Abstract: Bambara groundnut (*Vigna subterranea* (L.) Verdc.) originated in West Africa but has become widely distributed throughout the semi-arid zone of sub-Saharan Africa; it has appealing flavour and a high nutritive value comparable with other widely consumed legumes. In this study, the effect of moisture content on some quality parameters of stored bambara nut flour was investigated. Cream coloured bambara nut samples were cleaned by picking out dirt and other unwanted particles and processed into flour by milling and sieving. The initial moisture content of the flour was gotten to be 3.80%. The flour was then divided into four samples of 300 g each. The first portion served (3.80% moisture) served as control while the three remaining samples were reconditioned to moisture contents of 5.22, 7.78 and 9.04%, respectively and labeled A, B and C. The samples (A, B and C) were then packaged in high density polythene bag and stored under same storage conditions (29°C, 75%RH) with the control which was left unpacked. Proximate composition and microbial analysis of the flour were carried out at two weeks interval for a period of six weeks. Results obtained showed that moisture content had significant effect on the proximate composition, the moisture content for the control increased from 3.80±0.02 to 4.20±0.38; while A, B and C decreased from 5.22±0.49 to 4.65±0.12, 7.78±0.19 to 4.32±0.29 and 9.04±0.61 to 6.42±0.11, respectively after 6 weeks of storage; also, the total viable counts for both aerobic bacteria and fungi of the stored bambara nut flour was significant (p<0.05).

Keywords: Bambara nut flour, high density polythene, microbial count, moisture content, proximate composition, quality

Introduction

Bambara nut (*Vigna subterranea*) is an indigenous crop that is predominantly cultivated in semi-arid African regions by small scale farmers. It belongs to the family Fabaceae and sub family of the *papilionaceae* legumes (Atikuet al., 2004). It is cultivated from Senegal to Kenya and from the Sahara to South Africa and Madagascar for centuries but popular in Sub-Saharan Africa (Atikuet al., 2004). It is economically vital because it is an inexpensive source of high quality protein and is highly valued among the Northern and Eastern states of Nigeria (Oloyede et al., 2010). In Nigeria, bambara nuts are widely produced in Borno, Benue, Plateau and Taraba. It is probably the most drought-resistant of the grain legumes and may be found growing successfully where annual rainfall is below 500 mm and optimum between 900–1000 mm per year (Oloyede et al., 2010).

One of the major attributes of bambara nut is its high yield even in areas with notable dry season and poor soil and resistance or low susceptibility to pest and diseases which has led to the development of interest by researchers (Akande et al., 2009). Omoikhojo (2008) reported that it is the third most eaten legume in Africa after groundnut and cowpea. It originated in the Sahelian region of present day West Africa, from the bambara tribe near Timbuktu who now live mainly in Central Mali, hence its name bambara nut. It is also known as Juko beans (South Africa), NuyoCibemba (Republic of Zambia), Gurjirvoor Kwarrau (Hausa, Nigeria), Okpa (Igbo, Nigeria), Epawro (Yoruba, Nigeria) and Njimo beans (Zimbabwe) (Omoikhojo, 2008).

Bambara nut is chiefly cultivated for its edible and nutritional seeds. The seeds are contained in pods that are approximately 1.5 cm long and may be wrinkled and slightly oval or round; each pod contains one or two seeds. The colours of the seed usually vary from dark-brown, red, white, black, cream or a combination of these colours. The seeds contain sufficient amounts of protein, carbohydrate and fats; hence it is seen as complete food (Omoikhojo, 2008).

Considering its nutritional composition, bambara nut is an unusual example of a complete food and it has been alleged that people can live on it alone. The seed whether ripe or immature contains about 20% protein, 60% carbohydrates and 7% oil (National Research Council, 2006). The legumes are an important source of dietary protein, particularly when intake from animal sources is low or not available (Graham and Vance, 2003).

