

Effect of processing methods on antinutrients and oligosaccharides contents and protein digestibility of the flours of two newly developed bambara groundnut cultivars

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Abstract

The effects of malting and autoclaving on the levels of antinutrients and oligosaccharides and protein digestibility of newly developed Bambara groundnut seeds compared to Bambara groundnut commercial (BGC) seeds were investigated. The newly developed seeds (Accessions No: TVSU 5 – Bambara Groundnut White (BGW) and TVSU 146 – Bambara Groundnut Brown (BGB)) were obtained from International Institute of Tropical Agriculture (IITA), Nigeria, planted for 160 days and harvested. Autoclaving lowered ($p < 0.05$) the level of antinutritional factors of the samples. Malting was more effective in reducing the level of antinutrients with BGC exhibiting the greatest reduction values compared with that of BGB and BGW. The oligosaccharides contents of the samples were reduced ($p < 0.05$) after malting and autoclaving. Greatest loss in raffinose and stachyose content was observed after malting of BGB (82 and 71%, respectively) compared with that of BGW (76 and 56%, respectively) and BGC (67 and 64%, respectively) after malting. Also, processing methods improved the in vitro protein digestibility (IVPD) of the samples with BGB (92.31%) having the higher IVPD value than that of BGC (90.52%) and BGW (89.59%) after malting ($p < 0.05$). Malting and autoclaving was thus found to be an effective technique in improving the nutritional values of the newly developed Bambara groundnut seeds.

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Keywords

Bambara groundnut

Malting

Autoclaving

Antinutrients

Oligosaccharides

Protein digestibility

Introduction

Intense efforts are currently made in the search of cheap protein sources with good nutritional and functional properties, to attenuate the problem of protein malnutrition widely spread in developing countries (Siddhuraju *et al.*, 1996). In this regards, many studies have reported that many food grain legumes consumed in Africa are very important sources of nutrients, especially proteins and excellent sources of complex carbohydrates (Minka *et al.*, 1999; Minka and Bruneteau, 2000).

Bambara groundnut (*Voandzeia subterranean* L.) is a tropical food legume in Nigeria and other tropical areas. It is an herbaceous, intermediate, annual plant, with creeping stems at ground level. Bambara groundnut can grow under conditions unsuitable for groundnut and had tolerance for drought and poor soil (Agbenorhevi *et al.*, 2007). The seed contains about 19.60-21.42% crude protein and about 3.33-6.50% crude fat (Fadahunsi and Sanni, 2010; Okonkwo and

Opara, 2010; Fadahunsi *et al.*, 2011). It is usually fried or boiled with salt and eaten as snacks or pounded into flour and used in the preparation of soup, porridge and various fried or steamed food products such as akara, moin-moin and okpa in Nigeria. Adebowale *et al.* (2005) reported that bambara groundnut flour has been used in making bread in Zambia.

The nutritive value of legumes depends upon the processing methods, presence or absence of antinutritional or toxic factors and possible interaction of nutrient with other food component and most of the antinutritional factors can easily be removed or reduced by traditional cooking, boiling, autoclaving, malting, germination and soaking methods (Mulimani and Devendra, 1998; Singh *et al.*, 2012). It has been shown that, antinutritional and flatus factors (raffinose, stachyose, and verbascose) are removed considerably by utilizing this process in some other legumes (Singh *et al.*, 2012). The in vitro digestibility assay can provide a rapid and inexpensive means to determine and compare the

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protein digestibility of food products. This assay could be used to determine the effect of processing conditions on the protein digestibility of food products with the same formulation (Akporhonor *et al.*, 2006). The in-vitro protein digestibility of bambara bean flour was 71.58% with a multi enzymes system comprising of trypsin, chymotrypsin and peptidase (Mune-Mune *et al.*, 2007). Heat treatment increases the IVPD of legumes suggesting the presence of heat stable factors such as trypsin inhibitors which lower IVPD (Tresina and Mohan, 2012).

