

Full Length Research Paper

Evaluation of canned fermented oil bean seed slices during ambient storage

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African oil bean (*Pentaclethra macrophylla* Benth) seed slices were prepared, fermented and canned in three different media using conventional batch retort procedures. The canned products were subjected to sensory evaluation using a 20 man panel of judges, and to six months' ambient storage with monthly analysis being carried out for total viable counts, peroxide values, free fatty acids and acid values of the sample oils, as well as for hardness and appearance indices of the canned seed slices. The results show that after 6 months storage the total viable counts were 9.3×10^3 cfu/g, 1.7×10^4 cfu/g, and 6.0×10^3 cfu/g for the brine-canned, refined groundnut oil-canned and tomato sauce-canned samples, respectively. Peroxide values and free fatty acids had lowest concentrations in the product canned in refined groundnut oil; whereas the same product was least accepted organoleptically in terms of aroma and overall acceptability. The hardness and appearance indices show increased softening and colour darkening with prolonged storage period, although the samples were still considered acceptable after 6 months tropical ambient storage.

Key words: fermented oil bean seeds, canning, quality attributes, storage.

INTRODUCTION

Many tropical underutilized crop plants have seeds that are recently attracting worldwide attention because of their high nutrient potentials, especially with regard to solving the prevalent protein-energy malnutrition (PEM) that has ravaged populations in the less developed parts of the world. Some of these seeds have been exploited as soup bases, such as gbegiri from *Vigna unguiculata* (Akanbi, 1992); others are fermented and used as condiments and seasonings, such as okpei from *Prosopis africana* (Achi, 1992), and ogiri from *Ricinus communis* (Ogunfa, 1985).

The most prominent use of the African oil bean (*Pentaclethra macrophylla* Benth) seeds is the fermentation into ugba, a nutritious condiment and local snack, which is consumed by more than 40 million people in West tropical Africa (Enujiugha et al., 2002). Fermentation of African oil bean seed cotyledons plays many roles, including bringing about better nutrient availability and digestibility with significant softening of the cotyledons; formation of hydrolytic non-toxic products with flavour, texture and co-

lour that appeal to the consumer; and reduction in anti-nutritional and toxic factors (Enujiugha and Akanbi, 2002). However, the fermented ugba has restricted availability and exploitation because of its high rate of deterioration, as it is known to lose acceptability beyond 2 weeks ambient storage (Enujiugha, 2000). Softness increases progressively with storage duration at a very rapid rate, possibly due to the fact that the microbial and plant enzymes still remain active after fermentation and the major fermentative organisms still remain viable. Accompanying the increasing softness is the production of off-flavours and off-colours, as well as slimy dextrinous appearance.

The African oil bean seed contains more than 52% oil in its cotyledons (Enujiugha and Ayodele-Oni, 2003), with polyunsaturated fatty acids (especially linoleic and oleic acids) constituting more than 88% of the fatty acids in the seed oil (Enujiugha, 2003). This means that the seed and its products have potential predisposition to oxidative rancidity with attendant off-flavours, especially during tropical ambient (ca. 28 - 30°C) storage. Thermal processing has been applied in the preservation of the fermented oil bean seed slices with remarkable improvements in nutrient bioavailability, digestibility and function-

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nality (Enujiugha and Akanbi, 2005). However, the preservation of the quality and freshness of the fermented product is a critical factor in the choice of processing and preservation technique. Since thermal processing is viewed as an achievable technique for the maintenance of the preferred form and quality of ugba while at the same time ensuring the elongation of its shelf life (Enujiugha and Akanbi, 2002), it would be worthwhile to carry out storage studies on the canned product with a view to evaluating the impacts on some spoilage parameters.

As an oilseed, the major indicators of spoilage for the canned product are likely to be those that are useful in rancidity measurements, such as peroxide value and free fatty acids content. Also, since the fermented product becomes progressively softer with storage period, the measurement of softness via thumb pressure will show the level of acceptability during the storage period. In the present study, the quality of the canned ugba slices was evaluated with a view to determine the optimum storage duration under tropical ambient conditions.

MATERIALS AND METHODS

Preparation of the fermented seed slices

Freshly harvested African oil bean seeds were obtained from local farmers at Nri in eastern Nigeria and fermented in the laboratory using a modification of the previously outlined procedure (Enujiugha and Olagundoye, 2001). In short, parboiling of whole seeds was done for 4 h using heating plate, and after dehulling and slicing (3.5 - 5.0 x 0.2 - 0.4 cm), cooking of sliced cotyledons was done for 6 - 8 h. Slicing was done manually with a kitchen knife. The cooked oil bean seed slices were soaked in tap water overnight, washed in three changes of water, and spread out for 4 h natural inoculation on a matted surface, before being fermented at 30°C for 4 days.