Okpuzo et al. (2010) reported that when compared to other food legumes, bambara nut is rich in iron and the protein contains high lysine and methionine and can thus complement cereals. Over 100,000 metric tonnes of bambara nut is produced in Nigeria but it remains an underutilized legume because of the following constraints: hardness to cook, strong beany flavour, presence of anti-nutrients and dehulling difficulty (Enwere and Hung, 1996; Alozie et al., 2009). The dry seeds can be ground into flour and blended into many customary dishes, or used to make moi-moi. In eastern Nigeria, it is used to prepare a local delicacy called okpa. According to Eltayebet et al. (2011), adding bambara nut flour to wheat flour biscuits improved both the texture and flavor. Flour and some other materials used in producing food products need to be properly stored and well packaged before utilization to ensure its safety, quality and storage stability. To recognize the adequate potential of bambara nut flour in food processing, it is therefore vital to take into cognizance the effect of moisture content on some quality parameters of stored Bambara nut flour.

Materials and Methods

Sample preparation

Bambara seeds were purchased from Kure Ultra-Modern Market, Minna, Niger State, Nigeria. The bambara seed samples were thoroughly cleaned by picking out dirt and other unwanted particles and sorted manually to remove extraneous matters like dirt, immature seeds and small branches. They were milled and sieved with 300 um sieve into very fine flour as described by Abdel Rahmanet al. (2011). The initial moisture content of the initial sample was determined using the method described by Onwuka (2005). The initial moisture content of the initial sample (1200 g) was determined to be 3.8%. The sample was divided into four parts (300 g) and was labeled Control, A, B and C. The first three parts were reconditioned, using the method described by Akinoso (2006) and...
to moisture contents of 5.22% 7.78 and 9.04% while the fourth part was left to serve as control. After reconditioning, the samples (which were in triplicates) were mixed with a sterilized stirrer packaged in a high density polythene bags and put in the refrigerator at 4°C for about 3 days.

**Storage stability studies**

The processed Bambara nut flour samples were packaged (300 g) and stored for six weeks at room temperature. Analysis was carried out at two weeks interval for the duration of storage. All analysis of microbial stability and proximate compositions of Bambara nut flour were determined in triplicates.

**Analytical methods**

**Determination of proximate composition**

Moisture content was obtained using the standard oven method as described by AOAC (2012). The initial moisture content of the Bambara nut flour was determined as 3.8 %. The sample was then divided into four parts. The first part was to serve as the control while 300 g each of Bambara nut flour were reconditioned to desired moisture content levels of 5.22%, 7.78% and 9.04% labeled A, B and C, respectively using the method described by Akinoso (2006). The moisture content of the flour was increased by adding distilled water to the flour samples and calculated from equation 1.

\[ Q = \frac{A(b - a)}{100 - a} \]

A= Initial mass of the sample in grams (g)  
= Initial moisture content of the sample  
b = Desired moisture content of the sample  
Q = Mass of water to be added in kilograms (kg)

Crude protein, Ash, Fat, Crude fibre, Carbohydrate and dry matter of the Bambara nut flour were determined according to the methods described by Association of Analytical Chemists (AOAC, 2012).

**Microbial analysis**

Microbial analysis (total bacterial and fungal counts) was determined according to the methods described by Onwuka (2005).

**Statistical analysis**

The data were subjected to statistical analysis using SPSS software version 20.0. A one-way analysis of variance (ANOVA) was done to determine the differences. Significantly different means were separated using DMART. Significance was accepted at p<0.05.

**Results and Discussion**

**Changes in proximate composition**

The effect of moisture content on the proximate composition of bambara nut flour is presented in Table 1. Moisture content and storage duration had significant (p<0.05) effect on the proximate composition of Bambara nut flour. The moisture content of all the four samples increased during the first four weeks of storage and suddenly decreased in the sixth week of storage. This increase in moisture content could be as a result of variation of the storage temperature and relative humidity. The moisture content of the control increased from 3.80±0.02 to 4.20±0.58 while that of the other samples A, B and C decreased from 5.22±0.49 to 4.65±0.12, 7.78±0.19 to 4.32±0.29 and 9.04±0.61 to 6.42±0.11, respectively after 6 weeks of storage. The increase in moisture content observed in the control sample could be as a result of increase in the relative humidity of the environment during the storage period considering the fact the control was left unpacked (open).

