Effect of Processing on Anti-nutritional Factors and Sensory Qualities of ‘Hepho’, a Black Climbing Bean (Lablab purpureus L.) Flour

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Abstract
Food quality of most legumes depends upon the processing methods applied and presence or absence of anti-nutritional or toxic factors. It is widely accepted that simple and inexpensive processing techniques are effective methods of achieving desirable qualities of processed legume seeds. The aim of this study was to evaluate the effect of processing on anti-nutritional factors (tannin and Phytate) and sensory characteristics (taste, bitterness, appearance, texture and overall acceptability) of hepho (Lablab purpureus L) flour. Hepho is the Afan Oromo name for black climbing bean which is an indigenous legume in Ethiopia. The processing techniques employed were traditional cooking (TC) and pressure cooking (PC) of the dehulled and undehulled hepho bean while the raw sample was served as a control. All the processing methods significantly reduced (P <0.05) the anti-nutritional factors under investigation. Of the two processing techniques, the reduction of the antinutritional factors by PC was evaluated to be higher than that of TC. The sensory analysis showed that there is no significant difference (P > 0.05) between dehulled TC and dehulled PC as well as unde hullled TC and unde hulled PC. But there is a significant difference (P<0.05) between dehulled and undehulled PC methods. The overall acceptability rating was higher for dehulled seeds in both the treatments. From this it can be concluded that simple processing techniques can drastically reduce some anti-nutritional factors thereby increase the biological value of legumes and the same processing techniques can result in increase of the overall acceptability of dishes produced from legumes.

Keywords: Hepho (Lablab purpureus L.), processing methods, ant-nutritional factors, sensory characteristics.

1. Introduction
Legumes are important sources major nutrients for both human and animals, including valuable but incompletely balance protein, particularly in vegetarians’ diet (Akande and Fabiyi, 2010). The nutritive value of legumes depends upon the processing methods, presence or absence of anti-nutritional or toxic factors and possible interaction of nutrient with other food components (Akande and Fabiyi, 2010).

It is well known that plants generally contain anti-nutrients acquired from fertilizer and pesticides and several naturally-occurring chemicals. Some of these chemicals are known as “secondary metabolites” and they have been shown to be highly biologically active. They include saponins, tannins, flavonoids, alkaloids, trypsin (protease) inhibitors, oxalates, phytates, haemaglutinins (lectins), cyanogenic glycosides, cardiac glycosides, coumarins and gossypol (Soetan and Oyewole, 2009). The presence of relatively high concentration of these toxic factors a ffect the nutritional quality by interacting with intestinal tract and also reduce protein digestibility and amino acid absorption (Audu and Aremu, 2011). Some of these plant chemicals have been shown to be deleterious to health or evidently advantageous to human and animal health if consumed at appropriate amounts (Soetan and Oyewole, 2009). To improve the nutritional quality and to provide effective utilization of legume grains, it is essential that anti-nutritional factors would be removed or reduced. The anti-nutritional factors need to be destroyed either by heat or other treatments otherwise concentration of toxins will exert adverse physiological effects when ingested by man and animals (Alonso et al., 2000b). In order to inactivate or reduce anti-nutrients, various conventional, simple processing methods have been used in legume seeds (Akande and Fabiyi, 2010).

Hepho is the Afan Oromo name for black climbing bean (Lablab purpureus L.) which is a common bean belonging to the Leguminosae (Fabaceae) family (Zelalem, 2002). It is an indigenous, less popular and one of the principal food and cash crop legumes grown in both lowland and medium altitude areas of Ethiopia (Shimelis and Rakshit, 2007). Hepho or lablab is well known in the North-West parts and Wollega Zones (Western and Eastern Wollega Zones of Oromia region and Bemnishangul-Gumuz region) in Ethiopia (Deres, 2012). Hepho is cultivated on a limited scale by intercropping with maize and under fence (supportive) around home (Zelalem, 2002). Its seed usually reach a harvestable stage within five to six months from planting depending on the environment and the plant dies after the seeds have matured (Tadese and Bekele, 2003).