Canning of the fermented product

The fermented oil bean seed slices were canned using 300 x 208 kk lac/lac cans with 300 kk lined ends (Carnaud Metal Box Nig. Plc., Ikeja - Lagos, Nigeria) in three different media (reflecting the use to which the fermented product is put); namely, tomato sauce, refined groundnut oil and brine solution. Can size was not varied since Taiwo et al. (1997) observed that variations of can size did not affect the D-value (decimal reduction time) in the thermal processing of cowpeas in tomato sauce. The tomato sauce used in the present study was prepared from the following ingredients: tomato puree (30% solids), 75%; flour, 5%; sugar, 14.5%; salt, 3%; vinegar, 1%; and spices, 1%; to give a total solids content of 14.0 ± 0.5% (Taiwo et al., 1997). The brine solution used in the study contained 1.5% sodium chloride and 3% sucrose (w/v) as described by Ekanayake and Nelson (1990) and Lu et al. (1984). The refined groundnut oil used was a commercial brand purchased from a local market at Ile-Ife, Nigeria.

Portions of the fermented ugba slices (about 100 g each) were placed in the cans and the appropriate medium added as described by Aubourg et al. (1990), with a headspace of about 15 mm. The cans were covered with lids, exhausted in a steam chamber for 15 min, and sealed immediately by means of a semi-automatic sealer (Dixie Model 23-500, MD-75, USA). Thermal processing was done at 116°C for 60 min in a still retort and the cans were cooled under

running water at 38°C and stored in a cool place (about 15°C) for 1 week preceding quality evaluation and sensory analysis.

Sensory evaluation and storage studies

A 20 man panel of judges based on familiarity with ugba flavour, colour and texture was constituted to assess the products on the characteristic sensory parameters of appearance, aroma, taste and texture, as well as overall acceptability. The samples for evaluation included commercially-available fermented ugba (control) and the canned product in tomato sauce, brine solution and refined groundnut oil after the 8 days post-canning stabilization period. Table water was provided between samples to cleanse the palate; also the sample presentation order was randomized for each assessor (Enujiugha, 2006). The products were assessed on a 7-point Hedonic scale, with 7 equaling *like very much* and 1 equaling *dislike very much*.

For the storage studies, the canned products were stored under shelf (tropical ambient) conditions (ca. 28 - 30°C) for a period of 6 months, and monthly analysis was carried out for physicochemical properties and total viable counts. The samples taken immediately after the 7 days stabilization period served as control (or day zero samples).

Microbiological analysis

A 10 g sample of the canned product was taken and homogenized in 90 ml of 0.1% peptone water (DIFCO, 0118 - 17). From the 10⁻¹ dilution, other decimal dilutions (10⁻², 10⁻³, 10⁻⁴, 10⁻⁵) were prepared and 0.1 ml of inoculums from each dilution level was used in the evaluation (Enujiugha and Badejo, 2002). The total viable counts were determined by the pour plate method, using Plate Count Agar (DIFCO, 0479 - 17) as the medium. The plates were incubated at 30°C for 24 - 48 h (Kilinc and Cakli, 2005), and examined for growth. Separate colonies were identified using colony counter and the results were expressed as cfu/g.

Physico-chemical analysis

The seed oils of the canned products were extracted using Soxhlet apparatus and the rancidity indices (peroxide value, free fatty acids content and acid value) were determined using the methods of Pearson (1976). The peroxide values were expressed as milliequivalents of peroxide oxygen per kg of sample (mEq/kg) while the free fatty acids were expressed as g oleic acid per 100 g of sample (g/100g). The acid values were expressed as mg NaOH per g of sample (mg NaOH/g). Hardness/softness index was determined using thumb pressure while the appearance/colour index was determined via visual observation.

Statistical analysis

All experiments were carried out in triplicates, and except for the sensory evaluation data, all other data obtained were expressed as means ± s.d. Data obtained from sensory evaluation were subjected to analysis of variance (ANOVA) as described by Snedecor and Cochran (1976). Differences among means were separated using Duncan's multiple range test (Duncan, 1955); significance was accepted at 5% level ($P \leq 0.05$).

