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INFLUENCE OF ALKALINE-COOKING ON THE PROXIMATE COMPOSITION AND FUNCTIONAL PROPERTIES OF UNDER-UTILISED SEED OF OIL BEAN (*HEXALOBUS CRISPIFLORUS)* FLOUR

Adebola Ajayi [1], Oluwagbemiga K. Sokeye[1] Basheer F. Umar [2] and Olalekan J. Adebowale[1]
[1]Department of Food technology
[2]Department of Science Laboratory Technology,
The Federal Polytechnic, P.M.B. 50, Oja-Odan Road, Ilaro, Ogun State, Nigeria
E-mail address: jimiolalekan@yahoo.com

ABSTRACT

The effect of alkaline-cooking on the proximate composition and functional properties of two under-utilised seeds flour were studied using a 2 x 4 factorial experimental design with cooking time (0, 30 min) and lime concentrations (0, 0.33, 0.5 and 1.0%). Proximate composition (crude protein, crude fat, moisture, ash and crude fibre), pH, energy value and the functional properties (Bulk density, water absorption, oil absorption, foaming capacity and swelling power) were determined using standard methods. The cooking time and lime concentration significantly ($p \leq 0.05$) influenced the moisture, pH, ash, protein and crude fat of the samples. Nixtamalization significantly ($p \leq 0.05$) increased the foaming capacity (1.0 – 2.4%) and swelling power (2.9 – 3.7%). Conversely, there were decrease in the values obtained for the bulk density (0.7 – 0.5g/cm^3), water absorption (5.2 – 4.2ml/g) and oil absorption capacity (6.0 – 5.6ml/g). The effect of lime concentration on the foaming capacity, swelling power, ash and protein were not too significant ($p \leq 0.05$); however, a slight increase in protein content with pronounced fat contents were observed.
***Keywords**: Hexalobus crispiflorus, alkaline-cooking, nixtamalization, lime concentration proximate composition, functional properties.*

INTRODUCTION

There have been greater efforts at making the under-utilised legume and oil seeds to have valuable usage in the developing countries, particularly Africa where they are highly abundant. This development emanated as a result of the prevalent malnutrition in Africa due to lack of sufficient animal protein, hence the search for alternative sources of protein from lesser-known legume and oil seeds in lieu of expensive and scarce animal protein (Adebowale and Lawal, 2004). These under-utilised legumes and oilseeds are usually found in the rural communities of the developing countries and have their respective native names as well as their uses in the local context. However, with the collaborative efforts of the researchers from various institutions, the botanical names of most of these legumes and oilseeds were established.

Amongst these under-utilised seeds, that were found in the south western Nigeria are *Hexalobus crispiflorus* (oilseed), *Cajanus cajan* (pigeon pea), *mucuna pruriens* (mucuna bean), *Sphenostylis stenocarpa (*African yam bean*)* and *Vicia faba* (Broad bean), with the former being consumed majorly by the Yewa people of south western Nigeria, where it is referred to as '*Apara*' by the natives. The cotyledon, after dehulling is boiled to soften, salted and eaten as local snack and could also be grinded into paste and used as thickener in soup. This development has stimulated research on the utilisation of some of these under-utilised seeds in Nigeria, such as winged bean (Okezie and Bello, 1988); jack bean (Adebowale and Lawal, 2003, 2004); pigeon pea (Oshodi and Ekperigin, 1989; Akoja, 2008); African yam bean (Adeyeye et al., 1994; Eromosele et al., 2008; Arogundade et al., 2009). A major drawback to the use of these seeds in the communities is the length of time it takes to soak, cook and prepared them. In many cases these seeds because of their hard coat required overnight soaking followed by at least an hour to two or more of preparation. The fact now is that, consumers are striving to have greater varieties and healthier foods in the diet with the convenient of ready-to-eat and fast cooking meals (Boyce *et al*., 2010).