Hepho, like most common legumes, are consumed in different forms and used for the preparation of
various diets in Ethiopia. It is consumed in different forms; such as sauce or ‘Wett’ prepared from both dehulled and undeulled seeds with addition of salts, pepper and butter. It can also be prepared in the form of ‘Nifro’ by boiling hepho bean together with maize.

Black climbing bean (Lablab purpureus L.) is one of the least exploited legumes in Ethiopia despite its high level of protein and common minerals such as phosphorus, calcium and iron (Akinjayeju and Ajayi, 2011). This low consumption of black climbing bean has been attributed partly to the high presence of anti-nutritional factors and hard-to-cook phenomenon which requires long time of cooking to make it safe and soft enough for consumption (Akinjayeju and Ajayi, 2011). Treatments like cooking and dehulling improves the nutritional quality (protein quality, palatability, digestibility), sensorial quality and reduces the anti-nutritional factors of legume beans ((Abiodun and Adepeju, 2011). For proper utilization and acceptability of legums, it is desirable to study the cooking and sensory properties since they play important role in nutrient composition and overall acceptability during preparation and processing (Akinjayeju and Ajayi, 2011). Therefore, this study was aimed to evaluate the effect of traditional cooking (TC) and pressure cooking (PC) on anti-nutritional factors (tannin and Phytate) and sensory characteristics (taste, bitterness, appearance, texture and overall acceptability) of hepho (Lablab purpureus L) flour.

2. MATERIALS AND METHODS

2.1 Sample Collection

Mature seeds of Lablab purpureus L., ‘hepho’ beans were collected from growers in Bandira, Kubena Hambelta and Horo Hambelta districts of Ethiopia and authenticated by a botanist of the department of Botany, Addis Ababa University, Ethiopia. The samples were packed in polyethylene bags, kept in an ice-box to prevent moisture loss and taken to laboratories for experimentation.

2.2 Samples Preparation

The seeds were thoroughly cleaned and sorted to remove stones and injured seeds. It was then divided into three portions and treated as follows. The first portion was treated as raw and served as a control. The second portion was manually dehulled after boiling in water for 30 minutes followed by hand-rubbing to separate the seeds from the seed coat. The dehulled seeds were then dried in oven-drier (Gallenkamp Hotbox Oven, size 2, Gallenkamp, UK) at 60°C for 8 hours. The third portion was properly cleaned and treated as undehulled. The raw was milled into fine powder using electric grinder (NIMA-8300 Burman, Germany) until to pass through 0.425 mm sieve mesh size and the dehulled and undeulled portions were subjected to the processing methods.

2.3. Traditional cooking

The dehulled and undeulled ‘hepho’ or lablab seeds were traditionally cooked separately in distilled water in the ratio 1:10 (w/v) for 1 to 2hrs until they became soft when felt between the fingers following the method justified by the local women. The cooking water was drained off and the seeds were sun dried and ground into fine powder by using an electric mill (NIMA-8300 Burman, Germany) until to pass through 0.425 mm sieve mesh size. Samples were preserved in air-tight bottles in the refrigerator for analysis.

2.4 Pressure cooking

The dehulled and undeulled seeds of ‘hepho’ or lablab were pressure cooked separately in pressure cooker at 101.31 Kpa (15 psi), 121°C in distilled water (1:5 w/v) for 15 min. The cooking water was drained off and the
seeds were sun dried and ground into fine powder by using an electric mill (NIMA-8300 Burman, Germany) until it pass through 0.425 mm sieve mesh size. Samples were preserved in air-tight bottles in the refrigerator for analysis.

2.5 Determination of anti-nutritional factors (Phytate and Tannin)

2.5.1 Determination of Phytate content

The phytate content was determined following the method described by Nwosu (2011). The phytic acid in the samples was precipitated with excess FeCl$_3$ after extraction of 1g of each sample with 100mL 0.5N HCl. The precipitate was converted to sodium phytate using 2 mL of 2% NaOH before digestion with an acid mixture containing equal portions (1mL) of conc. H$_2$SO$_4$ and 65% HClO$_4$. The liberated phosphorus was measured calorimetrically (Jenway 6051 Colorimeter) at 520nm after colour development with molybdate solution. The percentage phytate was thus calculated as:

\[
\text{% Phytate} = \left( \frac{100}{W_t} \right) \times \left( \frac{au}{as} \right) \times C \times \frac{V_t}{V_a}
\]

