MICROBIOLOGICAL, TEXTURAL AND SENSORY CHANGES DURING THE FERMENTATION OF AFRICAN LOCUST BEAN (Parkia biglobosa) TO PRODUCE SONRU, WEST AFRICAN TRADITIONAL CONDIMENT

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ABSTRACT

African locust bean (Parkia biglobosa) was traditionally fermented in basket with jute bags to produce Sonru, a food condiment from Benin. Due to hygienic problems, a wooden box was developed to replace basket with jute bags to process African locust bean into Sonru. Changes in microbial, textural and sensory characteristics of the fermented cotyledons from wooden box compared with that from basket with jute bags were investigated at different fermentation times (0, 6, 12, 18 and 24 hours). Bacillus species were found as the main microorganism involved in the fermentation of Sonru. The use of the box allowed increasing Bacillus growth during the fermentation and improving texture (softness) of the product. Penetration forces were significantly lower in samples fermented from the wooden box than that from the traditional method (basket with jute bags). Concerning sensory quality attributes, samples fermented from the wooden box at 12 hours showed higher scores of softness and aroma with when compared to traditional Sonru. The wooden box was suitable to improve the fermentation process of African locust bean to produce Sonru.

Keywords: Fermentation, Sonru, locust bean, wooden box, sensory, softness

INTRODUCTION

African people have depended on non timber forest products (NTFPs) for food security. There is diversity of the NTFPs that are used for food and medicine (Addis et al., 2013; FAO 1996). NTFPs are rich in vitamins (mainly A, B, and C), minerals, fibers, carbohydrates and proteins (Chadare et al., 2017). They are affordable for both urban and rural population in Africa countries and offer high nutritional quality for many families (Guil et al., 1997; Ogle and
Grivetti 1985). African locust bean tree (*Parkia biglobosa*) is one of the most NTFPs which seeds are used as protein source condiment after fermentation and consumed by various socio-cultural groups of West African countries with different names Afitin, Sonru, Iru in Benin (Azokpota *et al.*, 2008; Azokpota *et al.*, 2006; Hongbété 2001), Iru, Dawadaw in Nigeria (Ajayin *et al.*, 2015; Daramola *et al.*, 2009), Soumbala in Burkina Faso (Diawara *et al.*, 1998; Ouoba *et al.*, 2003) and Netetu in Senegal (Ndir *et al.*, 2000). This African fermented flavour and taste condiment is similar to traditional fermented soya product, Natto in Japan and Thua-nao in Thailand (Tsai *et al.*, 2007).

In the north Benin, Sonru process involves a fermentation during 48h where cooked African locust bean cotyledons with traditional additive “Yanyanku” obtained from malvacene dried beans (*Hibiscus sabdariffa*), are spread on jute bags in basket or in fermentation box (Hongbété *et al.*, 2017b). After fermentation, the cover is removed and the container is left for cooling at ambient temperature for about 6 h. Traditionally, the processing technology is mainly produced as home based practice where the know-how has been passed from one generation to another (Hongbété 2001). In the North Benin, Sonru has increasingly gained interest in food technologies due to its nutritional properties and potential use as food flavouring agent.

However, basic information related to textural and sensory changes during Sonru fermentation, has not been reported. The objective of this study was to investigate the changes in African locust bean with additive during fermentation to produce Sonru in Fermentation box or in basket.

**MATERIALS AND METHODS**

**Sonru Production**

African locust beans were purchased in local market at Parakou (Benin) and were used to produce Sonru according to the traditional process as described the Fig.1. Dried African locust beans were first boiled for about 10 hours. They were then cooled overnight in the boiled water. After removing the testa by fulling, cotyledons were sorted, washed and boiled for about 2 hours. The cooked cotyledons were mixed with yanyanku before dividing into two batches. The first batch was fermented in wooden box (modern method) (Hongbété *et al.*, 2017a). The second batch was spread on basket trays, wrapped with jute sacks and clothes to maintain a hot atmosphere (traditional method). Wrapped cotyledons were fermented for about 18 hours. Then, the fermented cotyledons were left to cool at room temperature (25 ± 3 °C) for about 6 hours. Duplicate experiments were performed. The fermented cotyledons were sampled at 0, 6, 12, 18 and 24 hours for microbiological, physicochemical, textural and sensory analyses. The temperature of the fermented product was measured with thermometer (76 mm immersion, Brannan, UK) at each sampling time, as a quantitative variable.
**Microbiological analysis**