Alkaline cooking with lime, traditionally called nixtamalization, is the primary processing step during manufacture of several maize products (Sefa-Dedeh *et al.,* 2004). The process of nixtamalization is popular in Mexico and Central America and has been applied in maize for centuries which have been reported to have led to increased bioavailability of niacin, improved protein quality, increased calcium and reduction of aflatoxin concentrations in foods (Afoakwa et al*.,* 2007). Previous work by Sefa-Dedeh *et al.* (2001) showed a drastic reduction in Amylograph viscosity when nixtamalized maize dough (*masa*) was fermented. The process has been adopted as a means of resolving the problem of low protein quantity of cereal-based food caused by the high starch content of cereals, cowpea fortification of traditional foods in Ghana and has led to improvements in protein quantity and quality of foods which are usually made from maize (Sefa-Dedeh et al*.,* 2003). Likewise, other studies conducted by Afoakwa et *al.* (2002) reported that the combined use of fermentation and cowpea fortification led a significant improvement in the nutritional quality of maize-based foods. Though, the alkaline cooking process is not new to the people of south western Nigeria as the use of potash is common in cooking cowpea, basically to hasten its softness. Alkaline cooking can therefore be employed to improve the nutritive quality of the staples. Therefore, the rationale of this study was to investigate the influence of alkaline cooking on the proximate composition and functional properties of underexploited oilseeds in south western Nigeria.

MATERIALS AND METHODS

Materials

Hexalobus crispiflorus seeds used for this work were obtained from a local market in Ilaro, Ogun state, Nigeria. The seeds were cleaned and sorted to remove foreign materials, infested and immature seeds. Cleaned seed were then stored at 4^0C (relative humidity 65-100%). Lime [$Ca(OH)_2$] laboratory grade (Sigma Chemicals,USA) was used.

Methods

Experimental design and sample preparation

A 2 × 4 factorial experimental design with cooking time (0, 30 min) and lime concentration (0, 0.33, 0.5 and 1.0%) as described by Sefa-Dedeh *et al.* (2004) was adapted for this investigation. Oilseed (500g) was steeped in the lime solutions for 12 h. Another batch of the samples was cooked in lime [$Ca(OH)_2$] solution at 0, 0.33, 0.5 and 1.0% concentrations, respectively, for 30 min. The boiled samples were then steeped in the cooked liquor for 12 h. After steeping, the seed samples were washed thoroughly to remove the excess lime. The nixtamal obtained was drained and milled with a disc attrition mill to produce dough. The dough was dried in an air oven at 50^0C for 14 h to moisture content between 9 and 12%. The dried samples were milled into fine flour (250µm) using a hammer mill and the flour sample was analyzed.

Chemical analysis

The moisture, crude protein (N ×6.25), ash, crude fat, crude fibre contents and pH were determined by the Association of Official Analytical Chemists Approved Methods (AOAC, 2000). Carbohydrate was determined by difference [100 – (%moisture + %protein + %ash + %fat + %fibre)]. Energy value was calculated using the Atwater factors (4 × %Carbohydrate + 9 × %Fat + 4 × %Protein) (Osborne and Voogt, 1978). Determinations were done in triplicate.

pH

Exactly 10g of dried flour was mixed with 100 ml distilled water. The mixture was allowed to stand for 15 min, shaken at 5 min intervals and centrifuged at 3000 rpm for 15 min using the centrifuge (KOKUSAN model H-103N series, Kokusan Inc., Tokyo, Japan). The supernatant was decanted and its pH was determined with digital pH meter (Model 250 pH meters, USA).

Functional Properties

Bulk density

This was carried out using the procedure of Narayana and Narasinga (1984). A specified quantity of the flour sample was transferred into an already weighed measuring cylinder (W_1). The flour sample was gently tapped to eliminate space between the flour and the level was noted to be the volume of the sample and then weighed (W_2). The study was conducted in triplicate.

$$\text{Bulk density } (g/cm^3) = \frac{W2 - W1}{Vol.\ of\ sample}$$

W_1= weight of sample before tapping; W_2 = weight of sample after tapping

Water and Oil absorption capacity

Water and oil absorption capacities were determined by the method of Sathe et al. (1982). A 0.5g milled seed flour sample was added to each of 5 ml distilled water and 5 ml oil (Executive Chef Unilever Plc) in separate 10 ml graduated centrifuge tubes. The mixtures were stirred with glass rods to disperse the samples in both solvents. After standing for 30 min at room temperature (27±2^{0}C), the mixtures were centrifuged at 5000×g for 30 min. Volumes of both supernatants were noted, and then excess water or oil absorbed were expressed as the percentage water or oil bound by 100g sample. Densities of water and oil used were 1g/ml and 0.88g/ml respectively. Determinations were done in triplicate.