Where
- $W_t$ = weight of sample used
- $au$ = absorbance of test sample
- $as$ = absorbance of standard phytate solution
- $C$ = Concentration of standard phytate solution
- $V_t$ = Total volume of extract
- $V_a$ = Volume of extract analyzed

2.5.2 Determination of Tannin content

The Folin-Denis spectrophotometric method that was described by Ezegbe (2012) was used for the determination of tannin content. A measured weight of each sample (1.0g) was dispersed in 10mL distilled water and agitated. This was left to stand for 30min at room temperature, being shaken every 5min. At the end of the 30mins, it was centrifuged at 1000rpm for 5minutes and the extract was obtained. 2.5mL of the supernatant (extract) was dispersed into a 50mL volumetric flask. Similarly 2.5mL of standard tannic acid solution was dispersed into a separate 50ml flask. A 1.0mL Folin-Denis reagent was measured into each flask, followed by 2.5mL of saturated Na$_2$CO$_3$ solution. The mixture was diluted to mark in the flask (50mL) and incubated for 90min at room temperature. The absorbencies were measured at 250nm in a Genway model 6000i electronic spectrophotometer. Readings were taken with the reagent blank at zero. Then the tannin content was calculated as follows;

\[
\text{% Tannin} = \left( \frac{Au}{As} \right) \times \left( \frac{100}{W} \right) \times \frac{V_f}{V_a}
\]

Where
- $Au$ = Absorbance of test sample
- $As$ = Absorbance of standard tannin solution
- $W$ = weight of sample used
- $V_f$ = total volume of extract
- $V_a$ = Volume of extract analysed

2.6 Preparation of ‘Hepho’ Meal (Nifro) and Sensory Evaluation.

Raw, dehulled and undehulled hepho beans were subjected to traditional cooking and pressure cooking and ‘Nifro’ was prepared as practiced by the local women in Ethiopia. The ‘Nifro’ was evaluated for taste, bitterness, appearance, texture and overall acceptability. The sensory evaluation was conducted by 12-member semi-trained test panelists, of which seven (5 females and 2 males) of them were selected from ‘hepho’ consumers and five (3 females and 2 males) were among students and staffs from Department of Food Technology and Process Engineering, Wollega University, Ethiopia. All recommended preconditions for sensory tests were set by a researcher prior to start the test and the 12 panelists were kindly instructed to follow it. A 5-point hedonic scale was used to rank for taste, bitterness, appearance, texture and overall acceptability (1: like very much, 2: like moderately, 3: like slightly, 4: dislike moderately and 5: dislike very much). The sensory evaluation was conducted between 3:00 and 4:00pm during which panelists are neither too satisfied nor too hungry assuming that they had their lunch.

2.7 Statistical Analysis

The results obtained from the analyses were subjected to Analysis of Variance (ANOVA) using SPSS version 16.0 (SPSS Inc., Chicago IL, USA). Ranking preference test was used to analyze the responses of the panelists for the sensory evaluation (Akinjayeju and Ajayi, 2011). Significance difference was accepted at the 0.05 (P<0.05) level of probability.
3. Results and Discussions

3.1 Anti-nutritional factors

Phytate and tannin content of raw and processed hepho flour were presented in Table 1. All the processing methods applied in this study significantly reduced (P<0.05) these two anti-nutritional factors.