Ten (10) g of each fermented cotyledons sample collected at different fermentation times (0, 6, 12, 18 and 24 hours) were introduced aseptically in a sterile stomacher bag and 90 mL sterile peptone-bacteriological salt solution was added. The mixture was homogenised for two min, using a stomacher (Lab Blender, 400 Circulator Seward, England). One ml of the suspension was
serially used for microbial counts according to ISO norms. Total viable counts (TVC) were enumerated using Plate Count Agar (PCA, Oxoid CM0325, Basingstoke, Hampshire, England), *Staphylococci* were grown on Baird-Parker (BP, Oxoid, CM275) supplemented with 5% steril egg yolk-tellurite emulsion (Oxoid, SR 54). *Bacillus* spp. were enumerated using Dextrose Tryptone Agar (DTA, Oxoid, CM75) supplemented with 0.2% soluble starch (Merck, D-6100). PCA plates were incubated at 30 °C for 72 hours (ISO-4833 2003) while Baird Parker (ISO-6888 1999) and Dextrose Tryptone Agar plates were incubated at 37 °C for 48 hours.

**Chemical analysis**

The pH was immediately measured on the wet samples according to Nout et al. (Nout 2001). The dry matter content was determined after oven drying the samples at 105°C using AOAC method 27.005 (AOAC 1995). Temperature of cotyledons was measured using infrared thermometer (Brannan, High Temperature InfraRed Thermoter 50.0 to 550°C).

**Instrumental softness analysis**

A puncture test was performed to determine the softness of the fermented cotyledons according to the method described by Hongbété et al. (2001). Softness indices of fermented cotyledons were assessed with uniaxial compression and penetration tests which were performed on cotyledons samples using a Stevens texture analyzer (Stevens, LFRA, Harlow, U.K.) equipped with a cone penetrator moving at 0.2 mm.s⁻¹ to a final penetration depth of 2 mm. Twelve measurements were made on cotyledons at each fermented time.

**Sensory analysis**

Softness, aroma and appearance were considered to be the most important attributes to describe the sensory quality of Sonru. The panel was composed of 20 ordinary consumers of Sonru. They had been previously trained for softness and aroma attributes as described by Hongbété et al (2011). The appearance of the fermented African locust beans was assessed according to the method described by Charoenkul et al. (2006), slightly modified. The fermented cotyledons of African locust beans were visually scored using a 9-point hedonic box scale from 1: “dislike extremely” to 9: “like extremely”.

**Statistical analysis**

Data were analysed using Statistica (version 6, StatSoft France, 2004) and significance was accepted at probability p < 0.05 with one-way analysis of variance (ANOVA) by using least significant difference (Student–Newman–Keuls test).
RESULTS AND DISCUSSION