Consequently, bambara groundnut seed contains antinutritional factors such as trypsin inhibitors, which are a serious obstacle to the use of bambara groundnut seeds (Haddad and Allaf, 2007). Udensi *et al.* (2008) reported that phytate was not significantly reduced by autoclaving even at increased autoclaving time and phytate content in legumes has been involved in reducing the bioavailability of minerals and inhibiting the activity of several enzymes. Retention of phytic acid for cooked faba bean- liquor mixture was dependent on both cooking temperature and time (Ziena *et al.*, 1991). The tannin content of bambara groundnut can be reduced during cooking and soaking but the total energy consumption will be increased (Mazahib *et al.*, 2013). According to Adeyeye (2011), the oxalate content of bambara groundnut was 5.02-8.59 mg/100g while groundnut seed flour has 4.08-6.42 mg/100g.

Despite the great uses of bambara groundnut, much of the available data and information on the anti-nutrient and nutritional compositions are limited to the commonly used variety. This is because of the possible effects of variety/genetic origin, climate, soil, pesticides and fertilizers on the nutritional composition of the new varieties. Therefore, the aim of this study was to determine the effect of malting and autoclaving on the levels of antinutrients and oligosaccharides and protein digestibility of newly developed bambara groundnut cultivars.

Materials and Methods

Collection of raw materials

Two new varieties of bambara groundnut seeds (Accessions No: TVSU 5 – Bambara Groundnut White (BGW) and TVSU 146 – Bambara Groundnut Brown (BGB) were collected from the International Institute of Tropical Agriculture (IITA), Ibadan, Oyo State, Nigeria. The seeds were multiplied by planting on a special and demarcated farm at Kelebe, Osogbo, Osun State, Nigeria. The ripe, matured and dried pods were harvested after 160 days. The pods were sun dried and threshed to obtain the seeds. Bambara

groundnut commercial (BGC) seeds were purchased from a local market in Oyo, Nigeria. All the seeds were washed with clean tap water to remove dirt and other adhering materials, sun dried and stored in air tight containers for analysis. All chemicals used were of analytical grade and obtained from Sigma chemicals, USA

Malting

Bambara groundnut seeds (500 g) was separately mixed with distilled water (1:3 w/v) and soaked for 6 h at room temperature (32°C). The soaked sample was harvested, drained and further malted in a malting chamber for 72 h. During malting the sample was sprayed with formaldehyde to prevent mould growth. The sample was wet with water and turned twice daily to keep the samples moist. The malted bambara groundnut sample was then dried at 55°C in a hot air oven (UNISCOPE SM 9053, England) to about 10% moisture content (Hassan *et al.*, 2006), milled using Marlex Excella grinder (Marlex Appliances PVT, Daman) and sieved to pass through a 0.2 mm screen sieve (Endecott Ltd., London, UK). The flour was packaged in air tight containers 4°C for further analysis.

Autoclaving

Bambara groundnut seeds (500 g) was autoclaved at 121°C and 15 psi in sample: water ratio (1:10) for 20 min, drained, dehulled and dried at 55°C in a hot air oven (UNISCOPE SM 9053, England) to about 10% moisture content (Udensi *et al.*, 2008). The dried sample was milled using the Marlex Excella grinder (Marlex Appliances PVT, Daman) and sieved to pass through a 0.2 mm screen sieve (9229, Endecott Ltd., London, UK). The flour was packaged in air tight container 4°C for further analysis.

Preparation of raw seeds flour

Bambara groundnut seeds (500 g) was soaked in water for 30 min and dehulled by rubbing between palms. The seed coats were washed away with tap water and dried in an oven (UNISCOPE SM 9053, England) maintained at 55°C, milled using Marlex Excella grinder (Marlex Appliances PVT, Daman). The flour obtained was then passed through a 0.2 mm screen sieve and packaged in air tight containers 4°C for further analysis.