Foaming capacity and stability

The foaming capacity and stability were studied according to the method of Okezie & Bello (1988). 1g of each flour sample was blended with 50 ml distilled water for 5 min. The whipped mixture was transferred into a graduated cylinder gently. Foam volume changes in the cylinder were recorded at intervals of 0, 30, 60, 90 and 120 min to obtain foam capacity of the sample.

$$\text{Foam capacity (\%)} = \frac{volume\ after\ whipping\ (ml)\ x\ volume.before\ whipping\ (ml) x\ 100}{Volume\ before\ whipping}$$

Swelling power

This was determined with the method described by Oloyo and Akoja (2005) with little modifications. One gram of the flour sample was mixed with 10 ml distilled water in a centrifuge tube and heated at 80^0C for 30 min. This was continually shaken during the heating period. After heating, the suspension was centrifuged at 1000×g for 15 min. The supernatant was decanted and the weight of the paste taken. The swelling power was calculated as:

$$\text{Swelling power} = \frac{weight\ of\ the\ paste}{Weight\ of\ dry\ flour}$$

Statistical analysis

The studies were carried at out at room temperature (25 ± 2^0C) in three replications and an average of the three replicates. Statistical analyses were performed using SPSS 12.0 for windows (SPSS Inc., Chicago, IL). Differences between means were evaluated as significant at $p \leq 0.05$.

Results and Discussion

Moisture content

The moisture contents (MC) of the lime-treated *Hexalobus crispiflorus* seed flour samples increased with increasing lime concentration, an indication that lime facilitates the absorption of water (Sefa-Dedeh et al., 2004). The MC of lime-treated sample flours ranged marginally from 6.0% to 8.0% while that of untreated sample flour was 5.0%. The seed sample absorbed more water at lime concentration from 0% and 0.33%, and increased slightly too maximum moisture absorption at a lime concentration of 1.0%. The highest seed moisture absorption was observed with the 1.0% lime concentration treatment. Samples treated with 0.33% and 0.5% had slightly lower MC than that treated with 1.0% lime. Sefa-Dedeh et al. (2004) reported that, in the presence of lime there is possibility of an osmotic potential developing in the grain and this will cause the maize to absorb more water until equilibrium is attained. The higher moisture content of the lime-treated samples can therefore be attributed to osmotic effects. The effect of lime on moisture absorption has also be studied by Chang and Hsu (1985), who reported that maize cooked in lime solution absorbs more water than maize cooked in ordinary water.

Analysis of variance showed that cooking and lime concentration significantly ($p \leq 0.05$) the moisture content of the nixtamal (figure 1). Multiple range analysis of the results revealed that alkaline-cooking had a significant effect on the moisture content of the sample. This is an indication that, the effect of the lime concentration on the moisture content of sample treated with lime was significantly different from the untreated sample (0% lime or raw sample). The effects of the various concentrations of lime on the moisture content were, however, comparable (Sefa-Dedeh et al., 2004).

Protein content
The protein content increased gradually from 22.6% in the raw sample to a range of 23.9-25.7% in the sample treated with various lime concentrations (figure 1). Highest protein content was recorded in sample treated with 1.0% lime. Most researchers have reported a small increase in nitrogen content of nixtamal, which has been attributed to a concentration effect. Bressani et al. (1958) reported increased protein content from 9.6%, 10.3% and 10.7% for raw maize, nixtamal and tortilla, respectively. Work done by other researchers' showed comparable amount of protein, when alkaline-cooked corn products were compared to original grain (Gomez et al., 1987; Sefa-Dedeh et al., 2004; Serna-Saldivar et al., 1987).

When the data from protein measurements were subjected to analysis of variance, the results indicated that the treatment had significant effect ($p \leq 0.05$) on the protein content of the flour produced. This could be due to the fact that the changes in protein quality of the lime-treated sample were slight.