3.1.1 Phytate content

The phytate content of raw ‘hepho’ bean seeds was determined to be 304.80mg/100g and among the processing methods employed, pressure cooking was found to be more effective in reducing the phytate content. The highest reduction (66%) was observed in the dehulled PC followed by undehulled PC (61%) reduction (Table 1). This report was in agreement with the works of Akinjayeju and Ajayi, 2011) on black beans (Phaseolus vulgaris) and Osman (2007) reported pressure cooking of lablab beans caused reduction phytate content by 52.29%. In this study, the traditional cooking also reduced the phytate content where dehulled TC cause reduction by 36% followed undehulled TC by 24% (Table1). This finding was well agreed with work of Mubarak (2005) that normal boiling decreased the phytate content of mung bean by 25.36%. The evident reduction in phytate during cooking may be caused by degeneration by heat or the formation of insoluble complexes between phytate in addition to leaching into the cooking medium, and other components, such as phytate-protein and phytate-protein mineral complexes (Abiodun and Adepeju, 2011) and also the reduction might be due to the hydrolysis and heat destruction of some molecules of isositol hexaphosphate to penta, tetra and triphosphate components (Ejigui et al., 2005). The reduction of phytate during processing of bean seeds can enhance the bioavailability of proteins and dietary minerals (Soetan and Oyewole, 2009).

3.1.2 Tannin Content

The tannin content of raw ‘hepho’ bean seed was determined to be 368.04 mg/100g and among the processing methods employed, pressure cooking was found to be effective in reducing the tannin content. The reduction caused by dehulled PC was found to be 42% followed by undehulled PC which was 41% (Table1). This value was in agreement with the works of past Authors such as Mittal et al., (2012) for chickpea processed by pressure cooking. Osman, (2007) reported the reduction of tannin content of labal beans by pressure cooking was higher than the present finding (60 to 70%). Significant reduction was also observed, 27% reduction by dehulled TC. This agreed with the work of Abiodun and Adepeju, 2011) for bambara nut. This could be attributed to the predominant presence of tannin in the seed coat and it’s also heat labile and degrade upon heat treatment (Rakic et al., 2007). Reduction of tannin content in the hepho bean was expected to improve its nutritional value because tannins form complexes with proteins and reduce their digestibility and palatability Abiodun and Adepeju 2011). Tannin is known to contribute for bitterness of beans (Ajala, 2009) and hepho is much bitter when eaten undehulled.

Table 1. Anti-nutritional composition of raw and processed Hepho bean seeds (mg/100g)

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Phytate</th>
<th>Tannin</th>
</tr>
</thead>
<tbody>
<tr>
<td>Raw</td>
<td>304.81±0.72</td>
<td>368.04±2.49</td>
</tr>
<tr>
<td>DTC</td>
<td>196.19± 0.04c</td>
<td>269.57± 0.00c</td>
</tr>
<tr>
<td></td>
<td>(-36%)</td>
<td>(-27%)</td>
</tr>
<tr>
<td>UTC</td>
<td>231.90±1.41b</td>
<td>293.04±52.19b</td>
</tr>
<tr>
<td></td>
<td>(-24%)</td>
<td>(-20%)</td>
</tr>
<tr>
<td>DPC</td>
<td>103.72±1.02e</td>
<td>215.30± 0.00d</td>
</tr>
<tr>
<td></td>
<td>(-66%)</td>
<td>(-42%)</td>
</tr>
<tr>
<td>UPC</td>
<td>118.95±1.00d</td>
<td>217.90±3.16d</td>
</tr>
<tr>
<td></td>
<td>(-61%)</td>
<td>(-41%)</td>
</tr>
</tbody>
</table>

“Mean not followed by the same superscript letters in the same column are significantly different (P<0.05).

NB: DTC stands for De-hulled Traditionally Cooked, UTC for Undehulled Traditionally Cooked, and DPC for Dehulled Pressure Cooked and UPC for Undehulled Pressure Cooked.” “(+) and( -) indicate increased and decreased from raw mean”

3.2 Sensory Characteristics

The mean scores and calculated variance ratios of the sensory characteristics of “Nifiro” prepared from the dehulled TC, undehulled PC, dehulled PC and undehulled PC samples were summarized in Table 2. The mean scores obtained show that “Nifiro” prepared from dehulled seeds were more acceptable to panelists in almost all parameters evaluated as compared to that prepared from undehulled seeds which recorded higher mean scores, an indication of reduced acceptability (5 for dislike very much). There was a significant difference between dehulled samples and undehulled samples for all parameters. This finding was in agreement the earlier reported observations by Akinjayeju and Ajayi, 2011 on “moinmoin” prepared from black bean.
The low overall acceptability rating of the “Nifiro” from undehulled sample could be attributed to the high presence of tannin in hepho which is responsible for its bitter taste (Ajala, 2009) and also of beany flavor in most legumes which is prevalent mostly in the seed coat and considered offensive to most consumers (Akinjayeju and Ajayi, 2011). This can be justified from the present finding that the rating for bitterness for all samples was very low (Table 2), an indicator of high tannin content.

