Effect of Combined Processing Methods on the Proximate and Mineral Composition of Pigeon Pea (Cajanus Cajan) Flour

Nwanekezi Emmanuel C.1, Ubbaonu Collins N.2, Arukwe Dorothy C.1,*

1Department of Food Science and Technology, Imo State University, Owerri, Nigeria
2Department of Food Science and Technology, Federal University of Technology, Owerri, Nigeria

Email address: dorarukwe@gmail.com (Arukwe D. C.)
*Corresponding author

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Abstract: The combined effects of different processing methods on the proximate and mineral composition of pigeon pea flour samples were investigated. The pigeon pea seeds were soaked (control), soaked and boiled, soaked and fermented, soaked, boiled and fermented, soaked and sprouted, soaked sprouted and boiled, soaked, sprouted and fermented and soaked, sprouted, boiled and fermented. The proximate result (dry weight basis) showed increased protein. The highest protein content (33.21%) was observed in the soaked, sprouted and fermented pigeon pea flour (SoSpFFPFPF) and this was significantly higher (p<0.05) than that (32.20%) of the soaked, sprouted, boiled and fermented pigeon pea sample (SoSpBFPPPF) and other flours, ash, fibre but decrease in fat and carbohydrate contents of sprouted, fermented, and combined sprouted/fermented processes while the boiled processes had decreased protein, ash, fibre, fat and increased carbohydrate contents. The results equally showed that sprouting and fermentation significantly (p<0.05) increased the minerals analyzed and combined sprouting/fermentation tremendously increased them (calcium 141 – 176mg/100g, magnesium 115.5 – 166.28 mg/100g, potassium 1205 – 1577mg/100g). The boiled processes recorded decreased mineral contents. This study gives an insight on the combined effects of sprouting, boiling and fermentation on the nutrients composition of pigeon pea. The results of this study leads to a better understanding of this legume (pigeon pea) thereby leading to their increased utilization at domestic and industrial levels.

Keywords: Combined Effects, Different Processing Methods, Proximate, Mineral, Fermentation

1. Introduction

Legumes are known to be the third largest family among flowering plants consisting of approximately 650 genera and 20,000 species [8]. The legumes used by humans are commonly called food legumes or grain legumes. The food legumes are divided into two groups: pulses and oil seeds. Pulses are edible seeds of cultivated legumes [14]. Oil seeds are those legumes used primarily for their oil content. Of the thousand known legume species, less than twenty are used extensively today as food, the major ones being soybean, chickpea, mung bean, groundnut, black-eye pea or cowpea, winged bean and real-pea. Some minor species are underutilized though they are often cultivated by traditional farmers and used to alleviate hunger and overcome malnutrition. They include African yam bean, lima bean, pigeon pea and bambara groundnut.

Pigeon pea is grown for food and is produced in Nigeria at subsistence level [1]. It is commonly called “fio fio” in south eastern Nigeria. Pigeon pea has fairly high protein content and a good source of dietary minerals such as calcium, phosphorus, magnesium, iron, copper, sulphur and potassium [29]. Minerals are constituents of skeletal tissues, co-factors to enzymes, carrier protein/protein hormones and electrolytes in body fluids and cells [22]. Minerals unlike protein cannot be synthesized in the body and can be obtained through dietary means.
The most serious malnutrition problem in Nigeria and extensively Africa is the protein-calorie malnutrition and mineral deficiency (hidden hunger) especially among children and pregnant/nursing mothers. The United Nations System Standing Committee on Nutrition (SCN) reported that malnutrition is directly or indirectly related to more than 50% of all children mortality and is a contributing factor to diseases in developing countries [27]. The commonly relied source of protein from animals has become very expensive and out of reach of the common man. Therefore harnessing plant protein and minerals from pigeon pea to meet the needs of the populace has become necessary. But pigeon pea contains some toxicants and anti-nutrients which make them unacceptable by consumers. It has long become necessary to develop improved methods of detoxifying pigeon pea so that their potential for use in improving protein and mineral malnutrition can be achieved. The aim of this study was to evaluate the combined effects of sprouting, boiling and fermentation on the proximate and mineral composition of pigeon pea.