Determination of antinutrients

Oxalate was determined by the method described by Falade *et al.* (2005) with slight modification. Two grams of the sample was extracted with 190 mL of distilled water plus 10 mL of 6 M hydrochloric acid

(HCl) in boiling water for 2 h, filtered and made up to 250 mL with water. An aliquot (50 mL) of the filtrate was titrated against ammonium hydroxide (NH₄OH) until the salmon pink of the methyl red indicator changed to a faint yellow. The solution was heated to 90°C and 10 mL of 5% (w/v) calcium chloride (CaCl₂) solution was added to precipitate the oxalate overnight. The precipitate was washed free of calcium and then washed into a 100 mL conical flask with 10 mL of hot sulphuric acid (H₂SO₄) (25% v/v) and then with 15 mL distilled water. The final solution was heated to 90°C and titrated against a standardized 0.1 M potassium permanganate (KMnO₄) until a faint purple colour of the solution persisted for 30 s. The oxalate was then calculated as the sodium oxalate equivalent from the mole ratio.

Vanillin-HCl method was used to assay tannin content according to Price and Butlet (1977) method. Catechin was used as reference standard. Phytate content was determined by the method described by Wheeler and Ferrel (1971) using 2.0 g dried sample. A standard curve was prepared expressing the results as iron III nitrate (Fe(NO₃)₃) equivalent. Phytate phosphorus was calculated from the standard curve assuming a 4:6 iron to phosphorus molar ratio. The spectrophotometric method of Brunner (1984) was used for saponin analysis using 1g of sample. Standard saponin solutions (0–10 ppm) were prepared and used as reference standard. The absorbance of sample and standard solutions were read after colour development in a Jenway V6300 spectrophotometer at a wavelength of 380 nm.

Trypsin inhibitor was determined by the method of Mbata *et al.* (2009) with slight modification. Approximately, 1.0 g portion of the sample was extracted by soaking overnight at 4°C in 50 mL of 0.01 M sodium hydroxide (NaOH) and pH was adjusted to 8.4–10.0. The suspension was diluted so that 2 mL of the sample extract inhibited 40–60% of standard trypsin used in the analysis. Synthetic benzoyl-dl-arginine-p-nitroamide was used as substrate for the inhibition of trypsin. A residual enzyme activity was determined in 2 mL aliquot of the sample extracts by measuring at 410 nm. Trypsin inhibitor activity (TIA) in term of milligrams pure trypsin inhibited per sample (g) was calculated as:

$$\text{TIA} = \frac{(2.632 \times D \times A1)}{S(\text{mg})\text{pure trypsin inhibited/g sample}}$$

Where A1 = change in absorbance due to trypsin inhibition/mL diluted sample extract

D = dilution factor and S = weight of sample (g)

Oligosaccharides content determination

Five grams of sample were added to 50 ml of 70% ethanol (v/v) and stirred for 12 h. The content of the flask was filtered through Whatman No. 1 filter paper and the residue was further washed with 25 ml of 70% ethanol. The combined filtrate was evaporated in a rotary vacuum evaporator at 40°C, freeze dried and re-suspended in 10 ml of distilled water. Ten microlitres of the above syrup was spotted in triplicate on chromatographic plates (19 × 19 cm) pre-coated with cellulose powder-G. The plates were kept in a chromatographic chamber containing n-propanol: ethyl acetate: water (6:1:3) as the solvent system (Tanaka *et al.*, 1975). The developed plates were sprayed with 1% α-naphthol in ethyl alcohol containing 10% orthophosphoric acid to locate the sugar spots. For quantitative estimation, the area (2x3 cm) corresponding to each oligosaccharide was scraped and soaked in 2 ml of distilled water kept overnight and filtered through Whatman No. 1 filter paper. The eluted individual oligosaccharides (raffinose and stachyose) were estimated by the method of Tanaka *et al.* (1975).

