Effect of salt treatment on fungal spoilage, proximate and mineral compositions of fermented African locust bean (Parkia biglobosa) seeds during ambient storage

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INTRODUCTION

The prevailing population pressure in Nigeria as in other less-developed countries has resulted in an increasing demand for wild under-exploited nutritious plant products with aesthetic and organoleptic appeal in the daily diet (Enuigbua 2005). The common edible portions of most under-utilized plants are the seeds which in some cases are cooked or roasted and eaten directly as snack foods e.g. coconut nut and banana fruit groundnut while some are cooked and fermented for use as soup and sauce ingredients e.g. African oil bean, locust bean, castor bean and melon. There are various plant seeds that are fermented and used as food in some rural parts of Nigeria among which are ‘iru’ from African locust bean (Parkia biglobosa), ‘ogiri’ from castor bean (Ricinus communis), ‘ogosi’ from mesquite seed (Prosopis africana) and ‘Ugba’ from African oil bean (Pentaclethra macrophylla) (Eze et al. 2014).

African locust bean (Parkia biglobosa) is a popular food condiment in West African countries which is widely consumed for its high nutritional value to millions of Nigerians both in rural and urban areas (Faqbemi 1989). The fermented locust bean known as ‘iru’ and ‘dawadawa’ among the Yoruba and Hausa people of Nigeria respectively is basically used among these people as food seasoning and condiment (Nti et al. 2012). Apart from the flavouring attribute of the fermented locust bean, it also contributes significantly to the intake of protein, carbohydrate calcium, phosphate, iron and essential fatty acids particularly vitamin B riboflavin and vitamin A (Odebunmi et al. 2010). Nevertheless, its production in rural areas is discouraged by the way it rapidly spoils or deteriorates and consequently leads to loss of produce and farmers because storage of fermented locust beans without any form of preservation such as salting or refrigeration is about 48hours due to inherent high moisture content (>55%) and vulnerability to peroxidation due to high fat content (>37%) (Oni 1997). Hence, this study was undertaken to investigate effect of salt treatment on fungal spoilage, proximate and mineral compositions of fermented locust beans during storage. In a completely randomized experimental set up, 20g of fresh African locust beans were separately weighed and distributed into clean transparent small plastic containers. Two grams (2g) of common salt were then added and the containers covered before storage at 28+2°C and at75+5%RH. Unsalted samples served as control. By day 5 of storage, Rhizopus sp, Aspergillus niger and A. flavus were the fungi isolates found associated with the deterioration of the unsalted locust bean as evident by discolourations and mycelia appearance. Interestingly, salted samples did not show any sign of deterioration even by day 28 of storage. Results also showed significant decrease (except in moisture contents) in the proximate and mineral compositions of the unsalted samples when compared with the salted.

RESULTS

Fungi associated with the spoilage of African locust bean

Unsalted locust bean samples started showing signs of spoilage as evident by discolourations and presence of fungi mycelia by day 5 of storage. The various fungi identified to be associated with the spoilage of the unsalted fresh samples as reported in Table 1 were Aspergillus niger, A. flavus and Rhizopus sp. A. niger appeared colourless when young on MEA and later turned black with age due to the production of black conidia. Microscopically the conidia were globose to elliptical in shape and short chains of two or more spores on erect hyphae while A. flavus formed greenish colonies on MEA and microscopically, the conidia were globose and short chains of two or more spores on erect hyphae. The hyphae were branched and septate. Meanwhile, Rhizopus sp. appeared whitish, fluffy and cottony in texture on MEA. Microscopically, the sporangiole was erect and the sporangia were oval and angular in shape. However, salted samples did not show any sign of deterioration even by day 28 of storage.

