Effect of Soaking Time on the Proximate, Mineral Compositions and Anti-nutritional Factors of Lima Bean

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Abstract
Effect of soaking time on some composition of lima bean seed flour was investigated. Lima bean seeds (Phaseolus lunatus) were soaked in de-ionized water for 12, 24, 36, and 48hrs respectively followed by draining, drying and milling. The un-soaked seeds were milled and served as control. Proximate and mineral compositions as well as Phytate, tannins and oxalates were determined using standard procedures. Results were compared with the raw and soaked lima bean respectively. The moisture levels in lima bean increased with soaking time from (11.35% to 15.56%) and were significantly different (P=0.05) in all samples. Variation in Protein were not significantly different (P=0.05) ranging from 7.92% to 10.70%. All the mineral contents increased significantly (P=0.05) with increase in soaking time. Consequently, all the anti-nutritional factors investigated reduces significantly with soaking time (P=0.05).

Keywords: Anti-nutritional factors, Lima beans, Mineral Compositions and soaking time.

Introduction
Legumes belong to the family leguminosae in the tropics, legumes are next important crops after cereals [1]. They are low cost dietary vegetables proteins and minerals when compared with animal products, such as meat, fish, and egg.[2].

Indigenous legumes therefore are an important source of affordable alternative protein to poor resource people in many countries [3], especially in Africa and Asia where they are predominantly consumed. In the developing countries, research attention is being paid to better utilization of legumes in addressing protein malnutrition and food security issues.

Lima bean (Phaseolus lunatus) is one of the lesser known legume grown for its seed and has been limited in use due to the presence of anti-nutritional factors commonly found in legumes. Included among these anti-nutritional factors are trypsin inhibitors, which inhibits the proteolytic activity of the digestive enzyme trypsin and can lead to reduced availability of amino acids and reduced growth [4]. Tannins are known to inhibits the activities if some enzymes like trypsin, amylase and lipase, [5] resulting from the formation of complexes with protein . Phytate content of legumes has been known to lower the bio- availability of minerals [6], [7] and inhibits the activity of several enzymes [8]. Saponins when present in large quantity in food legumes impart bitter taste to the plant foods [9]. Reduction to safe level of the anti-nutritional factors is essential to improve the nutritional quality of lima bean and effective utilization of its full potentials as human food. There is limited information on processing effects on the anti-nutrients in the seeds especially our traditional food processing practices.

The objectives of this study is therefore to investigate the effect of soaking period on the proximate, mineral composition and anti-nutritional factors of lima bean (Phaseolus lunatus).

Materials and Methods.
Matured dried seeds of lima bean were purchased from Ikere central market, Ikere –Ekiti Nigeria. The seeds were processed as follows. Five hundred grammes (500) of whole seeds were soaked in distilled water at room temperature (25-32) °C in a 1:30 (bean : water) ratio for 6, 12, 18, and 24 hrs respectively. The soaked seeds were drained , rinsed, dehulled and dried at 55°C to about 10 % moisture content. The dried seed were milled in attrition mill, sieved to pass through 1mm mesh size and packaged in polyethylene container for further analysis.

Chemical analysis.
The tannins content of the seed flours was determined by modifying the procedure of[10], while phytic acid was determined by the method of [11].The oxalate was determined according to the method of [12]. The proximate compositions (crude protein, moisture , fat content and ash) was determined using [13]. The mineral composition was determined using flame photometer (potassium and sodium) while calcium and Magnesium was by the varsanate EDTA complexion titration method of [14] and phosphorus using molybdovanadate (yellow) Spectrometry described by [14].

Results and Discussion.
The proximate compositions of un-soaked and (control ) soaked lima bean seeds is shown in Table 1. Results from this study showed that soaking lima bean seeds for varying periods increased the percentage moisture content from 11.35% (raw sample) to 15.56% after 48hrs of soaking. There was a significant difference (P=0.05) in the moisture content of all the samples with respect to soaking time. A previous study carried out by [15], showed that increase in percentage moisture was a function of treatment in soaked sorghum. There was no...
significant difference in (P=0.05) in the percentage of protein content of both soaked and un-soaked lima bean. This might be as a result of fermentation of the soaked bean seed O. [16] who reported on fermented African yam bean showed that fermentation had no effect on the crude protein content of the African yam bean. [17], reported that fermentation process usually do not significantly change the total protein content and amino acid composition of substrate from the results, there was an increase in the percentage protein at 12hrs of soaking. This might be attributed to the presence of fermenting micro-flora in the soaking water which led to the increase in the protein content.