Ash
Trends observed in the ash contents were 2.0% and a range from 2.0% to 5.0% for untreated and lime-treated samples respectively (figure 1). The ash content of the sample flour treated with lime increased as lime concentration was increasing. The highest ash content was observed with 1.0% lime-treated sample flour. This trend was similar to the pattern observed in the moisture content determinations. Some findings on changes in ash contents of nixtamal have showed an increase in total ash content from maize to tortilla (Sefa-Dedeh et al., 2004). Statistical analysis conducted on the results, indicated that the effect of cooking and lime concentration on the ash content of the treated samples was, however significant ($p \leq 0.05$).

Fibre
Values of 4.2% and a range from 3.6% to 3.8% were observed for untreated and lime-treated sample flours respectively (figure 1). There was decrease in the fibre content with increasing lime concentration on the samples. Lowest fibre content was recorded with sample treated with 1% lime.

Fat
The fat content increased largely from 38% in untreated sample to 46% in 0.33% lime treated. Slight increment was observed in treated sample from 46% to 48% at 0.33% to 0.5% lime treatment respectively (figure 1). However, highest fat content was recorded as 52% for sample treated with 1% lime concentration. This trend could be a better processing method to increase the yield of oilseed during extraction, as it may not be unconnected with the fact that the lime treatment could have ruptured the oil cells in the seed sample.

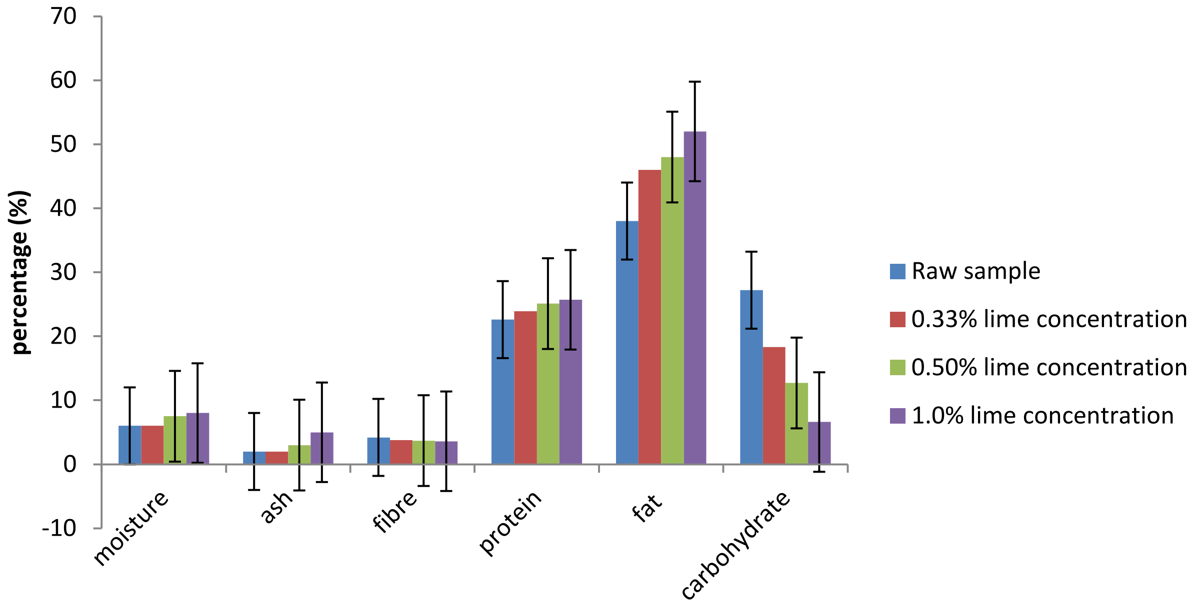

Figure 1. Effect of nixtamalization on the proximate composition of *Hexalobus crispiflorus* seed flour