2. Materials and Methods

2.1. Materials Collection

The pigeon pea (Cajanus cajan) seeds for this research were purchased from a market (Ahia ohuu) in Aba, Abia State. The chemicals used were obtained from Food Science and Technology laboratory, Imo State University, Owerri and National Root Crops Research Institute, Umudike, Umuahia.

2.2. Sample Preparation

Sixteen kilograms of pigeon pea seeds were sorted to remove dirt and other foreign particles after which they were washed. The grains were then soaked in water (1.3v/v) for 3 hours using a large container and the water drained.

2.2.1. Production of Soaked Pigeon Pea Flour (Control)

Two (2) kilograms of the soaked grains were dehulled and dried in an oven at 60°C for 7 hours. The dried seeds were milled into flour using disc attrition mill (Asiko All, Addis Nigeria). The flour was then sieved with standard sieve (1.0mm mesh) and packaged in polyethylene bag for further studies.

2.2.2. Production of Soaked and Boiled Pigeon Pea Flour

Two (2) kilograms of the soaked grains were dehulled and boiled in water for 1 hour at 100°C. The water was drained and the seeds dried in an oven at 60°C for 7 hours. The dried seeds were milled into flour using disc attrition mill (Asiko All, Addis Nigeria). The flour was then sieved with standard sieve (1.0mm mesh) and packaged in polyethylene bag for further studies.

2.2.3. Production of Soaked and Fermented Pigeon Pea Flour

Two kilograms (2kg) of the soaked grains were dehulled, crushed, wrapped in plantain leaves and allowed to ferment for 4 days. After fermentation, the grains were dried in an oven at 60°C for 7 hours. The fermented dried cotyledons were milled into flour with disc attrition mill (Asiko All, Addis Nigeria) and standard (1.0 mm) mesh sieved before packaging in polyethylene bag for further studies.

2.2.4. Production of Soaked, Boiled and Fermented Pigeon Pea Flour

Two kilograms (2kg) of the soaked grains were boiled with water for 1 hour and the water drained. The grains were wrapped in plantain leaves and allowed to ferment for 4 days as described by [16]. After fermentation, the seeds were dehulled and dried in an oven at 60°C for 7 hours. The dried seeds were milled into flour using disc attrition mill (Asiko All, Addis Nigeria). The legume flour were sieved with standard sieve (1.0 mm mesh) and packaged in polyethylene bag for further studies.

2.2.5. Production of Soaked and Sprouted Pigeon Pea Flour

Sprouting was carried out according to the method described by [5]. Eight (8) kilograms of the soaked grains were spread in a single layer on a moistened jute bag and allowed to germinate (sprout) at room temperature for 3 days. During this time, the grains were sprayed with water at intervals of 12 hours until the last day of sprouting. After sprouting, the seeds were dehulled and rootlets removed. Then the cotyledons were divided into four portions of 2kg each. Then the portion (2kg) for the production of soaked and sprouted pigeon pea flour were dried in an oven at 60°C for 7 hours and milled into flour using a disc attrition mill (Asiko All, Addis Nigeria) and 1.0 mm mesh sieved before packaging into polyethylene bag for further studies.

2.2.6. Production of Soaked, Sprouted and Boiled Pigeon Pea Flour

Two (2) kilograms of the sprouted pigeon pea seeds were boiled for 1 hour with water at 100°C, drained and dried in an oven at 60°C for 7 hours. The dried sprouted boiled cotyledons were milled with disc attrition mill (Asiko All, Addis Nigeria), sieved with standard sieve (1.0mm mesh) and packaged in polyethylene bag for further studies.

2.2.7. Production of Soaked, Sprouted and Fermented Pigeon Pea Flour

Two (2) kilograms of the sprouted dehulled grains were wrapped in plantain leaves and allowed to ferment for 4 days as described by [16]. After fermentation, the cotyledons were dried in an oven at 60°C for 7 hours and milled into flour with disc attrition mill (Asiko All, Addis Nigeria) and standard 1.0mm mesh sieved before packaging in polyethylene bag for further studies.

2.2.8. Production of Soaked, Sprouted, Boiled and Fermented Pigeon Pea Flour

Two (2) kilograms of the sprouted dehulled grains were boiled in water for 1 hour at 100°C and the water drained. The sprouted, dehulled and boiled cotyledons were wrapped in plantain leaves and allowed to ferment for 4 days as...
described by [16]. After fermentation, the cotyledons were dried in an oven at 60°C for 7 hours. The dried sprouted-boiled-fermented grains were milled into flour with disc attrition mill (Asiko All, Addis Nigeria) and standard 1.0 mm mesh sieved before packaging in polyethylene bag for further studies.

