EFFECTS OF FERMENTATION WITH R. OLIGOSPORUS AND R. STOLONIFER ON PROXIMATE AND PASTING PROPERTIES OF DEHULLED AND UNDEHULLED VELVET BEANS (MUCUNA UTILIS) FLOUR

Balogun, OlA*; Olatidoye, OPA; and Otunola, ETB

ADepartment of Food Technology, Yaba College of Technology, Yaba, Nigeria.
BDepartment of Food Science and Technology, Ladoke Akintola University Ogbomosho, Nigeria.

ABSTRACT

The effects of fermentation on selected properties of velvet beans (Mucuna utilis) were studied. The bean seeds were subjected to solid state fermentation using Rhizopus oligosporus and Rhizopus stolonifer, both singly and in combination respectively incubated at 30°C for 72 hours. Samples were analyzed for proximate and pasting properties using established procedures at twelve hourly intervals during fermentation. After fermentation, samples were dried in hot air oven at 55°C for 24 hours, milled and sieved appropriately. Results showed that the moisture contents of the fermented velvet bean flour reduced significantly as fermentation progressed from 6.02% in the undehulled unfermented sample to 5.02% in the undehulled sample fermented with the mixed culture; and from 5.92% in the unfermented dehulled sample to 5.08% in the dehulled sample fermented with R. oligosporus. The ash content increased significantly (P<0.05) from 3.00% in the unfermented, undehulled sample to 4.30% in the undehulled sample fermented with R. oligosporus, and from 2.90% in unfermented dehulled sample to 2.30% in both dehulled sample fermented with R. oligosporus and dehulled sample fermented with the mixed culture. The carbohydrate content generally decreased across all the samples as the period of fermentation increased (P<0.05). Peak viscosity decreased throughout all the samples, in both the dehulled and undehulled samples. The value
range in the undehulled samples is from 120.25 to -2.93 RVU while the pasting temperatures increased from 55.95°C in the unfermented sample to 95.50°C. Also in the dehulled samples, the range of values for peak viscosity is from 11.35 to -2.02 RVU, and however a decrease in the values of pasting temperatures for these set of samples observed from 98.80°C to 84.45°C. All other pasting characteristics values, which include final viscosity, set back and peak time decreased significantly at P < 0.05, except for the peak time which was observed to have increased in the undehulled sample. The result of this study suggests that the fermentation process could be a viable option in the detoxification of velvet beans and also effective in improving its nutritional status. This study reveals that the nutritional profile of samples of M. utilis can also be explored as an alternate protein source to alleviate protein-energy-malnutrition among economically weaker sections of peoples in developing countries.

**Keywords:** Fermentation, proximate, physicochemical properties, velvet bean, Rhizopus species
1. INTRODUCTION

The dearth in food supply especially of protein is of such magnitude the developing nations have to depend mostly on cereals, grains, starch roots and tubers for energy and protein need (Auret and Behar Syndrome, 1953). The net effect of this protein deficit in the developing countries is manifested in the prevalence of various forms of Protein Calorie Malnutrition (PCM) diseases such as kwashiorkor, marasmus and mental deficiencies (Bressssani, 1975). In view of prevalent food shortage, attention is currently being focused on the exploitation of lesser known and non-traditional plant resources (Becker, 1986). This has necessitated exploration alternate sources of protein to bridge the gap for protein requirement of the various section of vegetarian population. In this context, alternate sources like untraditional legumes (under exploited/tribal pulses) assume significance. The average Nigerian does not consume enough protein of animal origin, and animal protein is more efficient than plant protein in providing the amino acids necessary for tissue development, repair and function (FAO, 1994). There is, therefore, a continual need to focus on the exploitation of lesser-known or non-traditional plant resources that are not subject to competition between man and livestock. *Mucuna utilis* (velvet bean), a tropical legume, is little known and has a low human preference for food, but has a high potential as an energy/protein source in livestock feed (Emenalom and Nwachukwu 2006). It is comparable to soybean in terms of amino acid and mineral profile (Siddhuraju et al., 1996; Iyayi and Taiwo 2003). However, the use of velvet beans as a source of protein for monogastrics is limited by the presence of antinutritional factors like trypsin inhibitors, haemagglutinins, phytic acids, hydrocyanic acid and tannins (Emiola et al., 2003; Siddhuraju et al., 1996).