RESULTS

Table 1 shows the results of sensory evaluation of the

Table 1. Results of sensory evaluation of the ugba samples*.

Aroma	Taste	Colour	Texture	Overall	Acceptability
Brine	4.90 ^{bc}	5.15 ^a	5.20 ^b	5.50 ^a	5.40 ^a
Sauce	5.20 ^{ab}	5.05 ^a	4.55 ^b	5.45 ^a	5.10 ^{ab}
G/oil	4.30 ^c	4.50 ^a	4.65 ^b	5.05 ^a	4.40 ^b
Retail**	5.75 ^a	5.15 ^a	6.25 ^a	5.60 ^a	5.55 ^a

*Means in column with the same following letters in superscript are not significantly different (P 0.05). Also 7 = like very much; 1 = dislike very much. **Commercially-available fermented seed slices.

Table 2. Total viable counts of the canned samples during ambient storage (cfu/g)*.

Storage Duration (months)	Total viable counts (cfu/g)		
	Brine-canned	G/oil-canned	T/sauce-canned
0	0	0	0
1	1.3 x 10 ²	2.9 x 10 ²	2.2 x 10 ²
2	1.8 x 10 ²	3.0 x 10 ³	2.5 x 10 ²
3	2.0 x 10 ²	3.2 x 10 ³	8.1 x 10 ²
4	3.5 x 10 ³	5.4 x 10 ³	2.0 x 10 ³
5	4.0 x 10 ³	9.5 x 10 ³	3.7 x 10 ³
6	9.2 x 10 ³	1.7 x 10 ⁴	6.0 x 10 ³

*Weighted means from replicated experiments (n = 3; P 0.05)

canned ugba samples compared with the commercially-available fermented ugba. The canned ugba in brine was comparable to the commercially-available ugba in terms of overall acceptability. The ugba samples canned in brine and sauce were found not to be significantly different (P ≥ 0.05) from the commercial sample in terms of flavour (aroma and taste) and texture. Most of the panelists remarked negatively on the darker brown colours of the canned samples compared to the commercial ugba. On a 7-point Hedonic scale, with 7 equaling *like very much* and 1 equaling *dislike very much*, the best rated sample in terms of aroma was the commercial *ugba* sample, closely followed by the fermented product canned in tomato sauce. However, in terms of colour and stickiness, the brine-canned sample was more preferred to the sauce-canned ugba. The product canned in refined groundnut oil was the least accepted by the panelists in terms of aroma and overall acceptability.

Table 2 gives the results of the total viable counts in the canned ugba samples during the six months' ambient storage. At the end of the storage period the total bacterial counts were 9.2 x 10³ cfu/g, 1.7 x 10⁴ cfu/g and 6.0 x 10³ cfu/g for the canned ugba samples in brine, refined groundnut oil and tomato sauce, respectively. The higher acidity of the tomato sauce and the higher salt concentration of the brine solution may have affected the luxuriant growth of the microorganisms in the samples. There were gradual increases in bacterial density in all the examined products, although the increases were not very significant (P 0.05) within the first three months of

storage.

The rancidity indices for the canned products (being oilseed-based) comprise peroxide and acid values as well as the free fatty acids content, as these are reflective of oxidative rancidity, total acidity and hydrolytic rancidity, respectively. The peroxide values of the seed oils of the canned ugba samples during 6 months of ambient storage are presented in Table 3. At the end of the storage period, the peroxide values for the samples were 11.63 meq/kg for brine-canned sample, 9.54 meq/kg for the vegetable oil-canned sample, and 10.02 meq/kg for the tomato sauce-canned sample. The peroxide value of the seed oil of brine-canned samples after 5 months storage and that of the tomato sauce-canned samples after 6 months storage were slightly higher than the value (10 meq/kg) recommended for fresh oils (Pearson, 1976). Table 4 shows the free fatty acids (FFAs) contents of the samples during the 6 months storage period. The free fatty acids increased from 0.16 ± 0.05 g to 3.12 ± 0.10 g in the brine-canned samples; from 0.12 ± 0.01 g to 2.54 ± 0.10 g in the groundnut oil-canned samples; and from 0.13 ± 0.02 g to 3.98 ± 0.05 g in the tomato sauce-canned samples within the 6 months storage period. Onyeike and Acheru (2002) obtained a value of 0.37 ± 0.10 g/100g for the raw oil bean seed cotyledons.