3. Proximate Analysis

The proximate (Moisture, fat, Protein, ash, crude fibre, and carbohydrate) analysis of all the samples was carried out according to the method of [4]. Two (2) grams each of the samples were

4. Mineral Analysis

The sample for the analysis was prepared by ashing about 0.5g of the sample in a muffle furnace (Carbolite bambord, Sheffield, England) for 6hrs at 550 to 600°C and allowed to cool in a desiccator. Five millilitres (5ml) of 1M HNO₃ was then added and the solution evaporated to dryness using a hot plate. The crucible with its content were returned to the furnace and incinerated for 10-15mins. On cooling, 10ml of 1M HCl solution was added and filtered into 50ml volumetric flask with Whatman No. 1 filter paper and funnel. The residue on the filter paper was rinsed with 0.1 M HCl solution into 100ml volumetric flask and made up to mark with 0.1M HCL.

4.1. Determination of Potassium and Sodium by Flame Photometry

The sample for the analysis was prepared by ashing about 0.5g of the sample in a muffle furnace (Carbolite bambord, Sheffield, England) for 6hrs at 550 to 600°C and allowed to cool in a desiccator. Five millilitres (5ml) of 1M HNO₃ was then added and the solution evaporated to dryness using a hot plate. The crucible with its content were returned to the furnace and incinerated for 10-15mins. On cooling, 10ml of 1M HCl solution was added and filtered into 50ml volumetric flask with Whatman No. 1 filter paper and funnel. The residue on the filter paper was rinsed with 0.1 M HCl solution into 100ml volumetric flask and made up to mark with 0.1M HCL.

4.2. Determination of Calcium and Magnesium by Compleximetric Method Using EDTA

The method described by [26] was employed and the experiment was carried out in triplicate. In this method, calcium was determined by Versenate method using calcium indicator. Calcium and magnesium were determined together and then magnesium was obtained by difference between calcium and magnesium values.

A quantity of EDTA (0.931g) was dissolved in 1 litre of distilled indicator. Broichrome Black 1 (0.25g) was dissolved in 50ml methylated spirit. Buffer solution was prepared by dissolving 65.70g of NH₄Cl(s) in water, 570ml of 0.88N NH₄OH solution added before diluting to 1 litre. Five millilitres (5ml) of the sample solution was pipetted into a conical flask and diluted to 100ml with water. Then, 15ml of buffer solution, 10 drops of indicator and 2ml of triethanolamine were then added. The solution was titrated until the colour of the solution changed from red to a clean blue colour. A blank titration was carried out and subtracted from the sample reading.

Calculation was done as follows:

$$\text{(Ca + Mg) g/kg} = \frac{x ml \times \text{volume of solution}}{10 \times \text{aliquot x sample mass (g)}}$$

Where X = volume of EDTA solution used in titration.

5. Results and Discussion

5.1. Proximate Composition of Pigeon Pea Flour Samples Processed By Different Methods

The mean values of proximate composition of soaked (S,PPF), soaked and boiled (S,BPPF), soaked and fermented (S,FPPF), soaked, boiled and fermented (S,BFPF), soaked, sprouted (S,S,PPF), soaked, sprouted and fermented (S,S,FPPF) and soaked, sprouted, boiled and fermented (S,S,BFPF) pigeon pea flour samples are presented in Table 1. There were significant differences (p<0.05) in the protein contents of these pigeon pea flour samples. The control sample (soaked pigeon pea flour) had a protein value of 24.15% which was significantly (p<0.05) higher than that of the soaked and boiled pigeon pea flour which had a protein content of 22.78%. The highest protein content (33.21%) was observed in the soaked, sprouted and fermented pigeon pea flour (S,S,FPPF) and this was significantly higher (p<0.05) than that (32.20%) of the soaked, sprouted, boiled and fermented pigeon pea sample (S,S,BFPF) and other flours. The increase in protein by fermentation treatment might be due to synthesis of proteins by fermenting microorganisms while that due to sprouting could be attributed to the degradation of carbohydrate and fat by sprouting enzymes for energy leading to increase in the relative proportion of protein. This result was in line with the work of [6] who observed an increase in protein during germination of bean, chick pea and pea seeds. Also, [25] who reported increase in protein content of beniseed flour which was produced from fermented beniseed.