Agro-processing operations which describe the transformation of agricultural produce into different physical or chemical states have identified several unit operations (including fermentation) to bring about such changes. This is sequel to the fact that fermentation is a highly appropriate technique for use in developing countries and remote areas where access to sophisticated equipment is limited (FAO, 1998). Although, fermentation process has several inherent benefits in food processing operations (Campbell-Platt, 1980), the operations have the potential to alter the functional properties of starch embedded in velvet bean. It is also known that the functional properties of starches are increasingly exploited in several facets of human endeavor, particularly in the food industry where they replace other plants of microbial polymers, which are more expensive. Such starches according to Charles and Guy (1999a) are modified in a number of ways. The utilization of starch is based on its physico-chemical properties such as its ability to swell which is required in bread making, emulsifying power which is appreciated in salad dressing and moisture binding capacity required in baking powder amongst several uses (Agboola et al., 1994). Other properties of starch that usually affect the application in food industry include gelatinization, pasting, solubility, swelling, clarity, opacity.
and viscosity (Sabiramo and Del Rosario, 1986). This study therefore aims to evaluate the effect of fermentation with *Rhizopus* species on proximate and pasting properties of dehulled and undehulled velvet beans (*Mucuna utilis*) flour.

**2. MATERIALS AND METHODS**

**2.1. Materials**: The velvet bean (*Mucuna pruriens var. utilis*) used for this study was obtained from International Institute for Tropical Agriculture, IITA, Ibadan, Nigeria. The inoculum (R. oligosporus and R. stolonifer) were obtained from the Indonesian Embassy, Lagos, Nigeria while the chemicals used were of the analytical grade obtained from May and Baker Company, England and Sigma Chemical Company Limited, UK.

**2.2. Preparation of subculture for velvet bean fermentation**: The subcultures of *Rhizopus oligosporus* and *Rhizopus stolonifer* were prepared by the procedure of Siddhuraju and Becker (2001). 50ml of distilled water was added to 50g rice in a beaker. This was covered with sterile muslin cloth firmly tied with a twine. The beaker and its contents were then sterilized, at 115 psi for 15 minutes and at a temperature of 120°C, cooled and inoculated, separately, with each inoculum of *Rhizopus oligosporus* and *Rhizopus Stolonifer* before being incubated for 4 days at 30°C. It was then dried in a sterile oven at 40°C for 48 hours, pulverized in a clean sterile dry blender for 2 minutes until uniform dark grey granules were obtained. This was then stored in sterile sealed polythene bags in a sterile jar.

**2.3. Preparation of Fermented Velvet Bean**: This was done according to the method of de-Reu *et al.*, (1994). 500g of velvet bean was cleaned and washed with tap water. It was then steeped in water for 12 hours, drained, put in aluminum pot with some water and brought to boil. It was then drained and de-hulled. Water at a ratio of 1:4 w/v was added to the beans and allowed to steep for 24 hours. The steeped beans were then boiled in the steep water, drained and then spread out to dry a little at room temperature. The de-hulled, cooked beans were then poured the perforated polythene bag and inoculums sprinkled and thoroughly mixed. The container was then tightly sealed. They were then incubator at about 32°C for a periods ranging between 0 and 72hr. The unfermented velvet bean sample (0 hour fermentation) was then used as the control. At regular intervals of 12 hours samples were taken out for appropriate analysis, except between 24 and 36 hours when samples were taken at 6 hours intervals.

**2.4. Preparation of Fermented Velvet Bean Flour**: At the end of the fermentation period, samples from each fermented beans were taken and blanched for 20 minutes, and then sliced into smaller units. The slices were then drained and dried in an oven at a temperature of around 55°C for 24 hours, cooled and then milled to give fermented velvet bean flour.

The flour was packed in polythene bags, sealed and kept in the freezer until required for analysis.