Table 2. Mean scores and calculated variance ratios of sensory characteristics of “Nifiro” prepared from dehulled and undehulled TC and PC ‘hepho’ seeds.

<table>
<thead>
<tr>
<th>Sample</th>
<th>DTC</th>
<th>UTC</th>
<th>DPC</th>
<th>UPC</th>
<th>F_cal</th>
</tr>
</thead>
<tbody>
<tr>
<td>Taste</td>
<td>1.42±0.67&lt;sup&gt;a&lt;/sup&gt;</td>
<td>3.08±0.73&lt;sup&gt;b&lt;/sup&gt;</td>
<td>2.00±1.04&lt;sup&gt;a&lt;/sup&gt;</td>
<td>3.16±1.11&lt;sup&gt;b&lt;/sup&gt;</td>
<td>8.55</td>
</tr>
<tr>
<td>Bitterness</td>
<td>2.42±0.92&lt;sup&gt;a&lt;/sup&gt;</td>
<td>2.92±0.67&lt;sup&gt;b&lt;/sup&gt;</td>
<td>3.16±0.08&lt;sup&gt;a&lt;/sup&gt;</td>
<td>2.83±1.40&lt;sup&gt;b&lt;/sup&gt;</td>
<td>6.75</td>
</tr>
<tr>
<td>Appearance</td>
<td>1.83±0.71&lt;sup&gt;a&lt;/sup&gt;</td>
<td>3.33±0.82&lt;sup&gt;b&lt;/sup&gt;</td>
<td>1.75±0.99&lt;sup&gt;a&lt;/sup&gt;</td>
<td>3.16±0.90&lt;sup&gt;b&lt;/sup&gt;</td>
<td>18.45</td>
</tr>
<tr>
<td>Texture</td>
<td>2.00±0.73&lt;sup&gt;a&lt;/sup&gt;</td>
<td>3.46±0.67&lt;sup&gt;b&lt;/sup&gt;</td>
<td>1.42±0.99&lt;sup&gt;a&lt;/sup&gt;</td>
<td>3.08±0.80&lt;sup&gt;b&lt;/sup&gt;</td>
<td>15.75</td>
</tr>
<tr>
<td>Overall acceptance</td>
<td>1.83±0.65&lt;sup&gt;a&lt;/sup&gt;</td>
<td>3.08±0.83&lt;sup&gt;b&lt;/sup&gt;</td>
<td>1.50±0.71&lt;sup&gt;a&lt;/sup&gt;</td>
<td>3.17±0.68&lt;sup&gt;b&lt;/sup&gt;</td>
<td>14.67</td>
</tr>
</tbody>
</table>

Means with the same letters along the same row are not significantly different at p ≤ 0.05; 1 = liked very much, 2 = like moderately, 3 = like slightly, 4 = dislike moderately and 5 = disliked very much; Ftab (From Statistical Tables) F(0.05) = 7.82. For F_cal > Ftab, there is significant difference between the processing methods.

Fig. 3. “Nifiro” prepared from ‘Hepho’ bean seeds and presented to the panellists. (Source: Photo by the researcher).

4. Conclusions
This study showed that traditional cooking and pressure significantly reduced the anti-nutritional factors under the study (phytate and tannin). Pressure cooking appreciably reduced the anti-nutritional factors as compared to traditional cooking. Dehulling and cooking was found to cause a higher reduction in the anti-nutritional factors present in ‘hepho’ seeds than the undehulled treatment. It also showed that dehulling and cooking resulted in higher overall acceptability effect on the sensory characteristics of “Nifiro” prepared from ‘hepho’ seeds. From this it can be drawn that simple processing techniques can drastically reduce some anti-nutritional factors thereby increase the biological value of legumes and the same processing techniques can result in increase in the overall acceptability or sensory qualities of dishes produced from legumes.

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