Table 1. Proximate Composition of Pigeon Pea Flour Processed By Different Methods.

<table>
<thead>
<tr>
<th>Flour sample</th>
<th>Protein (%)</th>
<th>Fat (%)</th>
<th>Ash (%)</th>
<th>Crude Fibre (%)</th>
<th>Carbohydrate (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>S,PPF</td>
<td>24.15±0.01a</td>
<td>2.39±0.01a</td>
<td>4.38±0.01a</td>
<td>6.09±0.02a</td>
<td>62.99±0.06a</td>
</tr>
<tr>
<td>S,BPPF</td>
<td>22.78±0.01a</td>
<td>2.19±0.02a</td>
<td>3.27±0.02a</td>
<td>5.68±0.01a</td>
<td>66.08±0.03a</td>
</tr>
<tr>
<td>S,FPPF</td>
<td>28.98±0.01a</td>
<td>2.22±0.02a</td>
<td>4.85±0.01b</td>
<td>7.36±0.02a</td>
<td>56.86±0.07a</td>
</tr>
<tr>
<td>S,BFPF</td>
<td>26.03±0.02a</td>
<td>2.06±0.0a</td>
<td>4.26±0.03a</td>
<td>6.28±0.02a</td>
<td>61.37±0.03a</td>
</tr>
<tr>
<td>S,S,PPF</td>
<td>27.54±0.02a</td>
<td>1.70±0.01a</td>
<td>4.59±0.02a</td>
<td>8.30±0.01a</td>
<td>57.87±0.04a</td>
</tr>
<tr>
<td>S,S,BPPF</td>
<td>24.90±0.02a</td>
<td>1.22±0.0a</td>
<td>4.19±0.02a</td>
<td>6.80±0.03a</td>
<td>62.89±0.03a</td>
</tr>
<tr>
<td>S,S,FPPF</td>
<td>33.21±0.01a</td>
<td>1.40±0.01a</td>
<td>5.01±0.01a</td>
<td>8.79±0.02a</td>
<td>51.59±0.03a</td>
</tr>
</tbody>
</table>

[International Journal of Food Science and Biotechnology 2017; 2(3): 73-79 75]
[30] noted that the increase in protein value due to fermentation could be attributed to net synthesis of protein by fermenting microbes which might have resulted in the production of some amino-acids during protein synthesis. The highest protein percentage obtained in the sprouted/fermented pigeon pea flour was due to synergetic effect or extra protein produced in the pigeon pea seeds by individual sprouting and fermentation processes. This increase in protein confers nutritional advantage for the combined sprouted/fermented pigeon pea flours. Protein is used in the body for growth, healthy living, maintenance and production of tissues and cells.

The moisture content of the samples varied widely and was thought to have affected the results. Therefore, to nullify the effect of the large variation in the moisture contents of the samples, results were based on dry weight basis.

There were significant differences (p<0.05) in the fat contents of the pigeon pea flour samples (Table 1). The control sample (S_oPPF) had the highest fat content (2.39%) followed by samples S_oFPPF (2.22%), S_oBFPPF (2.19%) and S_oBFPFF (2.06%) respectively. The least fat content (1.22%) was recorded in sample S_rBFPPF followed by samples S_rBFPFF (1.30%), S_rFPPF (1.40%) and S_rPPF (1.70%) respectively.

The fat content values which ranged from 1.22% to 2.39% were different from 1.80% value reported by [23] for pigeon pea flour. This result is in agreement with the findings of [24] who reported reduced fat content for boiled pigeon pea flour. Another researcher, [7] indicated decreased fat content due to fermentation. The reduction in fat content of fermented pigeon pea flour could be attributed to the increased activities of the lipolytic enzymes which hydrolysed fat components into fatty acid and glycerol. The decrease in the fat content of sprouted pigeon pea flour samples as observed in this work was in agreement with the work of [9] who stated decrease in fat content of sprouted mung bean, pea and lentil seeds respectively. According to [13], the decrease in fat content of sprouted flour samples might be attributed to their utilization as energy sources during sprouting process. Soaking, sprouting, fermentation and boiling treatments reduced fat content of sorghum [10].