3. RESULTS AND DISCUSSIONS

3.1. Proximate Composition of fermented and unfermented velvet bean Flour

Data obtained on the proximate composition of velvet bean flour during fermentation with *Rhizopus oligosporus* and *Rhizopus stolonifer* are as illustrated in Tables 1 and 2. The value of protein content in the undehulled samples increased from 25.65% in the unfermented sample to 29.39% in the sample fermented with *R. stolonifer*. Also from 25.05% in the unfermented dehulled sample to the highest value of 36.0% in the sample fermented with mixed culture (*Rhizopus oligosporus* and *Rhizopus stolonifer*) after 72 hours of fermentation. These results were in agreement with the observations reported by Vadivel and Janardhanan (2005) that marked increases in protein content was noticed as the period of fermentation increased. It also shows the significant secondary effect of dehulling on fermentation of legumes. The fat content decreased in all the samples from 14.01% in the unfermented, undehulled sample and from 13.31% to 12.12% in the undehulled sample fermented with *R. oligosporus* and 10.03% in the dehulled sample fermented with mixed culture. This result is consistent with the observation of Otunola and Olanipekun, 2004. The decrease in the fat content could have been due to the action of lipase produced by the cultures *R. oligosporus* and *R. stolonifer* as fermentation progressed (Pugalenthi, 2005). However, an increase in fat content was observed in dehulled sample fermented with *R. stolonifer* (15.03%) and 15.05% in undehulled sample fermented with the mixed culture. The carbohydrate content generally decreased across all the samples as the period of fermentation increased (P<0.05). This agrees with observations of Otunola and Olanipekun, 2004. The reduction in carbohydrate content of fermented velvet bean flour could be attributed to the possible bio-conversion of the substrate enzymes from organisms responsible for the fermentation, that is, *R. oligosporus* and *R. stolonifer* into other substances, especially proteins and fats, in addition to the portion used for carbon and energy source of the organisms. The ash content increased significantly (P<0.05) from 3.00% in the unfermented, undehulled sample to 4.30% in the undehulled sample fermented with *R. oligosporus*, and from 2.90% in unfermented dehulled sample to 2.30% in both dehulled sample fermented with *R. oligosporus* and dehulled sample fermented with the mixed culture. There was a very remarkable difference between the values of ash contents of the of the dehulled and undehulled samples, the undehulled values had significantly lower values because of the removal of the seed coats, which are responsible for most of the ash contents of legumes. These results are consistent with values previously reported by Akinjayeju and Enude, 2002. The general low moisture contents of the fermented velvet bean flour further reduced significantly as fermentation progressed from 6.02% in the undehulled
unfermented sample to 5.02% in the undeveloped sample fermented with the mixed culture; and from 5.92% in the unfermented dehulled sample to 5.08% in the dehulled sample fermented with *R. oligosporus*.
Table 1: Proximate composition of undehulled samples fermented with *R. oligosporus* and *R. stolonifer*

<table>
<thead>
<tr>
<th>Sample</th>
<th>Protein Content (%)</th>
<th>Fat Content (%)</th>
<th>Moisture Content (%)</th>
<th>Ash Content (%)</th>
<th>Crude Fibre Content (%)</th>
<th>Carbohydrate Content (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>G</td>
<td>25.65c</td>
<td>14.01b</td>
<td>6.02a</td>
<td>3.00d</td>
<td>7.33c,d</td>
<td>44.09a</td>
</tr>
<tr>
<td>A</td>
<td>27.00b</td>
<td>12.12d</td>
<td>5.51b</td>
<td>4.30a</td>
<td>7.32c</td>
<td>43.75b</td>
</tr>
<tr>
<td>C</td>
<td>28.63a</td>
<td>15.05a</td>
<td>5.02c</td>
<td>3.70b</td>
<td>9.05a</td>
<td>38.37d</td>
</tr>
<tr>
<td>E</td>
<td>29.39a</td>
<td>13.33c</td>
<td>5.04c</td>
<td>3.30c</td>
<td>7.45b</td>
<td>41.49c</td>
</tr>
</tbody>
</table>

*Values with the same subscript in the same column are not significantly different (P<0.05)*

A = Undehulled sample fermented with *R. oligosporus*, C = Undehulled sample fermented with the *R. oligosporus* + *R. stolonifer*, E = Undehulled sample fermented with *R. stolonifer*

G = Unfermented undehulled sample (Control)

Table 2: Proximate composition of dehulled samples fermented with *R. oligosporus* and *R. stolonifer*

<table>
<thead>
<tr>
<th>Sample</th>
<th>Protein Content (%)</th>
<th>Fat Content (%)</th>
<th>Moisture Content (%)</th>
<th>Ash Content (%)</th>
<th>Crude Fibre Content (%)</th>
<th>Carbohydrate Content (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>H</td>
<td>25.05d</td>
<td>13.31b</td>
<td>5.92a</td>
<td>2.90b</td>
<td>7.56c</td>
<td>45.26a</td>
</tr>
<tr>
<td>B</td>
<td>35.20b</td>
<td>13.30b</td>
<td>5.08c</td>
<td>2.30a</td>
<td>7.22d</td>
<td>36.91c</td>
</tr>
<tr>
<td>D</td>
<td>36.00a</td>
<td>10.03c</td>
<td>5.56b</td>
<td>2.30a</td>
<td>8.90a</td>
<td>37.98b</td>
</tr>
<tr>
<td>F</td>
<td>34.54c</td>
<td>15.03a</td>
<td>5.41b</td>
<td>2.10b</td>
<td>7.40c</td>
<td>36.52d</td>
</tr>
</tbody>
</table>