In vitro protein digestibility determination

In vitro protein digestibility of sample was measured according to the method of Saunders *et al.* (1973). Two hundred and fifty milligrams of the sample was suspended in 15 ml of 0.1 M HCl containing 1.5 mg of pepsin, followed by gentle shaking for 1 hr at room temperature. The resultant suspension was neutralized with 0.5 M NaOH and treated with 4.0 mg pancreatin in 7.5 ml of phosphate buffer (0.2 M, pH 8.0). The mixture was shaken for 24 h at room temperature in a water bath shaker, and the undigested solids were separated by centrifugation, washed with distilled water and air dried. The protein content of the dried residue was determined by Kjeldhal procedure (AOAC, 2000) as described earlier. Protein digestibility was obtained by the following equation.

$$\text{In vitro protein digestibility (\%)} = \frac{(I-F)}{I} \times 100$$

where, I = protein content of sample before digestion and F is the protein content of sample after digestion.

Results and Discussion

Effect of autoclaving and malting on antinutrients contents of bambara groundnut seeds

Table 1 shows the effect of autoclaving and malting on antinutrients contents of BGC, BGB and BGW. Raw BGB contained a lower level of saponin

Table 1. Effect of malting and autoclaving on the antinutrients content of bambara groundnut seeds

| Samples | Time | Saponin | | Tannin | | Phytate | | Oxalate | | Trypsin inhibitor | |
|---------|-------------|------------------------|-----------|------------------------|-----------|-----------------------|-----------|------------------------|-----------|-------------------------|-----------|
| | | Total (mg/100g) | RD (%) | Total (mg/100g) | RD (%) | Total (mg/100g) | RD (%) | Total (mg/100g) | RD (%) | Total (Tiu/100g) | RD (%) |
| BGC | Raw seeds | 8.40±0.01 ^a | - | 1.65±0.01 ^a | - | 850±0.32 ^a | - | 0.39±0.08 ^a | - | 17.56±0.40 ^a | - |
| | Autoclaving | 5.00±0.01 ^b | 40 | 0.68±0.03 ^b | 59 | 778±0.01 ^b | 8 | 0.18±0.03 ^b | 54 | 2.96±0.79 ^b | 83 |
| | Malting | 4.30±0.01 ^c | 49 | 0.55±0.01 ^c | 67 | 553±0.11 ^c | 35 | 0.06±0.02 ^c | 85 | 1.14±0.46 ^c | 94 |
| BGW | Raw seeds | 9.10±0.04 ^a | - | 0.75±0.04 ^a | - | 813±1.42 ^a | - | 0.26±0.01 ^a | - | 13.15±0.03 ^a | - |
| | Autoclaving | 7.07±0.01 ^b | 22 | 0.39±0.02 ^b | 48 | 660±0.47 ^b | 19 | 0.12±0.03 ^b | 54 | 2.24±0.88 ^b | 83 |
| | Malting | 4.07±0.00 ^c | 55 | 0.29±0.02 ^c | 61 | 591±0.62 ^c | 27 | 0.09±0.01 ^c | 65 | 1.15±0.11 ^c | 91 |
| BGB | Raw seeds | 7.49±0.01 ^a | - | 1.00±0.06 ^a | - | 670±0.07 ^a | - | 0.30±0.01 ^a | - | 16.15±1.33 ^a | - |
| | Autoclaving | 5.50±0.04 ^b | 27 | 0.76±0.04 ^b | 24 | 530±0.04 ^b | 21 | 0.13±0.02 ^b | 57 | 3.07±0.16 ^b | 81 |
| | Malting | 3.12±0.07 ^c | 58 | 0.43±0.01 ^c | 57 | 490±0.06 ^c | 27 | 0.07±0.02 ^c | 77 | 2.59±0.55 ^c | 84 |

RD: Reduction

Values are means ± standard deviation of three determinations. Means in a column not sharing a common superscript letter are significantly ($p < 0.05$) different as assessed by LSD test. Bambara groundnut white (BGW); Bambara groundnut brown (BGB); Bambara groundnut commercial (BGC)

(7.49 mg/100g) than the raw BGC (8.40 mg/100g) and BGW (9.10 mg/100g). Higher significant ($p < 0.05$) reduction in saponin level was observed after autoclaving BGC (5.00 mg/100g) compared with BGW (7.07 mg/100g) and BGB (5.50 mg/100g). However malting was more effective in lowering the level of saponin in BGW (by 55%) and BGC (58%) compared with BGC (49%). The values obtained in this study were higher than the values of 0.43 mg/100g reported by Mbagwu *et al.* (2011) for bambara groundnut seeds after processing.