pH

The pH of the nixtamalized *Hexalobus crispiflorus* seed flour samples ranged from 5.88% to 6.42%. Sample soaked in water without lime had the lowest pH value was 5.67% while that boiled in 1% lime for 30 min and steeped in the cooking liquor had the highest. This observation was in agreement with the findings of Sefa-Dedeh et al. (2004), where the highest pH was recorded in maize boiled in 1% lime for 30 min. The pH of the lime-treated *Hexalobus crispiflorus* seed flour increased with increasing lime concentration (figure 2), which may be due to absorption and retention of lime. Work done by Serna-Saldivar et al. (1990) showed that the pH of alkaline-cooked maize and its products, is closely related to the amount of lime used and retained during cooking and steeping. However, Bedolla and Rooney (1984) reported that the nixtamalized maize flours from commercial markets ranged from 7.1 to 7.4 and in this experiment, pH ranged 5.88 – 6.42 was obtained for the *Hexalobus crispiflorus* samples boiled in alkali. This reason may not be unconnected with the fact that *Hexalobus crispiflorus* is an oilseed material.

Analysis of variance showed that cooking and lime concentration had a significant effect (p≤o.05) on the lime treated *Hexalobus crispiflorus* seed flour samples. Multiple range analysis on the effect of lime concentration revealed that pH values of the samples treated with lime were comparable and significantly different from those treated without the lime (0% lime or raw sample).

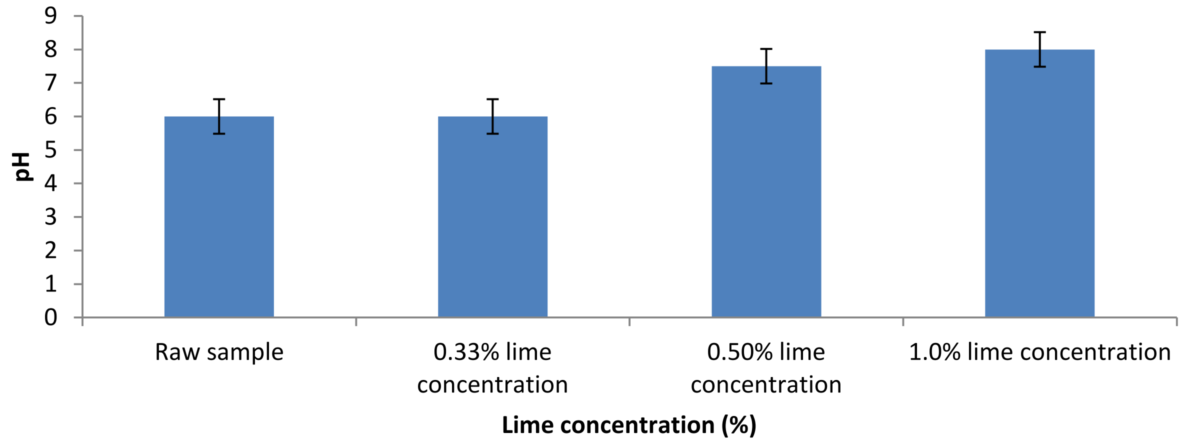

Figure 2: Effect of lime concentration on pH of raw and nixtamalized *Hexalobus crispiflorus*

Energy value

The energy value in the nixtamalized sample flours increase marginally with increasing lime concentration between 582.8 and 597.5kcal/100g flour (figure 3). Untreated sample flour had a value of 541.2kcal/100g flour sample. Highest energy value was recorded in sample flour treated with 1% lime for 30 min. This gradual increase in these energy values may not be unconnected with the earlier values recorded for the protein and fat (figure 1) which, similarly increased as lime concentration increases. The protein and fat values (%) were however used in the computation of the energy values (kcal/100g sample).