Table 5 gives the acid values of the extracted oils which is an indication of the total acidity as contributed by the fatty acids in the sample. The values obtained after 1 month ambient storage agree with the value (1.57 mg/g) obtained for fermented oil bean seeds by Okafor et al

Table 3. Peroxide values of samples during storage.

Storage Duration (months)	peroxide value (meq/kg)*		
	Brine-canned	G/oil-canned	T/sauce-canned
0	6.21 ± 0.40	6.30 ± 0.07	6.05 ± 0.12
1	7.15 ± 0.09	7.08 ± 0.51	7.00 ± 0.43
2	7.82 ± 0.20	7.20 ± 0.04	7.64 ± 0.22
3	7.97 ± 0.05	7.32 ± 0.06	7.90 ± 0.10
4	9.23 ± 0.57	8.02 ± 0.21	8.52 ± 0.04
5	10.04 ± 0.12	9.25 ± 0.20	9.07 ± 0.26
6	11.63 ± 0.80	9.54 ± 0.03	10.02 ± 0.70

*Figures represent means of triplicate determinations (mean ± s.d.) at 5% level.

Table 4. Free fatty acids (FFAs) contents of the products-oils during storage.

Storage Duration(months)	Free fatty acids content (g oleic acid)*		
	Brine-canned	G/oil-canned	T/sauce-canned
0	0.16 ± 0.05	0.12 ± 0.01	0.13 ± 0.02
1	0.92 ± 0.02	0.70 ± 0.05	0.65 ± 0.05
2	1.36 ± 0.05	1.20 ± 0.02	1.50 ± 0.10
3	2.01 ± 0.06	1.53 ± 0.10	2.48 ± 0.12
4	2.46 ± 0.10	1.63 ± 0.04	3.57 ± 0.03
5	2.97 ± 0.02	2.01 ± 0.07	3.70 ± 0.10
6	3.12 ± 0.10	2.54 ± 0.10	3.98 ± 0.05

*Figures represent means of triplicate determinations (mean ± s.d.) at 5% level.

Table 5. Acid values of oil samples from the canned products during storage.

Storage Duration(months)	Acid values (mg NaOH/g oil)*		
	Brine-canned	G/oil-canned	T/sauce-canned
0	0.30 ± 0.12	0.27 ± 0.07	0.22 ± 0.03
1	1.90 ± 0.06	1.43 ± 0.23	1.40 ± 0.09
2	2.73 ± 0.10	2.30 ± 0.05	2.98 ± 0.10
3	4.50 ± 0.03	3.15 ± 0.07	4.51 ± 0.12
4	4.47 ± 0.15	3.82 ± 0.04	6.98 ± 0.25
5	5.90 ± 0.08	4.55 ± 0.10	7.02 ± 0.13
6	6.43 ± 0.09	5.10 ± 0.07	7.92 ± 0.06

*Figures represent means of triplicate determinations (mean ± s.d.) at 5% level.

(1991) and Enujiugha (2000). After 6 months of tropical ambient storage, the acid values of the samples increased to 6.43, 5.10 and 7.02 mg/g for the brine-canned, groundnut oil-canned and tomato sauce-canned ugba, respectively. There were general gradual increases in acid values of the samples during the period of storage.

The results of the hardness (thumb pressure) and appearance (visual observation) indices of the canned oil bean samples during the 6 months storage period are presented in Table 6. The hard oil bean slices gradually became soft and melting as the storage period prolonged. The groundnut oil was able to preserve to some

extent the hardness of the canned seed slices more than both the brine solution and the tomato sauce. However, in all the cases, the texture changed with increasing storage duration. The canned oil bean slices were still acceptable after 5 months for the groundnut oil-canned product; after 4 months for the brine-canned product; and after 3 months for the tomato sauce-canned product. The results of the appearance index show that the desirable light brown colour of ugba was lost after 2 months storage in both brine-canned and tomato sauce-canned samples, and after 4 months in the groundnut oil-canned samples. The product became darker with increasing

Table 6. Hardness and appearance indices of the canned products during storage.

Storage Duration (months)	Hardness index			Appearance index		
	Brine	G/oil	T/sauce	Brine	G/oil	T/sauce
0	hard	hard	hard	Light brown	Light brown	Light brown
1	Slightly hard	hard	Slightly hard	Light brown	Light brown	Light brown
2	Slightly soft	Slightly soft	soft	brown	Light brown	brown
3	soft	soft	soft	Dark brown	Light brown	brown
4	soft	soft	Very soft	Dark brown	brown	Dark brown
5	Very soft	soft	Very soft	Dark brown	brown	Dark brown
6	melting	Very soft	Melting black	grayish	Dark brown	Dark brown

storage period. In the brine-canned sample the colour turned grayish black after 6 months ambient storage.