There was no significant difference (P=0.05) in the percentage of ash content of the un-soaked lima bean and samples soaked at 48hr, samples soaked for 36hrs showed an increase in the ash content. The percentage fat from the result was highest in lima bean soaked for 24hrs and decreased after 36hrs of soaking, this confirms to the result of [15], who observed increase in fat content after 12hrs of soaking sorghum. The percentage crude fibre content increases with the soaking time from 2.00 to 9.21% after 48hrs of soaking. This was contrary to [18], who reported that there is a decrease in crude fibre content of maize during fermentation. There was a significant increase in the crude fibre as soaking time increases. Generally the percentage carbohydrate level varied throughout the soaking periods and reduces on soaking from 71.91 to 66.77%.

The effect of soaking on the levels of anti-nutritional factors in lima bean is presented in Table 2. Generally soaking reduced all the anti-nutritional factors investigated to different levels with respect to soaking time. This might be as a result of leaching of the anti-nutrients in the soaked water. This conforms to the previous study carried out by [19], who observed that soaking cowpea in both acidic and alkaline solutions led to decrease in phytic acid. Phytate content was lowest in the sample soaked for 36hrs and 48hrs (0.291 %) and was highest in raw lima bean seed flour (0.415 %). [20] reported that phytates get reduced during soaking. The reduction in tannin content during soaking might be due to the leaching out of polyphenols into the soaking water under the influence of concentration [21], [22] since the tannin are polyphenols and polyphenolic compounds are water soluble in nature and mostly located in the seed coat [23].The percentage tannin in the raw lima bean was 0.499% and reduces to 0.251% after 48hrs of soaking. The oxalate content equally reduces from 0.542% to 0.325% after 48hrs of soaking.

### Table 1: Proximate composition of unsoaked lima bean and soaked lima bean seed flour.

<table>
<thead>
<tr>
<th>Soaking time( hrs)</th>
<th>Moisture (%)</th>
<th>Protein (%)</th>
<th>Ash (%)</th>
<th>Fibre(%)</th>
<th>Fat (%)</th>
<th>Carbohydrate(%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>11.35c</td>
<td>8.61c</td>
<td>3.29a</td>
<td>2.00a</td>
<td>2.19a</td>
<td>72.56a</td>
</tr>
<tr>
<td>12</td>
<td>12.00d</td>
<td>10.08a</td>
<td>2.71c</td>
<td>3.00c</td>
<td>2.15c</td>
<td>70.06b</td>
</tr>
<tr>
<td>24</td>
<td>13.25c</td>
<td>9.95b</td>
<td>3.21a</td>
<td>6.81e</td>
<td>2.01b</td>
<td>64.77f</td>
</tr>
<tr>
<td>36</td>
<td>14.36d</td>
<td>7.92a</td>
<td>3.02a</td>
<td>6.96e</td>
<td>2.00b</td>
<td>65.74p</td>
</tr>
<tr>
<td>48</td>
<td>15.56a</td>
<td>8.91c</td>
<td>3.01b</td>
<td>9.21a</td>
<td>2.01b</td>
<td>61.30l</td>
</tr>
</tbody>
</table>

Means with different superscript within the same column are significantly different (P=0.05).

### Table 2: Level of anti-nutritional factors in unsoaked and soaked lima bean seed flour.

<table>
<thead>
<tr>
<th>Soaking time (hrs)</th>
<th>Phytate (%)</th>
<th>Tannin (%)</th>
<th>Oxalate (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>0.415a</td>
<td>0.499b</td>
<td>0.542b</td>
</tr>
<tr>
<td>12</td>
<td>0.406w</td>
<td>0.389c</td>
<td>0.510d</td>
</tr>
<tr>
<td>24</td>
<td>0.321c</td>
<td>0.361c</td>
<td>0.453c</td>
</tr>
<tr>
<td>36</td>
<td>0.291w</td>
<td>0.351c</td>
<td>0.391b</td>
</tr>
<tr>
<td>48</td>
<td>0.291w</td>
<td>0.251c</td>
<td>0.231c</td>
</tr>
</tbody>
</table>

Means with different superscript within the same column are significantly different (P=0.05).