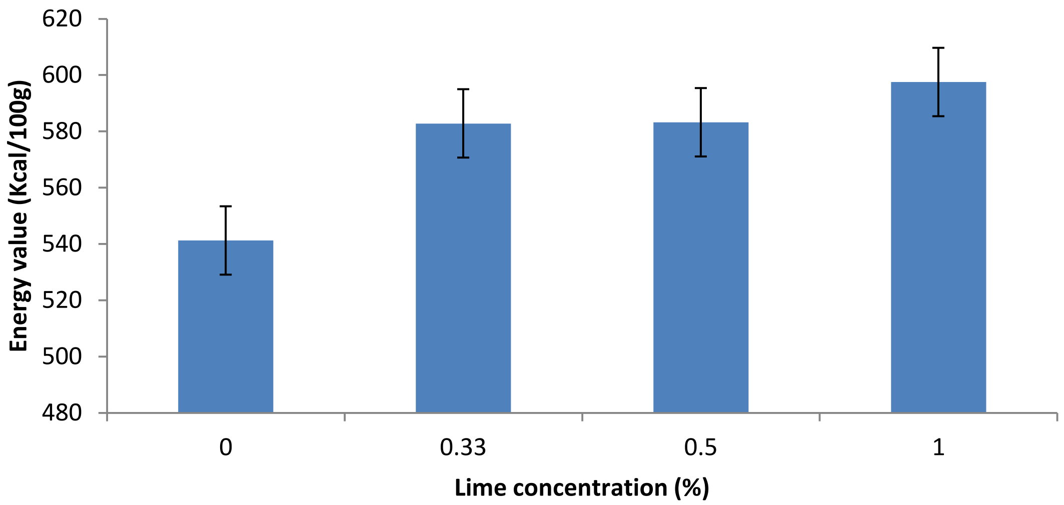

Figure 3. Effect of nixtamalization on the energy value of *Hexalobus crispiflorus* seed flour

Water and oil absorption capacity

The water absorption capacity (WAC) and the oil absorption capacity (OAC) of the samples decreased marginally with increasing lime concentration, this is not a reflection of the earlier values obtained in moisture content which increased gradually (figure 1). The WAC and OAC for the raw sample were higher than those of nixtamalized samples (figure

4). This is however contrary to the reports of Sefa-Dedeh et al. (2004), which showed reversed trends of increased WAC with increasing lime concentration while that of the cooked samples decreased a minimum and increased when it was cooked in 1% lime. The different trends observed in WAC of the alkaline-treated flours could be attributed to the effect on gelatinization, as well as Ca^{2+} and $Ca(OH)^{+}$- starch interaction (Sefa-Dedeh et al., 2004). Also, the starch hydroxyl sites in the sample might have been saturated, resulting in the decreased water absorption observed.

Analysis of variance on the data indicated that lime concentration and cooking significantly affected (p≤0.05) the WAC and OAC of the sample flours. The lime treatments however, did not have same effect on sample flours.

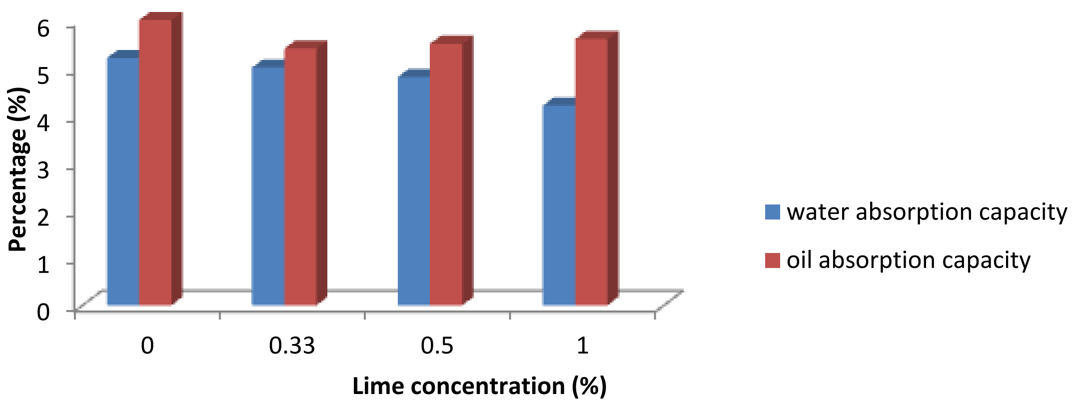

Figure 4. water and oil absorption capacity as affected by lime concentration

Foaming capacity and swelling power

Both the foaming capacity (FC) and swelling power (SP) were observed to increase with increasing lime concentration in the sample flours. FC of the flour samples ranged from 1.6% to 2.4% with the sample flour nixtamalized in 1% lime for 30 min and steeped in the cooking liquor had the highest (figure 5). Similarly, SP values ranged between 3.1% and 3.7% for the nixtamalized sample flours and highest value was recorded when sample flour was nixtamalized with 1% lime for 30 min.

