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Effects of processing methods on nutritive values of *Ekuru* from two cultivars of beans (*Vigna unguiculata* and *Vigna angustifoliata*)

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Beans contain substantial amount of protein, dietary fibre, B-vitamins, minerals, and anti-nutrients which limit their utilisation. Processing reduce the level of antinutrients in plant products but little information exist on effects of processing methods on nutrient and antinutrient composition of bean products. This study was undertaken to determine the effects of processing methods on nutrient and antinutrient composition, and micronutrient potential of bean paste (Ekuru) from two cultivars of beans, white (Vigna unguiculata) and red (Vigna angustifoliata) species of beans which were processed into white and brown Ekuru using standardised traditional methods. 100 g of raw V. unguiculata and V. angustifoliata contained 22.83 and 23.94 g protein, 1.94 and 2.11 g crude lipid, 30 and 29 mg sodium, 1.110 and 1.124 mg potassium, 390 and 130 mg phosphorus, 10.65 and 10.95 mg iron and 5.87 and 5.95 mg zinc, respectively. Ekuru products contained between 18.76 and 21.90 g protein, 0.81 and 1.38 g crude lipid, 20.00 and 90.00 mg sodium, 870 and 1124 mg potassium, 8.16 and 8.36 mg iron and 4.87 and 5.21 mg zinc, respectively. Processing decreased the nutrient content of the products compared with raw samples, the red bean product was higher in crude protein, crude lipid and ash; while the white bean product was higher in crude fibre and moisture content. The bean products were very low in antinutrients, with no trypsin inhibitors. Ekuru samples can be good sources of plant protein, nonhaeme iron and zinc, and their consumption should be encouraged.

Key words: Processing methods, nutritive value, antinutrients, Ekuru, Vigna unguiculata, Vigna angustifoliata.

INTRODUCTION

Malnutrition is a public health challenge especially in the developing countries of the world, and micronutrient malnutrition affect majority of people, with its effect being more pronounced among the rural poor communities. Good nutrition involves consuming foods and supplements rich in essential nutrients that provide

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Author(s) agree that this article remain permanently open access under the terms of the <u>Creative Commons Attribution License 4.0</u> International License adequate energy and support growth and body maintenance (Shills et al., 2006). Plant protein plays significant role in human nutrition, and legumes provide a range of essential nutrients including protein, low glycemic carbohydrates, dietary fibre, minerals and vitamins (Munro, 2007). Worldwide, most grown legumes are soybean, peanut, cowpea (beans) and chickpeas (Shutler et al., 1989). Beans have significant amount of both soluble and insoluble fibre (www.ext.colostate.com); its soluble fibre helps in lowering blood cholesterol by insulin creating more receptor sites (www.recipetips.com). The use of locally available foods and feedstuffs are being encouraged as a means of dietary diversification in combating the menace of micronutrient deficiencies. Beans have significant antioxidant properties which make it a terrific anti-aging food (FAO, 2009).

However, dry legumes contain several anti-nutritional factors such as glucosides, trypsin and chymotrypsin inhibitors, phytates and lectins (Vidal-Valverde et al., 1994), but some simple and inexpensive processing techniques such as soaking and cooking are highly efficient for the reduction of these anti-nutritional factors (Frias et al., 2000). Much information is available in the literature on nutrient content and potential contribution of different varieties of raw beans, but information on their prepared products is scanty. Beans can be prepared to local paste (Ekuru) by boiling and consumed with sauce. This study was undertaken to determine the effects of processing methods on nutrient and antinutrient composition and micronutrient retention of 'Ekuru' from two cultivars of beans, Vigna unguiculata and Vigna angustifoliata.

MATERIALS AND METHODS

Sample collection and preparation

Samples of white (V. unguiculata) and red (V. angustifoliata) varieties of beans were purchased from Bodija market in Ibadan, Oyo State, Nigeria. Each of the bean sample was divided into three portions - one portion each was used for raw sample analysis (Samples 1 and 2), second portion was dehulled (Samples 3 and 4), and the third portion used for Ekuru preparation. Each of the portions for Ekuru preparation was soaked in distilled water until soft and then dehulled. To 200 g of dehulled beans was added 50 g of onion, ground with a warring blender and stirred into a fluffy paste. Each of the paste was divided into two portions, giving a total of four Ekuru samples. One portion each from the two varieties of beans was prepared to white Ekuru (Samples 5 and 6), while 1 g of ground potash was added to each of the second portions to form brown Ekuru (Samples 7 and 8). The samples were wrapped with leaves and cooked for 40 min. After cooking, the moisture content of the four Ekuru samples was determined and the rest samples were dried to constant weight at 60°C, wrapped in cellophane nylon and refrigerated till when needed.