The mineral composition of unsoaked lima bean seed flour are shown in Table 3. Generally most mineral content increases upon soaking. This might be due to the decrease in the anti-nutritional factors with increase in soaking time as shown in Table 2. The anti-nutritional factors leached into the soaking water, thereby decreasing there concentration in the lima bean seed, thus releasing more minerals from their organically bound complexes due to their decreased concentration as these anti nutritional factors bind with minerals. This increase in mineral content is confirmed by the work of [18], who observed increase in mineral content of yellow maize after fermentation of the soaked seeds in water.

Sodium (Na) was highest in lima bean seed soaked for 48hrs (8.82%) and least in raw lima bean (3.98%). Potassium (K) was higher in lima bean soaked for 48hrs (9.08%) and least in raw lima bean (4.92%). Phosphorus (P) was highest in lima bean soaked for 48hrs (5.09%) and least in lima bean soaked for 12hrs (4.08%). This shows that at 12hrs, phosphorus still remained unchanged because soaking at that time was not sufficient. Concentration of calcium was increased with soaking time from (3.91 to 5.07%). Magnesium content increased from 7.02 to 8.64% as the soaking time increases.
CONCLUSION

Results from this study show that increase in moisture content with respect to time is a function of soaking, and there is significant reduction in the level of the antinutrient as soaking time increases. Therefore soaking and discarding of the soaked water has helped tremendously in increasing the mineral composition of lima bean as well as reducing the antinutrients which could be of health hazards to the consumer.

Table 3: Mineral composition of unsoaked and soaked lima bean seed flour.

<table>
<thead>
<tr>
<th>Soaking time (hrs)</th>
<th>Sodium (Na) (%)</th>
<th>Potassium (K) (%)</th>
<th>Phosphorus (P) (%)</th>
<th>Calcium (Ca) (%)</th>
<th>Magnesium (Mg) (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>3.98&lt;sup&gt;a&lt;/sup&gt;</td>
<td>4.92&lt;sup&gt;d&lt;/sup&gt;</td>
<td>4.08&lt;sup&gt;c&lt;/sup&gt;</td>
<td>3.91&lt;sup&gt;d&lt;/sup&gt;</td>
<td>7.02b</td>
</tr>
<tr>
<td>12</td>
<td>4.62&lt;sup&gt;b&lt;/sup&gt;</td>
<td>5.04&lt;sup&gt;c&lt;/sup&gt;</td>
<td>4.08&lt;sup&gt;c&lt;/sup&gt;</td>
<td>4.19&lt;sup&gt;d&lt;/sup&gt;</td>
<td>7.08b</td>
</tr>
<tr>
<td>24</td>
<td>5.02&lt;sup&gt;b&lt;/sup&gt;</td>
<td>5.61&lt;sup&gt;c&lt;/sup&gt;</td>
<td>4.92&lt;sup&gt;b&lt;/sup&gt;</td>
<td>4.81&lt;sup&gt;b&lt;/sup&gt;</td>
<td>7.08&lt;sup&gt;b&lt;/sup&gt;</td>
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<tr>
<td>36</td>
<td>8.82&lt;sup&gt;a&lt;/sup&gt;</td>
<td>8.71&lt;sup&gt;b&lt;/sup&gt;</td>
<td>5.04&lt;sup&gt;c&lt;/sup&gt;</td>
<td>5.06&lt;sup&gt;c&lt;/sup&gt;</td>
<td>8.04&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
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<td>48</td>
<td>8.82&lt;sup&gt;a&lt;/sup&gt;</td>
<td>9.08&lt;sup&gt;a&lt;/sup&gt;</td>
<td>5.09&lt;sup&gt;c&lt;/sup&gt;</td>
<td>5.87&lt;sup&gt;d&lt;/sup&gt;</td>
<td>8.64&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

Means with different superscripts within the same column are significantly different (P=0.05).

REFERENCES


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