The tannin content was higher in raw BGC (1.65 mg/100g) than BGW (0.75 mg/100g) and BGB (1.00 mg/100g). A significant ($p < 0.05$) reduction in tannin content of BGW (by 48%) and BGB (by 24%) after autoclaving was observed while BGC was reduced by 59%. Malting reduced the tannin level of the BGC, BGW and BGB by 67, 61 and 57%, respectively, ($p < 0.05$). Malting and autoclaving were very effective treatments in eliminating the tannin content of the samples. The tannin contents of the treated samples were lower than the values reported by Adewusi *et al.* (2008) for cowpea. Tannins are polyphenols and polyphenolics compounds which are soluble in water (Kumar, 2001). The reduction of tannin during malting and autoclaving may be attributed to leaching out of phenol into water used in autoclaving and malting (Uzogara *et al.*, 1990). Also, the loss of tannins may be due to the degradation or interaction with other components of seeds, such as proteins, to form insoluble complexes (Embaby, 2010). Tannins inhibit the activities of some enzymes like trypsin, amylase, lipase resulting from the formation of complexes with protein (Adewusi, 2008).

The phytate molecule is negatively charged at the physiological pH and is reported to bind essential, nutritionally important divalent cations, such as iron, zinc, magnesium and calcium. This forms insoluble complexes, thereby making minerals unavailable for absorption (Frontela *et al.*, 2008). Raw BGC had highest content of phytate (850 mg/100 g) while sample BGB (670 mg/100 g) had lowest content. These values were higher within the range of raw and processed bambara groundnut (566-811 mg/100g) reported by Yagoub and Abdalla (2007). Malting lowered ($p < 0.05$) the phytate content of BGW to 591 mg/100 g while the phytate content of both BGC and BGB reduced to 553 and 490 mg/100 g respectively. The phytate content of the autoclaved samples reduced ($p < 0.05$) to 778, 660 and 530 mg/100mg for BGC, BGW and BGB. Autoclaving slightly decreased the level of phytate in all the samples. Onwurafor *et al.* (2013) reported that phytate content of mung bean (*Vigna radiata*) flour was reduced from 87.10 to 41.21 mg/100g with malting. The apparent decrease in phytate content during autoclaving may be partly due either to the formation of insoluble complexes between phytate or other components such as phytate- protein- mineral complexes and phytate protein or to the inositol hexaphosphate hydrolysed to penta and tetra phosphate (Siddhuraju and Becker, 2001). On the other hand, it has been reported that phytic acid content was neither affected nor increased after treatments of bitter and sweet lupin seeds (Yagoub and Adalla, 2007; Martin-Cabrejas *et al.*, 2009; Embaby, 2010).

Raw BGC had highest amount of oxalate (0.39 mg/100 g) while the oxalate content of raw BGB and

Table 2. Effect of malting and autoclaving on oligosaccharides content of bambara groundnut seeds

| Samples | Methods | Raffinose | | Stachyose | |
|---------|-------------|------------------------|----------|------------------------|----------|
| | | Concentration (g/100g) | Loss (%) | Concentration (g/100g) | Loss (%) |
| BGC | Raw seeds | 2.05±0.02 ^a | - | 1.48±0.04 ^a | - |
| | Malting | 0.68±0.01 ^b | 67 | 0.53±0.03 ^c | 64 |
| | Autoclaving | 0.71±0.14 ^b | 65 | 0.60±0.09 ^b | 59 |
| BGW | Raw seeds | 1.30±0.03 ^a | - | 0.86±0.21 ^a | - |
| | Malting | 0.31±0.02 ^b | 76 | 0.35±0.04 ^c | 56 |
| | Autoclaving | 0.32±0.07 ^b | 75 | 0.50±0.11 ^b | 38 |
| BGB | Raw seeds | 1.79±0.06 ^a | - | 1.22±0.12 ^a | - |
| | Malting | 0.33±0.02 ^b | 82 | 0.35±0.07 ^c | 71 |
| | Autoclaving | 0.35±0.10 ^b | 80 | 0.42±0.18 ^b | 66 |