Proximate compositions of unsalted and salted locust bean samples during storage

Table 2 showed the proximate compositions of unsalted and salted locust bean samples during storage. The values of carbohydrate (18.08%), protein (9.80%), crude fibre (16.50%), fat (26.50%) and ash (4.80%) contents in salted locust bean samples by day 7 of storage were significantly higher (P<0.05) compared with proximate of unsalted
samples having values of carbohydrate (12.14%), protein (3.22%), crude fibre (11.83%), fat (7.44%) and total ash (1.78%) contents. However, there was a significant reduction in the moisture content of the unsalted locust bean samples from 63.50 to 24.50% in the salted samples.

As storage duration progressed to day 28, proximate compositions of the salted bean samples had further increased although non significantly to 19.50% (carbohydrate), 10.44% (protein), 5.32% (total ash) and 17.20% (crude fibre). Nevertheless, the moisture and fat contents had decreased to 22.52 and 24.18%, respectively.

**Mineral compositions of unsalted and salted locust bean samples during storage**

Mineral compositions of both unsalted locust beans and salted samples during storage were reported in Table 3. Results showed that there was a significant increase (P<0.05) in the concentrations (mg/L) of sodium (1498.99), potassium (297.32) and calcium (178.39) of salted samples by day 7 in storage when compared with the concentrations (mg/L) of sodium (1112.21), potassium (137.64) and calcium (108.61) in unsalted samples. Similarly, concentrations (mg/L) of iron (1.99), zinc (8.94), magnesium (108.24) and copper (4.96) in the salted samples by day 7 in storage were significantly higher when compared with the concentrations of iron (0.18), zinc (6.00), magnesium (95.21) and copper (2.98) in unsalted samples. However, phosphorous, lead, nickel and cadmium were not detected in both unsalted and salted samples.

As storage duration progressed to day 28, concentrations of sodium, potassium and calcium had further increased significantly to 1500.91, 463.64 and 363.64 mg/L respectively (Table 3) while respective concentrations of iron, zinc, magnesium and copper also recorded further significant increase to 4.02, 15.10, 138.60 and 8.26 mg/L.

### DISCUSSION

In this study, Rhizopus sp, Aspergillus niger and A. flavus were the fungi isolates found associated with the deterioration of the unsalted (fresh) locust bean samples as evident by discolouration and mouldy smell of locust bean seeds during storage. The emergence of these isolates on the deteriorating bean samples suggest possible contamination by spores in the air. The light spores of these isolates might be present in the surroundings (mg/L) of the production unit where the samples were processed and eventually settled on the preparation. This was supported by Ajayi (2014) who reported that the production of African locust bean is basically by traditional household level fermentation technology laden with food-borne hazards at virtually all points during processing.

The seeds usually come with a substantial microbial load comprising spores of aerobic spore-forming bacteria and mould spores, most of which are drastically reduced during the prolonged boiling to soften the testa as obtainable in related fermentations (Gernah et al. 2007). Cooling is there after done in open air and this may introduce particulate and microbial contaminants coupled with the nature of local calabash and similar utensils used.

Besides, the presence of these fungal isolates on the deteriorating bean samples was supported by the work of Ogbonnaet al. (2015) who reported genera of Aspergillus, Mucor, Penicillium and Rhizopusus mycota of stored locust bean seeds from markets in Jos Nigeria and these isolates have been implicated in post-harvest rot of some fruits and vegetables in storage (Adaskaveg et al. 2002). In fact, the significance of Rhizopus sp, Aspergillus niger and A. flavus on stored foodstuffs stems not only from their spoilage potentials but also from their potentials to produce a variety of mycotoxins. These mycotoxins are poisonous even in minute concentrations and pose a threat to the health of consumers taking such contaminated samples. For instance, A. flavus, one of the fungal isolates is capable of producing mycotoxins known as aflatoxin which causes aflatoxicosis.