There were significant differences (p<0.05) in the ash contents, with sample S_oFPPF recording the highest ash content (5.01%) followed by samples S_oPPF and S_oFPPF which had ash contents of 4.59% and 4.58% respectively. The lowest ash content (3.27%) was recorded in sample S_oBPPF. Sample S_oBFPPF had an ash content of 4.26% which was significantly different (p<0.05) from that of sample S_oSpBFPPF (4.19%). The increase in the ash content of sprouted pigeon pea flour as obtained in this study is in agreement with that of [20] who reported increase in percentage crude ash of sprouted pigeon pea flour. Also, the increase in ash content of fermented pigeon pea flours is in agreement with the reports of [20], [12] and [7]. [7] also opined that fermentation increased the ash content of pigeon pea flour from 4.61 to 5.52% after 5 days while [12] reported an increase from 2.1% to 2.9% for oil beans seeds fermented for 3 days.

The highest fibre content (8.79%) was recorded in sample S_oBFPF, followed by sample S_rBPPF (8.30%) and sample S_rFPPF (7.36%) respectively. The least fibre content (5.68%) was recorded in sample S_oPPF, followed by the control sample S_oPPF (6.09%) and sample S_rBFPPF (6.28%) respectively. The crude fibre contents of sample S_oBPPF and sample S_rBFPFP were 6.80% and 7.0% respectively. This result showed that crude fibre increased in the sprouted, fermented and combined sprouted/fermented pigeon pea flour samples. This could be due to depletion of carbohydrate and fat contents of the flour as a result of sprouting and fermentation thereby increasing the ratio of crude fibre in the flours. [15] had earlier stated that fermentation increases the crude fibre content of Afzelia africana. Sprouting was also reported to increase the crude fibre contents of mung bean and sorghum varieties [28]; [13], [3] indicated that crude fibre content of pigeon pea flour increased from 7.16 to 7.52% (dry weight basis) after sprouting.

The carbohydrate content of the pigeon pea flour samples varied significantly (p<0.05) from one another (Table 1). The highest carbohydrate content (66.08%) was recorded for the soaked/boiled sample (S_oBFPPF), followed in that order by the soaked sample (control, S_oPPF) with a value of 62.99%, soaked/sprouted/boiled sample (S_rBPPF) (62.89%) and soaked/boiled/fermented (S_rBFPPF) sample (61.37%). The least carbohydrate content (51.59%) was recorded in the soaked/sprouted/fermented sample (S_rSpFPPF), followed by the soaked/sprouted/boiled/fermented (S_rBFPPF) sample (55.17%), soaked/fermented (S_oFPPF) sample (56.86%) and soaked/sprouted (S_rSpPFP) sample (57.87%). This result showed that carbohydrate contents of the flour samples were decreased by sprouting, combined fermentation and sprouting /fermentation processes but increased when any of these treatments were combined with boiling of the seeds. This result is in agreement with the findings of [17] and [2] who reported some losses in the carbohydrate content of chickpea cultivars after sprouting. [19] equally reported percentage decrease in carbohydrate of malted pigeon pea
due to reduction in the values of other nutrients as malting progressed. The increase in carbohydrate content of pigeon pea due to boiling agrees with the reports of [1] and [24] who indicated increase in carbohydrate during the boiling of pigeon pea seeds. This increase in carbohydrate with boiling implies higher total digestible nutrient [2].

5.2. Mineral Contents of the Pigeon Pea Flour Samples Processed By Different Methods

The results of the mineral composition of pigeon pea flour samples processed by different methods are shown in Table 2. There were significant variations (p<0.05) in the calcium contents of the flour samples. The calcium contents ranged from 122 – 176mg/100g. The highest calcium content (176mg/100g) was recorded for the soaked/sprouted/fermented flour sample (S,FPPF) which was significantly different (p<0.05) from those of soaked/fermented (S,FPPF) (162mg/100g) and soaked/sprouted (S,S,FPPF) (153mg/100g) samples at 5% level of significance. The least calcium content (122mg/100g) was recorded in soaked/boiled pigeon pea flour (S,BPPF) while the control sample (S,PPF) had a value of 141mg/100g.