Chemical analyses

The eight samples were analyzed in triplicate for moisture, crude

protein, crude lipid, crude fibre and ash using standard methods of Association of Official Analytical Chemists (AOAC, 2005). The carbohydrate content was obtained by difference.

Proximate composition

The moisture content of the samples was determined by air oven method (Gallenkamp, Model OV - 440, England) at 105°C. The crude protein was determined by micro-Kjeldahl method through digestion of 5 g of sample with conc. H₂SO₄ and Kjeldahl catalyst in Kjeldahl flask for 3 h. The digest was made up to 100 and 5 ml portion was pipetted to Kjeldahl apparatus and 5 ml of 40% (w/v) NaOH solution was added. The mixture was steam distilled, and the liberated ammonia was collected in 10 ml of 2% boric acid, and titrated against 0.01 M HCl solution. The amount of crude protein was then calculated by multiplying percentage nitrogen in the digest by 6.25. Crude lipid was determined by weighing 5 g of dried sample into fat free extraction thimble and plugging lightly with cotton wool. The thimble was placed in the Soxhlet extractor fitted up with reflux condenser. The dried sample (at 60°C) was then extracted with petroleum ether and the crude lipid estimated as g/100 g dry weight of sample, and then converted to g/100 g fresh sample weight. The ash content was determined by weighing 5 g of sample in triplicate and heated in a muffle furnace at 550°C for 4 h, cooled to about 100°C in the furnace and then transferred into a dessicator to cool to room temperature, weighed, and ash was calculated as g/100 g original fresh sample. Crude fibre was determined using the method of Saura-Calixto et al. (1983). The carbohydrate content was obtained by difference. Gross energy of the samples was determined using ballistic bomb calorimeter (Manufacturer: Cal 2k – Eco, TUV Rheinland Quality Services (Pty) Limited, South Africa).

Mineral analysis

Potassium and sodium were determined by digesting the ash of the samples with perchloric acid and nitric acid, and then taking the readings on Jenway digital flame photometer/spectronic 20 (Bonire et al., 1990). Phosphorus was determined by vanado-molybdate colorimetric method (Ologhobo and Fetuga, 1983). Calcium, magnesium, iron, zinc, manganese, copper and selenium were determined spectrophotometrically by using Buck 200 atomic absorption spectrophotometer (Buck Scientific, Norwalk) (Essien et al., 1992) and their absorption compared with absorption of standards of these minerals.

Antinutrient analysis

Oxalate was determined by extraction with water for about 3 h and standard solutions of oxalic acid prepared and read on spectrophotometer (Spectronic 20) at 420 nm. The absorbance of the sample extracts were also read and amount of oxalate was estimated. Phytate was determined by titration with ferric chloride solution (Sudarmadji and Markakis, 1977); while trypsin inhibitory activity was determined on casein and comparing the absorbance with that of trypsin standard solutions read at 280 nm (Makkar and Becker, 1996). The tannin content was determined by extracting the samples with a mixture of acetone and acetic acid for 5 h, measured their absorbance and compared the absorbance of the extracts with the absorbance of standard solutions of tannic acid at 500 nm on spectronic 20 (Griffiths and Jones, 1977). Saponin was also determined by comparing the absorbance of the extracts with the standard at 380 nm (Makkar and Becker, 1996). Chi square test was performed on the results obtained and level of significance set at p<0.05.

Sample	Moisture	Protein	Fat	Fibre	Ash	Carbohydrate
Sample 1	13.7±0.03	22.8±0.32	1.9±0.03	4.5±0.03	3.7±0.02	53.4±0.03
Sample 2	12.2±0.20	23.9±0.80	2.1±0.02	4.3±0.03	3.7±0.03	53.7±0.03
Sample 3	16.4±0.03	20.8±0.14	1.6±0.03	1.4±0.02	2.0±0.02	57.8±0.10
Sample 4	15.3±0.03	21.8±0.01	1.6±0.03	1.4±0.02	2.0±0.02	57.9±0.07
Sample 5	69.7±0.04	21.2±0.40	0.8±0.03	0.8±0.03	1.0±0.01	7.5±0.20
Sample 6	67.9±0.03	21.9±0.06	1.0±0.02	0.7±0.02	1.1±0.02	7.4±0.08
Sample 7	65.2±0.08	18.8±0.12	1.3±0.03	0.7±0.03	1.2±0.02	12.9±0.20
Sample 8	64.9±0.08	19.8±0.10	1.4±0.03	0.6±0.03	1.3±0.02	12.0±0.06

Table 1. Proximate composition of whole, dehulled and processed bean varieties (g/100 g)*.

