumption. Hence, the diversification of their uses is encouraged.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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APPENDIX

Preparation of Arsenomolybdate Reagent

100 g of ammonium molybdate was dissolved in 1800 ml (1.8 liters) of distilled water. To this, 84 ml of concentrated hydrogen sulphate was added to the solution. 12 g of bisodium hydrogen arsenate (orthoarsenate, $Na_2H_2O_4.7H_2O$) was dissolved in 10ml of distilled water. This solution was mixed with ammonium molybdate, placed in the incubator between 24-84 h at 37ºC and stored in amber bottle.

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