B = Dehulled sample fermented with *R. oligosporus*, D = Dehulled sample fermented with *R. oligosporus* + *R. stolonifer*, F = Dehulled sample fermented with *R. stolonifer*, H = Unfermented dehulled sample (Control)

### 3.2. Pasting Characteristics

In the course of fermentation, it was observed that the peak viscosity decreased throughout all the samples, in both the dehulled and undehulled parts as shown in Tables 3 and 4. The value range in the undehulled samples is from 120.25 to -2.93 RVU, while...
the pasting temperatures increased from 55.95°C in the unfermented sample to 95.50°C. Also in the dehulled samples, the range of values for peak viscosity is from 11.35 to 2.02 RVU, and however a decrease in the values of pasting temperatures for these set of samples observed, from 98.80°C to 84.45°C. All other pasting characteristics values, which include final viscosity, set back and peak time decreased significantly at P < 0.05, except for the peak time which was observed to have increased in the undeckled samples. The observation appeared similar to those of Otunola and Olanipekun, (2002). The peak viscosity on fermentation and it became more obvious in the undeckled samples. This could probably be due to high protein contents with increasing period of fermentation, which invariably influences the visco-elastic properties of the flour. Higher amylose content due to starch concentration pattern lowers the initial pasting temperature (Otunola and Olanipekun, 2002, Akinjayeju and Bisiriyu, 2004). In line with these values which appear comparable to those of starches extracted from some staples, there therefore a possibility that these flours from fermented velvet beans could adequately be used as substituent in some food products where the other crops are being used. The pasting characteristics data suggest that fermentation and dehulling have the tendency to reduce the cooking time of the fermented flours. This is due to the fact that peak viscosity was observed to have reduced significantly in the fermented flours. The reduction could have been due to the activities of amylase which break down starch into simpler sugars, releasing bound water and so reducing viscosity in accordance with the report of Otunola and Olanipekun, (2002). Set back value, an indication of tendency to retrograde on cooling, was observed to have decreased in the fermented samples when compared with the control. The reduction was even more pronounced in the dehulled samples, however an unexpected significant increase was observed in the undeckled sample fermented with R. stolonifer. This suggests that the tendency to retrograde is significantly reduced in the fermented samples, excepting for the undeckled sample fermented with R. stolonifer which is expected to retrograde significantly compared to other fermented samples. The reduced peak time in the fermented dehulled sample flours suggests early gelatinization, which can render starch more susceptible to breakdown because it undergoes a longer period of sheer (Otunola and Olanipekun, 2002). The opposite will be expected in the fermented undeckled sample flours in which there was a significant increase in peak time compared with the control.

Table 3: Pasting characteristics of Fermented and Unfermented Undeckled velvet bean Flours

Table 4: Pasting characteristics of Fermented and Unfermented dehulled velvet bean Flours

<table>
<thead>
<tr>
<th>Sample</th>
<th>Peak Viscosity</th>
<th>Final viscosity</th>
<th>Set back</th>
<th>Peak time</th>
<th>Pasting temperature</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>-2.93c</td>
<td>-2.52c</td>
<td>0.76c</td>
<td>6.30a,b</td>
<td>95.50 a</td>
</tr>
<tr>
<td>C</td>
<td>-2.52c</td>
<td>-2.02c</td>
<td>0.80c</td>
<td>5.70b</td>
<td>95.40 a</td>
</tr>
<tr>
<td>E</td>
<td>25.25b</td>
<td>5.00b</td>
<td>4.57a</td>
<td>5.27c</td>
<td>51.05c</td>
</tr>
<tr>
<td>G</td>
<td>120.25a</td>
<td>12.00a</td>
<td>1.50b</td>
<td>2.00d</td>
<td>55.95b</td>
</tr>
</tbody>
</table>

A = Undehulled sample fermented with R. oligosporus, C = Undehulled sample fermented with the R. oligosporus + R. stolonifer, E = Undehulled sample fermented with R. stolonifer, G = Unfermented undehulled sample (Control).

4. CONCLUSION:

The observation made in present study protein, most of the essential amino acids, fatty acid such as linoleic, palmitic and oleic acids and some minerals when compared to some other oil seeds and nuts. This study reveals that the nutritional profile of samples of M. utilis can also be explored as an alternate protein source to alleviate protein-energy-malnutrition among economically weaker sections of peoples in developing countries.
REFERENCES