Total viable counts (TVC), *Bacillus* sp and *Staphylococcus* sp changes during the Sonru production in wooden box and basket were shown in Fig.2. a, b and c, respectively. During the 24 hours fermentation period, TVC, *Bacillus*, *Staphylococcus* counts increased statistically (p< 0.05) as the fermentation progressed. TVC increased from 3.7 Log cfu/g at the beginning of the fermentation to reach a maximum 8.4 Log cfu/g in Sonru from wooden box after 12 hours fermentation but this maximum was reached after 24 hours fermentation in that from basket with jute bags (Fig 2 a). *Bacillus* counts increased from 3.3 at the beginning of the fermentation to 7.7 and 6.6 Log cfu/g in Sonru from wooden box and basket with jute bags, respectively, until the end of fermentation. *Staphylococcus* counts increased from 2.35 to 4.05 and 8.48 Log cfu/g in Sonru from wooden box and in that from basket with jute bags until the end of fermentation, respectively. Similar results were reported by Azokpota et al. (2006) and Agbobatinkpo et al. (2012) who found an increase of microbial counts (TVC, *Bacillus* and *Staphylococcus*) during processing of African locust beans into Sonru. The Results recorded showed that TVC and *Bacillus* sp of samples from the wooden box increased rapidly in the first 12 hours of fermentation and the rate did not change much after that. *Bacillus* species were significantly (p< 0.05) higher in Sonru from wooden box than in that from basket after 12 hours of fermentation. *Bacillus* growth in samples from wooden box at 12 hours was similar to that in Sonru from basket at 24 hours. The higher value of *Bacillus* in samples from wooden box indicates that wooden box developed for Sonru production offers better fermentation conditions. Results showed the effectiveness of wooden box to create hygienic conditions for Sonru production. It means that the wooden box is a better fermentation material for African locust beans to reach a maximum microbial count within few times. *Bacillus* species are the main microorganisms involved in the fermentation process of African locust beans into fermented African locust beans condiment (Aderibigbe and Odunfa 1990; Azokpota et al., 2007; Daramola et al., 2009; Ouoba et al., 2004). High *Bacillus* level in African locust beans during fermentation has been reported by Ouoba et al. (2004); Oladunmoye (2007) and Azokpota et al. (2007). *Bacillus* sp has been reported to be responsible for the fermentation of some Asian soybeans products like Natto and meju (Kiers et al., 2000; Wang and Fung 1996). Leejeerajumnean et al (2000) reported that *Bacillus* sp are known to produce a diversity of metabolite that can contribute to flavours and aroma the end product.
Fig. 2: Microbiological changes (log cfu/g) during fermentation of African locust bean for Sonru production in wooden box (●) and basket with jute bags (○); (a) Total viable counts, (b) *Bacillus*, (c) *Staphylococcus*
Staphylococcus sp in fermented African locust beans could be originated from the environment contamination (fermentation material, processors clothes, seeds, yanyanku). The present results are in accordance with those of Azokpota et al. (2006) and Agbobatinkpo et al. (2012) who reported the presence of Staphylococcus sp in the 48 hours of fermentation period of African locust bean and from those of Omafuvbe et al. (2000) who reported the presence of Staphylococcus sp only at the beginning of the fermentation of soya bean to soy dawdawa. Staphylococcus spp are toxinogenic bacteria, responsible of the most common foods borne illness giving rise to nausea, vomiting, abdominal cramping, prostration and diarrhea (FAO 1994). An investigation is needed to have an idea about the contribution of Staphylococcus sp in the processing of African locust beans into Sonru. However Staphylococcus sp are known from other fermented foods such as Lanhouin a dried fermented salt fish product (Kindossi et al., 2016), fermented sausages (Benito et al., 2008; Coton et al., 2010) to contribute to flavour and aroma and appear to be relatively safe for consumption.

Table 1 shows the chemical changes of African locust bean at different fermentation times to produce Sonru in basket with jute bags and in wooden box. Results show significant increase in dry matter content during the fermentation. The dry matter content ranged from 36.2 to 38.3 % and 36.3 to 38.6 % dwb from the beginning of the fermentation to 24 hours for modern process and traditional process, respectively. But, no significant difference in dry matter content was observed between Sonru samples fermented from wooden box (modern method) and those fermented from basket (traditional method). The increase of dry matter could be clarified by the loss of water evaporation under the high temperature developed during fermentation and by the loss of residual water by draining through the perforated plate or basket during Sonru fermentation.
Table 1: Chemical changes of African locust beans at different fermentation times