Values are means ± standard deviation of three determinations. Means in a column not sharing a common superscript letter are significantly ($p < 0.05$) different as assessed by LSD test. Bambara groundnut white (BGW); Bambara groundnut brown (BGB); Bambara groundnut commercial (BGC)

BGW were 0.30 and 0.26 mg/100g respectively. The oxalate content of the samples showed appreciable reduction in oxalate level after malting and autoclaving. After malting and autoclaving, the levels of oxalate in sample BGB were reduced ($p < 0.05$) to 0.07 and 0.13 mg/100g respectively. There was a significant ($p < 0.05$) decrease in the level of oxalate to 0.18 and 0.06 mg/100g for autoclaved and malted BGC samples. The level of oxalate also decreased ($p < 0.05$) in samples BGW from 0.26 to 0.12 and 0.09 mg/100g in both autoclaved and malted samples. Onwurafor *et al.* (2013) reported that the oxalate content of mung beans flour was reduced from 6.24 mg/100g to 1.46 mg/100g after malting for 72 h while Olaleye *et al.* (2013) reported that autoclaving enhances the oxalate content of whole bambara groundnut seeds by increasing the level from 5.02 to 8.59 mg/100g.

The presence of protease inhibitors in the diet leads to the formation of the irreversible trypsin enzyme-trypsin inhibitor complex, causing a trypsin drop in the intestine and a decrease in the diet protein digestibility, leading to slower growth. Under this situation, the organism increases the secretory activity of the pancreas, which could cause pancreatic hypertrophy and hyperplasia (Liener, 1994). The trypsin inhibitors levels in both malted and autoclaved was lower than in the raw seeds. The levels of trypsin inhibitors in raw BGW, BGB and BGC were 13.15, 16.15 and 17.56 Tiu/g. A major beneficial effect of heat treatments of different seeds is the destruction of protease inhibitors, which interfere in protein digestibility. After processing treatments, the level of trypsin inhibitors was reduced ($p < 0.05$) to 2.24, 3.07 and 2.96 Tiu/g in autoclaved samples while it was

reduced ($p < 0.05$) to 1.15, 2.59 and 10.14 Tiu/g in malted samples. Adewusi *et al.* (2008) reported that autoclaving reduced trypsin inhibitors in *Mucuna flagellipes* from 108.80 Tiu/g to 18.00 (98.35% reduction) and 0.00 Tiu/g (100% reduction) after 60 and 90 min respectively. Heat treatments have been shown to be very effective in destroying trypsin inhibitor activity (Alonso *et al.*, 2000). Reactions involving deamidation splitting of covalent bonds, such as hydrolysis of peptide bonds at aspartic acid residues, and interchange or destruction of disulfide bonds, might be involved in the thermal inactivation (Alonso *et al.*, 1998).

Effect of malting and autoclaving on oligosaccharides contents of bambara groundnut seeds

The raffinose and stachyose contents of raw BGB (1.79 and 1.22 g/100g) and BGW (1.30 and 0.86 g/100g) were lower than that of BGC (2.05 and 1.48 g/100g) (Table 2). Malting the samples resulted in a significant ($p < 0.05$) reduction in the levels of stachyose and raffinose with greater loss found in BGB (82 and 71%) and BGW (76 and 56%) compared with BGC (67 and 64%). This may be due to the enzyme, galactosidase, which attack stachyose and raffinose during malting. Nnanna and Phillips (1988) reported that there was complete disappearance of two sugars- stachyose and raffinose in black gram after 72 h of germination. Germinated seeds of bambara groundnuts were found to produce low flatulence when fed to rats (Ruiz-Teran and Owens, 1996) and the low flatus produced is thus attributed to about 90% reduction in stachyose and verbascose. Also, Jood *et al.* (1985) reported that during the first 24 h of germination of black gram seeds, maximum