Amadi and Adebola (2008) reported that dry produce will exchange moisture with the atmosphere of storage until equilibrium is reached. The moisture content range of this study falls below the recommended moisture range (12.0–13.0) for flour stability. Lower moisture content in flour is a good indication of microbial stability and may also contribute to reducing the tendency of staling in baked food products (Ogiehor and Ikenebomeh, 2006).

There was significant (p<0.05) but inconsistent increase in ash contents of samples B and C, and a decrease in the control and sample A during the storage period. The increase in ash content was in the order, sample B (4.01±0.08) < sample C (4.30±0.46) while the decrease was in the order: control (3.05±0.16) < sample A (3.72±0.03). The effect of moisture content was not significant in ash content for all the samples. According to Mbataet al. (2007), addition of 30% Bambara groundnut improves the mineral and essential amino acid contents of maize ‘ogó’. There were little differences in protein and fat contents as they consistently increased during the storage period. The percentage protein difference was between 24.34±0.35-23.72±2.72 for the control sample, 25.70±0.71-25.75±0.25 for sample A, 26.59±0.72-28.43±0.73 for sample B and 26.30±0.25-27.5±1.19 for sample C. The results observed in this study could be as a result of effect of moisture and microbes in the package as reported by Uchechukwu-Aguwa (2015).

Fat contents also increased as the storage period increased. Changes in moisture content and storage duration had notable effect on both fat and protein contents of flours. There was significant amount of carbohydrate in all four samples across storage. The decrease in carbohydrate content could be as a result of the variation in the other parameters like ash, protein and fat during storage.

The control had highest carbohydrate content (62.78±2.83) at 29°C, 75%RH storage condition. The lowest carbohydrate content of the flour was observed in sample C (52.82±0.95). These values fall within the range of the average value of 63% reported for carbohydrate content of Bambara (Mbata et al., 2007).

**Changes in microbial quality**

Microbial count of bambara nut flour samples stored at ambient conditions (29°C, 75%RH) at different moisture content levels were influenced by the moisture content of the flour (Table 1). There was a progressive increase during storage in the total plate count for aerobic mesophilic bacteria and fungi in all the four samples of Bambara nut flour. This could be due to increase in moisture content during storage. The total viable bacteria counts increased from 1 to 1.4×10³; 2.0×10⁴; 3.0×10⁵ cfu/g and 3.0×10⁶ while the total fungal count increased from 1 to 3.7×10³; 3.8×10⁵; 4.2×10⁶ cfu/g and 4.0×10⁷ in the control, samples A, B, and C, respectively. The steady and gradual increase noted in the total viable bacterial and fungal counts in all the samples stored at different initial moisture content suggest a favourable micro environmental conditions and nutrient availability (Adejumọ and Raji, 2012).

The results of the study shows that sample B (Bambara nut flour with an initial moisture content of 7.78±0.19%) exhibited higher microbial stability with lower bacterial count of 3.0×10³ cfu/g compared to samples A and C and the lowest fungal count of 2.0×10⁶ cfu/g at the end of the six weeks storage period.
Evaluation of the Effect of Moisture Content on Stored Bambara Nut Flour

Table 1: Proximate composition of Bambara nut flour at different moisture levels under the same storage conditions