However, the non-deterioration of the salted locust bean samples by day 28 of storage cannot be connected with the salt treatment. The salt drew water out of the bean sample thereby lowering its moisture content to a level that inhibited fungal growth. The low water content is desirable according to the report of Qayyum et al. (2012) that high moisture content encourages growth of microorganisms which could reduce stability and shelf storage capability. Also, the salt treatment could have probably created hypertonic environment to any attacking moulds on the samples thus generating osmotic imbalance that eventually resulted to the destruction of the attacking moulds. This is in consonance with the report of Ademola et al. (2011) that salting is one of the oldest methods of preserving food and is used because most bacteria, fungi and other potentially pathogenic organisms cannot survive in a highly salty environment, due to the hypertonic nature of salt and that common salt is probably the oldest known antimicrobial agent. Consequently, this further confirmed the report of Elemo et al. (2011) that dehydration, salting and packaging of home produced food items such as locust beans (iru) in simple polyethylene bags is desirable to extend their shelf life.

Also, results from this study showed significant percent decrease (except in moisture contents) in the proximate compositions of the unsalted deteriorated locust bean samples when compared with the salted non-deteriorated samples. This was supported by the report of Ogbonna et al. (2015) that colonisation of the locust bean seeds by the storage fungi reduced significantly the nutrient composition of the seeds. This reduction in the nutrient values of the locust bean seeds is an evidence of utilisation of the nutrient contents of the seeds by the fungi. However, the increase in the moisture content of the unsalted deteriorated locust bean samples could be attributed to maceration of cellulose and pectin components of the beans cell wall by the fungal isolates. It could also be as a result of the biodeterioration activities of the spoilage fungi with the resultant elaboration of their metabolites into the surrounding bean seeds and the dissolution of the beans tissues into rot thereby increasing the moisture level of the bean seeds (Ogbonna et al. 2015). As storage duration extended to day 28, the carbohydrate, protein, crude fibre and total ash contents had further increased in the salted samples though insignificantly while the moisture and fat contents had also further reduced insignificantly. The further reduction in the moisture content probably accounted for further increase in the carbohydrate, protein, crude fibre and total ash contents of the salted samples because Adegunloye (2012) reported that lower moisture content causes reduction in solubility of nutrients in substrate while decrease in the fat contents could be as a result of oxidative activities (Zia-ur-Rehman et al. 2003).

Meanwhile, the relative significant increase in the concentrations of Na, K, Ca, Fe, Zn, Mg and Cu in the salted non-deteriorated samples throughout the storage period when compared with the unsalted samples that deteriorated by day 5 of storage in agreement with the observation of Ademola et al. (2011) who reported increase in concentrations of mineral elements in locust bean preserved and stored for up to 4 weeks. Addition of salt must have also contributed to the increase in the mineral concentrations particularly sodium and calcium. Besides, the lower concentrations recorded in the unsalted samples could not but be connected with the fact that the spoilage fungi that deteriorated the unsalted samples probably used these minerals as growth factors and this consequently resulted in their significant decrease in the unsalted samples.

<table>
<thead>
<tr>
<th>Table 1: Storage moulds found associated with the spoilage of African Locust Bean during storage at 28±2°C and 75±5% RH</th>
</tr>
</thead>
<tbody>
<tr>
<td>Samples</td>
</tr>
<tr>
<td>Aspergillus niger</td>
</tr>
<tr>
<td>Aspergillus flavus</td>
</tr>
<tr>
<td>Rhizopus sp.</td>
</tr>
</tbody>
</table>

Note: + = present, - = absent, RH = relative humidity
Table 2: Proximate compositions of unsalted and salted African locust bean during storage