DISCUSSION

Sensory evaluation has shown that fermented seed slices of African oil bean canned in brine and tomato sauce using conventional thermal processing procedures had fairly comparable consumer acceptability to retail samples of fermented ugba. Product colour and appearance seemed to be the major problem in consumer acceptance of the sensory quality of the canned ugba. It has been observed that browning during storage and processing is a significant problem in the food industry and is believed to be one of the main causes of quality loss during post-harvest handling of legumes (Nagai and Suzuki, 2003). The acceptable colour of ugba is light brown; darker brown colours are undesirable. The colour changes are believed to be the result of both enzymatic and non-enzymatic browning. Polyphenol oxidase (a major enzyme contained in oil bean seed cotyledons) catalyzes the oxidation of phenolic substances to quinines, which spontaneously polymerize to form a brown pigment (Enujiugha and Akanbi, 2005a). In the present study, groundnut oil as a medium seemed to prevent the undesirable browning activities up to 5 months of storage.

The total viable counts of the canned product in brine and sauce were still within safe levels even after 3 months of ambient storage, which is indicative of their edibility and stability. For the product canned in groundnut oil, the microbial load rose sharply after 1 month of storage, revealing its lower safety ratio. The increasing microbial load with longer storage periods could be attributed to the product's low acidity level. Ugba is known to be a low acid food (Enujiugha and Akanbi, 2002; 2005), and such foods, according to Garbutt (1997), may need the addition of the antibiotic nisin to effectively prevent growth of microorganisms, especially during prolonged storage. However, the results of the present study show that products could safely be consumed after 6 months of tropical ambient storage. This is premised on the fact that the conventional thermal

processing schedule applied was enough to destroy all spores of pathogenic organisms.

The lower peroxide value recorded for the oil-canned product compared to the other samples is an indication of its relative stability to oxidative rancidity. Peroxide value correlates to some extent with the off-flavour caused by aldehydes and other oxidative rancidity products. A rancid taste often begins to be noticeable when the peroxide value is between 20 and 40 meq/kg. The results of the present study indicate that the canned products could still be consumed after 6 months tropical ambient (ca. 28°C) storage. Free fatty acids (FFAs) were generally low in the samples throughout the storage period. Free fatty acids content is an indicator of the level of hydrolytic rancidity of an oil, and this is usually enhanced by the seed lipases in the presence of moisture. Already a previous study (Enujiugha et al., 2004) has demonstrated an active lipase in dormant seeds of the African oil bean. The results of the present study show that the canned ugba slices in tomato sauce is highly prone to hydrolytic rancidity beyond 3 months of tropical ambient storage compared to those canned in the other media. This could be attributed to more hydrophilic molecules in solution. NaCl exerts a more drying effect so that the brine-canned samples are less prone to hydrolysis. Acidity increased with storage, indicating continued hydrolysis of the triglycerides and other macromolecules in the seed oil. The high margin of increase recorded in the tomato sauce-canned sample after 4 months of storage compared to those canned in the other media could be attributed to increased degradation of the macromolecules and subsequent accumulation of fatty acids in the oil, and other oil-based degradation products.

The canned products were progressively getting softer with longer storage periods. Rodrigo et al. (1992) and Mallidis and Katsaboukakis (2002) observed that thermal processing of foods to the level necessary to achieve commercial sterility causes considerable softening of the tissues. The results of the present study agree with this observation. The softening could also be attributed to internal enzyme hydrolysis. Enujiugha et al. (2002) suggested that the generally reported softening of the seed slices could be a consequence of enzyme hydrolysis of

macromolecules such as polysaccharides, peptides and glycerides.

Conclusion

Fermented African oil bean seed slices (ugba) have been canned in three different media and stored for 6 months under ambient conditions, with remarkable results. The results of total viable counts (TVC) and peroxide values show clearly that the products kept longer in refined groundnut oil, followed by tomato sauce, and finally brine solution. The hardness and appearance indices show increased softening and colour darkening with prolonged storage period. The free fatty acids and acid values were lowest in the product canned in refined groundnut oil. The products were still acceptable after the storage period.

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