Values are means±SD of triplicate determinations. Sample 1 = raw white bean; Sample 2 = raw red bean; Sample 3 = dehulled white bean; Sample 4 = dehulled red bean; Sample 5 = processed white bean *Ekuru*; Sample 6 = processed red bean *Ekuru*; Sample 7 = processed white bean *'coloured Ekuru*; Sample 8 = processed red bean *'coloured Ekuru*'.

Table 2a. Mineral composition of raw and dehulled white and red beans (mg/100 g)*.

Deremeter	Sample					
Parameter	1	2	3	4		
Sodium	30.00±8.00	29.00±2.00	20.00±0.00	30.00±1.00		
Potassium	1110.00±60.00	1124.00±20.00	1110.00±10.00	1120.00±10.00		
Calcium	60.00±10.00	80.00±4.00	60.00±0.00	70.00±5.00		
Magnesium	240.00±2.00	270.00±20.00	150.00±3.00	210.00±30.00		
Phosphorus	390.00±2.00	130.00±20.00	300.00±3.00	100.00±20.00		
Iron	10.65±0.03	10.95±0.07	9.28±0.03	9.70±0.02		
Zinc	5.87±0.02	5.95±0.02	5.45±0.03	5.70±0.03		
Manganese	1.84±0.03	2.00±0.03	1.59±0.03	1.74±0.03		
Copper	0.65±0.03	0.70±0.03	0.52±0.03	0.60±0.02		

Values are means \pm SD of triplicate determinations. Sample 1 = raw white bean; Sample 2 = raw red bean; Sample 3 = dehulled white bean; Sample 4 = dehulled red bean; Sample 5 = processed white bean *Ekuru*; Sample 6 = processed red bean *Ekuru*; Sample 7 = processed white bean *'coloured Ekuru*'; Sample 8 = processed red bean *'coloured Ekuru*'.

RESULTS

The results of proximate nutrient composition of the raw, dehulled and processed V. unquiculata and V. angustifoliata are shown in Table 1. The two varieties of beans were high in protein and carbohydrates, moderate in ash and crude fibre but low in moisture and fat content. V. unguiculata [Sample 1 (white beans)] had higher value of moisture and crude fibre, while V. angustifoliata (Sample 2 (red beans)) was higher in crude protein, crude fat and carbohydrate content with no significant difference in all the nutrient values (p>0.05). The two varieties of beans had the same value of ash. Soaking and dehulling led to slight but significant increase in the moisture and carbohydrate content of the samples (Samples 3 and 4), with significant reduction in their crude protein, fat, fibre and ash content. Processing the beans to Ekuru led to significant increase in moisture content (p<0.05) and insignificant increase in protein, compared with the dehulled samples (Samples 5 and 6). However, processing significantly reduced the carbohydrate, crude fat, fibre and ash content of the products compared with the raw and dehulled samples (p<0.05). Brown *Ekuru* samples (Samples 7 and 8) had significantly lower value of crude protein and fibre compared with white *Ekuru*, but significantly higher value of fat, ash and carbohydrate content (p<0.05).

The mineral composition of the two varieties of beans and their *Ekuru* products are as shown in Tables 2a and 2b. The two bean varieties were very high in potassium and phosphorus, high in magnesium and iron but low in copper and very low in sodium and calcium. Dehulling resulted in significant reduction in sodium, magnesium, phosphorus, iron, copper (p<0.05), with slight insignificant reduction in potassium and zinc content of the beans. Processing the beans to *Ekuru* led to significant increase in sodium, calcium, and phosphorus content of the products, with significant reduction in potassium, iron and zinc content compared with the raw and dehulled samples. Addition of potash significantly increased the mineral content of red coloured *Ekuru* (Samples 7 and 8) of the two bean varieties compared with the white