Table 3. Effect of malting and autoclaving on *in vitro* protein digestibility of bambara groundnut seeds

| Treatments | Sample | <i>In vitro</i> digestibility (%) |
|-------------|--------|-----------------------------------|
| Raw seeds | BGC | 56.77±0.04 ^f |
| | BGW | 64.42±0.13 ^e |
| | BGB | 68.35±0.09 ^d |
| Malting | BGC | 90.52±0.13 ^{ab} |
| | BGW | 89.59±0.06 ^b |
| | BGB | 92.31±0.03 ^a |
| Autoclaving | BGC | 89.52±0.23 ^{bc} |
| | BGW | 88.48±0.37 ^c |
| | BGB | 90.31±0.03 ^{ab} |

Values are means ± standard deviation of three determinations. Means in a column not sharing a common superscript letter are significantly ($p < 0.05$) different as assessed by LSD test. Bambara groundnut white (BGW); Bambara groundnut brown (BGB); Bambara groundnut commercial (BGC)

reduction was observed in the level of verbascose followed by stachyose and raffinose. Malting for 72 h caused considerable losses of stachyose and raffinose and results in better digestibility (Sat and Keles, 2002).

Autoclaving significantly ($p < 0.05$) reduced the levels of raffinose in BGB (by 80%) and BGW (75%) while the stachyose was reduced by 66 and 38%, respectively compared to the raw samples (Table 2). However, lower loss in raffinose (65%) level was observed in BGC as compared with BGW and BGB, after autoclaving. The results obtained compares favourably with the report of Sat and Keles (2002) where it was reported that pressure cooking was significantly more effective on sugar degradation than conventional cooking. He also observed that decreases in stachyose and raffinose contents were higher in autoclaved samples that were not pre-soaked. Sat and Keles (2002) reported that during autoclaving the cotyledons were slightly damaged and may absorb more water. In this work, malting and autoclaving has significantly reduced the level of the oligomers-stachyose and raffinose in the new seeds which tends to suggest lesser flatulence production potential in this grain legume. Also, the low occurrence of these components may be of relevance in making bambara groundnut more acceptable for incorporation into weaning foods.

Effect of malting and autoclaving on in-vitro protein digestibility of bambara groundnut seeds

Protein digestibility is a primary determinant of the availability of amino acids and, therefore, protein digestibility is important in evaluating

the nutritive quality of a food protein. The *in vitro* protein digestibility of raw BGW (64.42%) and BGB (68.35%) were higher than that of BGC (56.77%) (Table 3). Autoclaving of the seeds significantly ($p < 0.05$) increased the IVPD of BGB, BGW and BGC to 89.52, 88.48 and 89.31%, respectively. Malting was more effective in increasing ($p < 0.05$) the IVPD of the samples to a range of 89.52 to 90.31% with BGB exhibiting the value. Heat processing has been reported to increase the digestibility of protein by destroying protease inhibition (Abbey and Benezi, 1988). The improvement in the protein digestibility of all the samples was due to the leaching-out of phytic acid, tannin and polyphenols during soaking prior to malting which is known to interact with protein to form complexes. In addition, increase in protein digestibility of malted samples could be attributed to the partial degradation of complex compounds to more simple and stable products. Micro flora may produce proteolytic enzymes during malting which may be responsible for the increase in protein digestibility. Reduction of pH during malting plays an important role in enhancing native proteolytic enzymes activity and consequently promotes the breakdown of proteins to smaller polypeptides which are easily digested by enzymes (Singh *et al.*, 2012). In addition, thermal processing promoted structural changes of protein such as globulin, thereby increasing chain flexibility and accessibility to proteases (Swaisgood and Catignani, 1991).

Conclusion

Processing methods are effective in reducing the antinutritional factors of the newly developed bambara groundnut seeds with malting showing the greatest reduction effect. Malting and autoclaving lowered the oligosaccharides contents of the samples with the newly developed samples having lower values than the control sample. The IVPD of the seeds were improved after malting and autoclaving. Thus, malting and autoclaving can be considered as a beneficial technique for improving the nutritional value of the seeds.

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