<table>
<thead>
<tr>
<th>Methods</th>
<th>Fermentation equipment</th>
<th>Fermentation times (Hours)</th>
<th>Temperature °C</th>
<th>Dry matter (% dwb)</th>
<th>pH</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>0</td>
<td>34.0±0.5&lt;sup&gt;a&lt;/sup&gt;</td>
<td>36.2±0.2&lt;sup&gt;a&lt;/sup&gt;</td>
<td>5.6±0.2&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>Modern</td>
<td>Wooden box</td>
<td>6</td>
<td>38.8±0.5&lt;sup&gt;d&lt;/sup&gt;</td>
<td>36.7±0.2&lt;sup&gt;b&lt;/sup&gt;</td>
<td>6.7±0.1&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td></td>
<td></td>
<td>12</td>
<td>44.5±0.3&lt;sup&gt;e&lt;/sup&gt;</td>
<td>37.5±0.2&lt;sup&gt;c&lt;/sup&gt;</td>
<td>7.5±0.2&lt;sup&gt;d&lt;/sup&gt;</td>
</tr>
<tr>
<td></td>
<td></td>
<td>18</td>
<td>46.5±0.3&lt;sup&gt;f&lt;/sup&gt;</td>
<td>38.2±0.2&lt;sup&gt;d&lt;/sup&gt;</td>
<td>8.1±0.1&lt;sup&gt;e&lt;/sup&gt;</td>
</tr>
<tr>
<td></td>
<td></td>
<td>24</td>
<td>35.5±0.3&lt;sup&gt;b&lt;/sup&gt;</td>
<td>38.3±0.3&lt;sup&gt;d&lt;/sup&gt;</td>
<td>8.3±0.2&lt;sup&gt;e&lt;/sup&gt;</td>
</tr>
<tr>
<td>Tradition</td>
<td>Basket with jute bag</td>
<td>0</td>
<td>34.5±0.3&lt;sup&gt;b&lt;/sup&gt;</td>
<td>36.3±0.1&lt;sup&gt;a&lt;/sup&gt;</td>
<td>5.7±0.2&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td></td>
<td></td>
<td>6</td>
<td>37.5±0.5&lt;sup&gt;c&lt;/sup&gt;</td>
<td>36.8±0.2&lt;sup&gt;b&lt;/sup&gt;</td>
<td>6.3±0.2&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td></td>
<td></td>
<td>12</td>
<td>39.3±0.5&lt;sup&gt;d&lt;/sup&gt;</td>
<td>37.8±0.2&lt;sup&gt;c&lt;/sup&gt;</td>
<td>6.8±0.1&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td></td>
<td></td>
<td>18</td>
<td>44.0±0.6&lt;sup&gt;e&lt;/sup&gt;</td>
<td>38.4±0.2&lt;sup&gt;d&lt;/sup&gt;</td>
<td>7.5±0.2&lt;sup&gt;d&lt;/sup&gt;</td>
</tr>
<tr>
<td></td>
<td></td>
<td>24</td>
<td>33.1±0.8&lt;sup&gt;a&lt;/sup&gt;</td>
<td>38.6±0.3&lt;sup&gt;d&lt;/sup&gt;</td>
<td>7.6±0.2&lt;sup&gt;d&lt;/sup&gt;</td>
</tr>
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</table>

Significant increase in pH value was observed during the fermentation of African locust beans, with the highest value for samples fermented in wooden box. The pH value ranged from 5.6 to 8.3 and 5.6 to 7.6 from the beginning of the fermentation to 24 hours for samples fermented in wooden box and those fermented in basket, respectively. Samples from wooden box at 12 hours showed similar value of pH (7.5) to those from basket at 18 and 24 hours of fermentation. The increase of pH during African locust beans fermentation was agreed with previous findings (Azokpota et al., 2006; Hongbé té 2001; Omafuvbe et al., 2000). The increase of pH that occurs during fermentation of proteinous food is attributed to the abundant production of ammonia which is responsible for the unique aroma sometimes described as ammoniacal or pungent (Azokpota et al., 2006; Daramola et al., 2009; Diawara et al., 1998; Kiers et al., 2000). Temperature in African locust beans showed significant increase during fermentation from 0 to 18 hours followed by decrease at 24 hours. The increase in temperature varies according to fermentation equipment. The highest value was observed for samples from wooden box. These results are in accordance with those of Azokpota et al. (2006) and Agbobatinkpo et al. (2012).
The results of sensory quality attributes of Sonru samples from two types of fermentation materials evaluated by the panel for preference for soft texture, aroma, and overall acceptability were shown in Table 2. The sensory scores of softness varied between 6.4 and 8.4 for Sonru (traditional) from basket with jute bags while softness scores of samples from wooden box varied between 3.8 and 8.3. The samples of Sonru (modern) obtained from fermentation box at 15 hours and samples of Sonru (traditional) from basket with jute bags at 24 hours recorded significantly higher score for overall acceptance (8.2 and 8.5, respectively) with the highest aroma scores (7.3 and 7.4, respectively). Soft texture is one of important quality attributes of Sonru that influence its acceptability by consumers (Agbobatinkpo et al., 2012; Hongbéte 2001). The wooden box developed could be used to improve Sonru process by reducing the fermentation duration.