<table>
<thead>
<tr>
<th>Samples</th>
<th>Duration (wks)</th>
<th>Moisture content (%)</th>
<th>Ash (%)</th>
<th>Fat (%)</th>
<th>Crude Fibre (%)</th>
<th>Crude Protein (%)</th>
<th>Carbohydrate</th>
<th>Dry matter</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>0</td>
<td>3.60±0.02</td>
<td>4.78±0.06</td>
<td>4.99±0.11</td>
<td>3.06±0.13</td>
<td>24.34±0.35</td>
<td>50.64±0.26</td>
<td>96.2±2.2</td>
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<tr>
<td></td>
<td>2</td>
<td>4.42±0.10</td>
<td>3.06±0.21</td>
<td>3.33±0.58</td>
<td>1.91±0.05</td>
<td>24.00±0.58</td>
<td>62.31±1.37</td>
<td>95.5±1.1</td>
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<tr>
<td></td>
<td>4</td>
<td>6.10±0.19</td>
<td>3.73±0.29</td>
<td>4.17±0.29</td>
<td>1.62±0.34</td>
<td>21.95±1.41</td>
<td>62.43±0.76</td>
<td>93.9±0.20</td>
</tr>
<tr>
<td>Sample A</td>
<td>0</td>
<td>4.20±0.58</td>
<td>3.05±0.16</td>
<td>3.91±0.16</td>
<td>1.45±0.19</td>
<td>23.72±2.72</td>
<td>62.78±2.83</td>
<td>95.8±0.58</td>
</tr>
<tr>
<td></td>
<td>2</td>
<td>5.22±0.49</td>
<td>3.75±0.18</td>
<td>7.3±0.25</td>
<td>2.80±0.19</td>
<td>25.70±0.71</td>
<td>55.24±0.75</td>
<td>94.78±0.49</td>
</tr>
<tr>
<td>Sample B</td>
<td>0</td>
<td>7.78±0.19</td>
<td>3.57±0.09</td>
<td>7.49±0.03</td>
<td>3.18±0.23</td>
<td>26.59±0.72</td>
<td>51.40±0.46</td>
<td>92.2±0.19</td>
</tr>
<tr>
<td></td>
<td>2</td>
<td>4.88±0.43</td>
<td>3.80±0.36</td>
<td>5.16±0.33</td>
<td>3.11±0.14</td>
<td>26.91±0.84</td>
<td>56.14±0.69</td>
<td>95.12±0.13</td>
</tr>
<tr>
<td></td>
<td>4</td>
<td>5.02±0.25</td>
<td>3.80±0.23</td>
<td>4.95±0.24</td>
<td>1.77±0.09</td>
<td>26.64±0.84</td>
<td>57.4±0.90</td>
<td>94.98±0.25</td>
</tr>
<tr>
<td>Sample C</td>
<td>0</td>
<td>4.32±0.29</td>
<td>4.01±0.08</td>
<td>4.83±0.17</td>
<td>2.01±0.07</td>
<td>28.43±0.73</td>
<td>56.4±0.90</td>
<td>95.68±0.29</td>
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<td>2</td>
<td>4.70±0.54</td>
<td>3.07±0.15</td>
<td>7.59±0.25</td>
<td>3.14±0.15</td>
<td>26.30±0.25</td>
<td>50.9±0.14</td>
<td>90.96±0.61</td>
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<td>4</td>
<td>7.19±0.25</td>
<td>3.39±0.20</td>
<td>6.61±0.35</td>
<td>3.18±0.21</td>
<td>26.82±1.13</td>
<td>52.82±0.95</td>
<td>92.81±0.25</td>
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<td></td>
<td>6</td>
<td>4.37±0.06</td>
<td>2.88±0.50</td>
<td>6.25±0.23</td>
<td>1.45±0.18</td>
<td>26.93±0.99</td>
<td>58.12±0.67</td>
<td>95.63±0.06</td>
</tr>
<tr>
<td></td>
<td>8</td>
<td>6.42±0.11</td>
<td>4.30±0.46</td>
<td>4.90±0.13</td>
<td>1.64±0.12</td>
<td>27.57±1.19</td>
<td>55.17±1.22</td>
<td>93.24±1.77</td>
</tr>
</tbody>
</table>

The values are given as means of triplicate determinations ±standard deviation. Similar superscripts in columns are not significantly different (p=0.05)

The results for the microbial analysis of triplicate determinations are represented in Figs. 1 and 2.

Fig. 1: The effect of moisture content on the total bacterial count of bambara nut flour

Fig. 2: The effect of moisture content on the total fungal count of bambara nut flour

Conclusion

A vital factor to be considered in extending the shelf life stability of Bambara nut flour is moisture content. Moisture content had significant effect on the carbohydrate, fat and protein contents of bambara flour on storage. Moisture content affected the microbial quality of the flour during storage. Flours with low moisture content had low microbial growth and therefore longer shelf-life and better quality. Sample B with an initial moisture content of 7.78±0.19% provided the lowest moisture content of 4.32±0.29% and highest stability of microbial quality in bambara nut flour during the six weeks of storage period. Moisture content of 8% is recommended for storing bambara nut flour for a long period in order to maintain quality, storage and microbial stability which are vital for use of the flour for food formulations.

References

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