<table>
<thead>
<tr>
<th>Samples</th>
<th>Moisture</th>
<th>CHO</th>
<th>Crude Fibre</th>
<th>Protein</th>
<th>Fat</th>
<th>Total ash</th>
</tr>
</thead>
<tbody>
<tr>
<td>Unsalted locust beans</td>
<td>63.50 ±0.25b</td>
<td>12.14 ±0.24a</td>
<td>11.83 ±0.29a</td>
<td>3.22 ±0.30b</td>
<td>7.44 ±0.06a</td>
<td>1.78 ±0.20a</td>
</tr>
<tr>
<td>by day 7 of storage</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Salted locust beans</td>
<td>24.50 ±0.24a</td>
<td>18.08 ±0.55b</td>
<td>16.50 ±0.55b</td>
<td>9.80 ±0.09a</td>
<td>26.50 ±0.09a</td>
<td>4.80 ±0.44b</td>
</tr>
<tr>
<td>day 7 of storage</td>
<td>0.14a</td>
<td>0.55b</td>
<td>0.55b</td>
<td>0.09a</td>
<td>0.16b</td>
<td>0.44b</td>
</tr>
<tr>
<td>Salted locust beans</td>
<td>22.52 ±0.16a</td>
<td>19.50 ±0.30b</td>
<td>17.20 ±0.60b</td>
<td>10.44 ±0.10a</td>
<td>24.18 ±0.10b</td>
<td>5.32 ±0.22b</td>
</tr>
<tr>
<td>day 28 of storage</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Note: Each value is a mean ± standard error of three replicates and means followed by the same letter are not significantly different (p> 0.05) using New Duncan’s Multiple Range Test.
CHO - Carbohydrate

Table 3: Mineral compositions of unsalted and salted African locust beans during storage

<table>
<thead>
<tr>
<th>Samples</th>
<th>Na</th>
<th>K</th>
<th>Ca</th>
<th>Fe</th>
<th>Zn</th>
<th>Mg</th>
<th>Cu</th>
</tr>
</thead>
<tbody>
<tr>
<td>Unsalted locust bean</td>
<td>1112.21</td>
<td>137.64</td>
<td>108.61</td>
<td>0.18±</td>
<td>6.00±</td>
<td>95.21</td>
<td>2.98±</td>
</tr>
<tr>
<td>by day 7 of storage</td>
<td>± 0.01a</td>
<td>± 0.03a</td>
<td>± 0.40a</td>
<td>± 0.01a</td>
<td>± 0.03a</td>
<td>± 0.02a</td>
<td>± 0.03a</td>
</tr>
<tr>
<td>Salted locust bean</td>
<td>1498.99</td>
<td>297.32</td>
<td>178.39</td>
<td>1.99</td>
<td>8.94</td>
<td>108.24±</td>
<td>4.96</td>
</tr>
<tr>
<td>by day 7 of storage</td>
<td>± 0.19b</td>
<td>± 0.21b</td>
<td>± 0.08b</td>
<td>± 0.01b</td>
<td>± 0.01a</td>
<td>± 0.01a</td>
<td>± 0.00a</td>
</tr>
<tr>
<td>Salted locust beans</td>
<td>1500.91</td>
<td>463.64</td>
<td>363.64</td>
<td>4.02</td>
<td>15.10</td>
<td>95.45</td>
<td>8.26</td>
</tr>
<tr>
<td>by day 28 of storage</td>
<td>± 0.15c</td>
<td>± 0.60c</td>
<td>± 0.66c</td>
<td>± 0.53c</td>
<td>± 0.60c</td>
<td>± 0.04a</td>
<td>± 0.20c</td>
</tr>
</tbody>
</table>

Note: Each value is a mean ± standard error of three replicates and means followed by the same letter are not significantly different (p> 0.05) using New Duncan’s Multiple Range Test.
Na – Sodium, K – Potassium, Ca – Calcium, Fe – Iron, Zn – Zinc, Mg – Magnesium and Cu – Copper.

CONCLUSION
This study showed that African locust bean (Parkia biglobosa) seeds can be preserved and stored for 28 days under ambient conditions after treatment with appropriate gram of common salt as experimented in the study without showing any evidence of deterioration or significant reduction in proximate and mineral values during storage. Consequently, salt treatment is hereby recommended for extending shelf life of locust bean seeds after harvest because salting was able to preserve the processed locust bean seed for 28days while unsalted seeds deteriorated fast by day 5 of storage.

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REFERENCES