**Table 2: Sensory quality scores of Sonru from traditional and modern methods**

<table>
<thead>
<tr>
<th>Products</th>
<th>Fermentation equipment</th>
<th>Fermentation times (Hours)</th>
<th>softness</th>
<th>Aroma</th>
<th>Acceptance</th>
</tr>
</thead>
<tbody>
<tr>
<td>Modern</td>
<td>Wooden box</td>
<td>12</td>
<td>8.4 ± 0.3(^d)</td>
<td>7.3 ± 0.2(^c)</td>
<td>8.2 ± 0.2(^{cd})</td>
</tr>
<tr>
<td></td>
<td></td>
<td>18</td>
<td>7.2 ± 0.3(^c)</td>
<td>7.1 ± 0.2(^c)</td>
<td>8.4 ± 0.3(^d)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>24</td>
<td>6.4 ± 0.2(^b)</td>
<td>6.5 ± 0.2(^b)</td>
<td>6.7 ± 0.2(^b)</td>
</tr>
<tr>
<td>Traditional</td>
<td>Basket with jute bags</td>
<td>12</td>
<td>2.5 ± 0.1(^a)</td>
<td>4.3 ± 0.1(^a)</td>
<td>4.1 ± 0.1(^a)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>18</td>
<td>6.6 ± 0.3(^b)</td>
<td>7.2 ± 0.2(^c)</td>
<td>6.7 ± 0.3(^b)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>24</td>
<td>8.3 ± 0.2(^d)</td>
<td>7.4 ± 0.2(^c)</td>
<td>8.5 ± 0.2(^d)</td>
</tr>
</tbody>
</table>

The results of the instrumental texture analysis using a cone probe for penetration tests are shown in Table 3. There are significant effects of fermentation equipment and fermentation time on the softness of the African locust bean cotyledons. Significant decrease in maximum penetration forces of the African locust bean cotyledons was recorded during fermentation time (from 0 to 24 hours). The maximum penetration force varied from 1.9 to 0.33 N (in samples from basket with jute bags) and from 1.88 to 0.24 N (samples from wooden box) from 0 to 24 hours. Texture of samples from wooden box appeared softer than that of samples obtained from basket, jute bag and cloths. Decrease in maximum penetration force was also reported in African locust bean fermented with additives to produce iru and sonru (Agbobatinkpo et al. 2012).
Table 3: Changes in maximum penetration Force of African locust beans cotyledons at different fermentation times (N)

<table>
<thead>
<tr>
<th>Fermentation times (Hours)</th>
<th>Traditional method (Fermentation in basket with jute bags)</th>
<th>Modern method (Fermentation in wooden box)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>1.90 ± 0.02f</td>
<td>1.88 ± 0.03f</td>
</tr>
<tr>
<td>6</td>
<td>1.63 ± 0.03e</td>
<td>1.45 ± 0.02d</td>
</tr>
<tr>
<td>12</td>
<td>0.92 ± 0.02bc</td>
<td>0.66 ± 0.01c</td>
</tr>
<tr>
<td>18</td>
<td>0.68 ± 0.02c</td>
<td>0.35 ± 0.02b</td>
</tr>
<tr>
<td>24</td>
<td>0.33 ± 0.01b</td>
<td>0.24 ± 0.01a</td>
</tr>
</tbody>
</table>

CONCLUSION

The wooden box can be used for the fermentation of African locust beans to produce Sonru. More than the traditional fermentation equipment (basket and jute bags), the wooden box improves the fermentation conditions of the beans with a fast growth of Bacillus species. It allows improving a softening of fermented beans, according to low values of penetration force of samples fermented with the wooden box compared with traditional equipment. Samples from the wooden box showed a comparable sensory quality to the traditional Sonru. However, the optimum fermentation conditions of the wooden box remain a challenge to be overcome.

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