The sodium contents of the flour samples ranged from 10.0 to 13.8mg/100g. The highest sodium content (13.8mg/100g) was observed in the soaked/sprouted/fermented flour sample (S,S,FPPF) while the lowest sodium content (10.0mg/100g) was recorded in the soaked/boiled flour sample (S,BPPF). There were significant differences (p<0.05) in the sodium contents of the samples.

Sprouting, fermentation and combined sprouting and fermentation processes increased the mineral contents of the pigeon pea flours with combined sprouting/fermentation processing method having the highest incremental effect (Table 2). These increments in the minerals due to sprouting and fermentation and combined sprouting/fermentation could be attributed to synthesis of minerals by increased activities of enzymes and microorganisms during sprouting and fermentation processes. The increased mineral content by sprouting, fermentation and combined sprouting/fermentation processes confer nutritional advantages on the pigeon pea flours. This is because the pigeon pea flours could be used to alleviate the problem of minerals deficiency in the diets. These results are in agreement with the work of [21] who indicated increased mineral content in bambara nut after fermentation. The increase in the minerals could be as a result of the increased metabolic activities of enzymes during sprouting which released more minerals from their bound complexes with antinutrients [18].

### Table 2. Mineral Content of Pigeon Pea Flour Samples Processed By Different Methods.

<table>
<thead>
<tr>
<th>Flour Sample</th>
<th>Calcium mg/100g</th>
<th>Magnesium mg/100g</th>
<th>Potassium mg/100g</th>
<th>Sodium mg/100g</th>
</tr>
</thead>
<tbody>
<tr>
<td>S,PPF</td>
<td>141+0.02</td>
<td>115.50+0.02</td>
<td>1205+0.03</td>
<td>11.5+0.02</td>
</tr>
<tr>
<td>S,BPPF</td>
<td>122+0.01</td>
<td>98.15+0.01</td>
<td>957+0.03</td>
<td>10.0+0.0</td>
</tr>
<tr>
<td>S,FPPF</td>
<td>162+0.02</td>
<td>145.2+0.01</td>
<td>1422+0.04</td>
<td>12.8+0.02</td>
</tr>
<tr>
<td>S,BFPPF</td>
<td>143+0.01</td>
<td>121.1+0.02</td>
<td>1324+0.02</td>
<td>10.5+0.01</td>
</tr>
<tr>
<td>S,S,PPF</td>
<td>153+0.02</td>
<td>136.50+0.02</td>
<td>1401+0.02</td>
<td>12.0+0.02</td>
</tr>
<tr>
<td>S,S,BPPF</td>
<td>140+0.01</td>
<td>118.15+0.03</td>
<td>1205+0.03</td>
<td>10.3+0.01</td>
</tr>
<tr>
<td>S,S,FPPF</td>
<td>176+0.02</td>
<td>168.28+0.02</td>
<td>1577+0.02</td>
<td>13.8+0.02</td>
</tr>
<tr>
<td>S,S,BFPPF</td>
<td>147+0.06</td>
<td>141.10+0.01</td>
<td>1307+0.02</td>
<td>11.2+0.01</td>
</tr>
<tr>
<td>LSD</td>
<td>0.017</td>
<td>0.158</td>
<td>0.026</td>
<td>0.017</td>
</tr>
</tbody>
</table>

Values are means + Standard deviations from the means. Means with different letter within a column are significantly different (p<0.05). LSD= Least significant difference

Key: S,PPF = soaked pigeon pea flour (control), S,BPFPF = soaked and boiled pigeon pea flour, S,FPPF = soaked and fermented pigeon pea flour, S,BFPPF = soaked, boiled and fermented pigeon pea flour, S,S,PPF = soaked and sprouted pigeon pea flour, S,S,BPPF = soaked, sprouted and boiled pigeon pea flour, S,S,FPPF = soaked, sprouted and fermented pigeon pea flour, S,S,BFPPF = soaked, sprouted, boiled and fermented pigeon pea flour

6. Conclusion

This result has shown that combined sprouting/fermentation processing method increased the proximate (protein, ash, fibre) and minerals (calcium, magnesium and potassium) contents of pigeon pea flour. Combined sprouted/fermented processed pigeon pea flour
has a good potential as a cheap and alternative source of protein and minerals in the diet. Consumption of this blend would therefore alleviate the problem of malnutrition prevalent in the country.

References