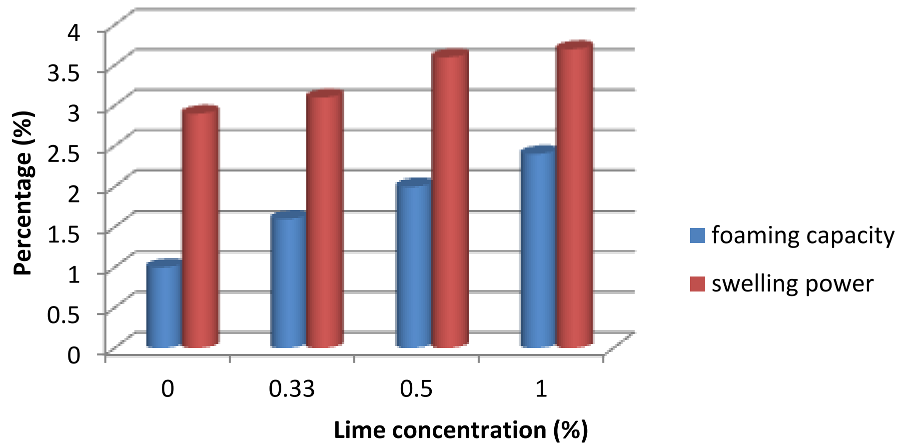

Figure 5. Effect of lime concentration on the foaming capacity and swelling power of Hexalobus crispiflorus seed flour

Bulk density

There was a slight decrease in the values of bulk density obtained as lime concentration increases. Values of 0.7g/cm^3 and 0.5-0.6 g/cm^3 were recorded for untreated and nixtamalized sample flours respectively (figure 6). The values decreased steadily with increasing lime concentration, however the values became constant between 0.50% and 1.0% lime concentration.

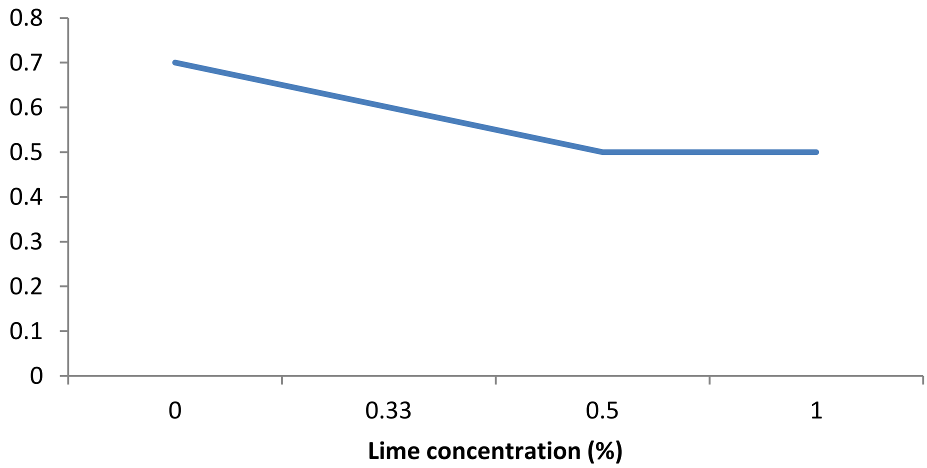

Figure 6. Influence of the lime concentration on the bulk density (%) of Hexalobus crispiflorus seed flour

CONCLUSION

Alkaline-cooking of the *Hexalobus crispiflorus* seed flour resulted in increased pH, moisture, foaming capacity and swelling power. Nixtamalization improved the energy value, protein and fat contents of the lime-treated sample flours. Hence, nixtamalization can therefore be employed in the improvement of the nutritional quality of *Hexalobus* *crispiflorus* seed flour-based traditional foods as well as widen the product base of the seed processing

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