Devenueter	Sample					
Parameter	5	6	7	8		
Sodium	30.00±0.00	20.00±1.00	90.00±3.00	20.00±1.00		
Potassium	900.00±20.00	870.00±10.00	970.00±10.00	920.00±10.00		
Calcium	700.00±20.00	670.00±9.00	800.00±30.00	750.00±10.00		
Magnesium	260.00±2.00	200.00±20.00	300.00±1.00	260.00±30.00		
Phosphorus	380.00±6.00	340.00±10.00	450.00±2.00	420.00±30.00		
Iron	8.21±0.02	8.16±0.02	8.36±0.02	8.30±0.03		
Zinc	4.94±0.02	4.87±0.04	5.21±0.03	5.21±0.02		
Manganese	1.32±0.01	1.27±0.02	1.42±0.03	1.40±0.03		
Copper	0.66±0.03	0.56±0.03	0.75±0.03	0.71±0.04		

Table 2b. Mineral composition of white and red bean products (mg/100 g)*.

Values are means±SD of triplicate determinations. Sample 1 = Raw white bean; Sample 2 = Raw red bean; Sample 3 = Dehulled white bean; Sample 4 = dehulled red bean; Sample 5 = Processed white bean *Ekuru*; Sample 6 = Processed red bean *Ekuru*; Sample 7 = Processed white bean *'coloured Ekuru*'; Sample 8 = Processed red bean *'coloured Ekuru*'.

Table 3. Antinutrient composition of whole, dehulled and white and red bean products (mg/100 g)*.

Sample	Phytate	Oxalate	Tannin	Trypsin inhibitors (TIU/mg)
Sample 1	0.32±0.01	0.07±0.00	0.43±0.02	20.60±0.02
Sample 2	0.26±0.03	0.07±0.00	0.46±0.02	21.40±0.02
Sample 3	0.28±0.00	0.06±0.00	0.33±0.02	11.30±0.20
Sample 4	0.21±0.02	0.06±0.00	0.38±0.02	12.70±0.12
Sample 5	0.03±0.00	0.02±0.00	0.01±0.00	0.00±0.00
Sample 6	0.02±0.01	0.02±0.00	0.01±0.00	0.00±0.00
Sample 7	0.07±0.02	0.02±0.00	0.01±0.07	0.00±0.00
Sample 8	0.02±0.00	0.01±0.00	0.01±0.00	0.00±0.00

Values are means±SD of triplicate determinations. Sample 1 = raw white bean; Sample 2 = raw red bean; Sample 3 = dehulled white bean; Sample 4 = dehulled red bean; Sample 5 = processed white bean *Ekuru*; Sample 6 = processed red bean *Ekuru*; Sample 7 = processed white bean *'coloured Ekuru*'; Sample 8 = processed red bean *'coloured Ekuru*'.

coloured ones (Samples 5 and 6), (p<0.05). The level of antinutrients in the raw and processed bean samples are shown in Table 3. The raw white and red beans were low in antinutrients, except for the trypsin inhibitors whose values were on the high side. The two varieties had the same level of oxalates, white bean had higher phytate value, while the red bean was higher in tannins and trypsin inhibitors values. Dehulling and boiling led to significant reduction in the levels of all the antinutrients (p<0.05), while boiling completely eliminated the trypsin inhibitors in all the *Ekuru* products.

DISCUSSION

The result of proximate composition of two raw cowpea varieties (Samples 1 and 2) are as shown in Table 1. The values obtained for the raw samples were in agreement with the values reported for some varieties of cowpea in the literature (Chel-Guerrero et al., 2002; Adeparusi and Olute, 2010; Ajayi et al., 2010). The moisture content of

the raw beans was low, and this underscores its long shelf life and less susceptibility to microbial attack. The two varieties of beans were high in protein content and can be good sources of plant protein. This justifies their use as source of protein in Nigeria, especially among the rural populace living the traditional lifestyle with low consumption of animal products due to poverty (Adewale, 2005). The raw beans were low in fat but high in crude fibre and ash. This finding is in agreement with that of Oshodi and Aletor (1993) (4.3% fibre), and Adeparusi and Olute (2010) (4.11 g fibre and 4.27 g ash /100 g) on Lima bean. Legumes generally contain low fat content in the range 1.0 to 2.0 g/100 g sample, except for chickpea (6.7 g/100 g, Costa et al., 2006); soybean and peanut (21 g and 49 g/100 g sample, respectively, Augustin and Klein, 1989). Dehulling and processing by boiling of the bean varieties resulted in significant reduction in all the nutrient values, the cooked samples (Samples 5 to 8) recording higher reduction in values of fat, crude fibre and ash. The observed reduction in nutrient values is believed to be due to increase in moisture content of the

cooked products. Raw and processed samples of V. angustifoliata (Samples 2, 6 and 8) were insignificantly higher in crude protein, fat and ash content than V. unguiculata samples (Samples 1, 5, and 7), while the latter was higher in crude fibre and carbohydrate content (p>0.05). Traditional treatment methods of cowpeas such as dehulling, soaking, cooking, fermentation and germination have been used to improve the nutritional quality of food legumes to various extents (Chi-Fai et al., 1997; Mubarak, 2005). Dehulled beans, white and coloured Ekuru samples (Samples 3 to 8) contained lower fibre content than raw beans, implying that the seed coat is comprised mainly of fibre, while the protein and fat are concentrated in the cotyledon (Mungudi et al., 2010). Addition of potash to Ekuru samples (Samples 7 and 8) led to significant reduction in crude protein and fibre content of the products compared with Ekuru white (Samples 5 and 6). This observation confirms the report that potash has an adverse effect on protein content of foods (Isikwenu et al., 2013). However, addition of potash slightly increased the crude fat, ash and carbohydrate content of the products.

The results obtained for the two varieties of beans in Table 2a are in agreement with report of the literature (Adegunwa et al., 2012). The two varieties of beans (Samples 1 and 2) are good sources of potassium, magnesium, phosphorus, iron and zinc (Table 2a). Raw white beans was significantly higher in phosphorus content, while red beans was significantly higher in potassium, calcium, magnesium, iron and zinc content. The very low sodium and very high potassium content of the raw bean varieties (Table 2a, Samples 1 and 2) is advantageous and make them suitable for consumption by the hypertensives that require low sodium and high potassium for intra and extracellular electrolyte balance. Dehulling led to significant reduction in the mineral content of dehulled samples (Samples 3 and 4). This was an indication that removal of the seed coat exposed the minerals to leaching into the soaking water. Dehulling of seeds has been reported to cause loss of minerals through leaching during soaking and cooking of legumes (Augustine and Klein, 1989; Mungudi et al., 2010). Processing the two varieties of beans to Ekuru (Samples 5 and 6, Table 2b) led to significant reduction in potassium, zinc, manganese and copper content, while it significantly increased the calcium, magnesium and phosphorus content of the products (p<0.05). Addition of potash to red Ekuru (Samples 7 and 8, Table 2b) significantly increased mineral content of the samples in comparison with white Ekuru (Samples 5 and 6).

The phytate, oxalate and tannin content of the two varieties of beans (Samples 1 and 2) were very low and cannot constitute any significant hindrance to nutrient digestibility or absorption. However, the trypsin inhibitors level was high in the two samples. High level of trypsin inhibitors in legumes has always been the limiting factor for their effective and efficient utilization (Leiner, 1996). Dehulling led to significant reduction in the levels of all the antinutrients studied (p<0.05), and the reduction was much pronounced in the trypsin inhibitor levels. The reduction in the antinutrient levels was attributable to the removal of the seed coat, indicating that the coat contained significant amount of these antinutritents, especially trypsin inhibitors. The trypsin inhibitor values of dehulled samples were lower than what was reported for pigeon pea (Oloyo, 2004). Processing the beans to Ekuru led to significant reduction in phytates, oxalates and tannin content of the products compared with the raw and dehulled samples (p<0.05), while it completely destroyed the trypsin inhibitors in all the Ekuru samples (Samples 5 to 8). Addition of potash to the two varieties of beans did not increase the level of antinutrients significantly (p>0.05).

Conclusion

Generally, proximate composition of *V. unguiculata* and *V. angustifoliata* compared well with other types of legumes. However, different processing methods and the length of cooking time significantly decrease crude protein, fibre, ash and anti-nutritional contents. The decrease in the amounts of anti-nutrients is a major beneficial effect of cooking beans because the nutrients will be more digestible and bioavailable. Processing beans to *Ekuru* improves its nutrient content, and consumption of *Ekuru* should be popularized and promoted among all and sundry, especially among the elites as a means to combat macro and micronutrient malnutrition.

Conflict of interests

The authors have no conflict of interests, as the work was done solely through self effort.

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