

Effect of wrapping material on chemical and microbiological qualities of fermented melon seed (*Citrullus vulgaris L. Series*)

Peter-Ikechukwu I. Anne^{1,2}, Olawuni Ijeoma^{1,2}, Osobie C. Charles^{1,2*}, Odimegwu N. Euphresia^{1,2}, Uzoukwu E. Anthonia^{1,2}, Eluchie Chioma^{1,2}

¹Department of Food Science and Technology, Federal University of Technology, Owerri,
P. M. B. 1526 Owerri, Imo State (NIGERIA)

²Central Laboratory Service Unit, National Root Crop Research Institute (NRCRI),
Umudike, Umuahia, Abia State (NIGERIA)

E-mail : ccharles4@gmail.com

ABSTRACT

The effect of six different wrapping materials on melon seed (*Citrullus vulgaris L. series*) fermented traditionally was evaluated. Melon seed was sorted, washed, boiled (4 hours), dehulled and wrapped in blanched plantain leaf. It was then boiled again for 2 hours, drained, cooled and allowed to ferment naturally for 72 hours (primary fermentation). The fermented melon seeds were subsequently mashed and wrapped in six different wrapping materials (unblanched leaf, blanched leaf, aluminum foil, dried leaf, transparent polyethylene, black polyethylene), kept in a warm surrounding for another 72 hours (secondary fermentation). The resultant samples locally known as *Ogiri* were subjected to chemical and microbiological evaluation. Result of the chemical analysis showed increase in pH from 6 to 8. Titratable acidity decreased from 1.02% to 0.36% for primary fermentation and varied during secondary fermentation, with dried leaf having the highest (0.32%) and black polyethylene the lowest (0.18%). Amino nitrogen increased progressively during fermentation from 5.15% to 7.35% after the primary fermentation. The more the fermentation period progressed, the more the increased. © 2015 Trade Science Inc. - INDIA

KEYWORDS

Ogiri-egusi;
Fermented;
Melon seed;
Packaging/wrapping;
Effects;
Chemical composition;
Microbiological quality.

INTRODUCTION

Early man developed his own packaging technology using leaves for wrapping purpose and the skins of animals for the first flexible packaging material in the transportation of water and wine. Early container made by plaiting rushes and reeds as well as small wooden casks became evident^[1]. Packaging is an essential part of processing and distribution of foods whereas preservation is the major role of packaging^[1].

One major important role of packaging irrespective of whether it is the traditional method or the modern method is the preservation of food. The early man has practiced the preservation of food through various ways which includes the fermentation of food, drying of food over a fire or with the aid of sunlight and salting and, thereafter, packaging with various materials. In Nigeria, there is a wide range of fermented indigenous staple foods such as *iru*, *ogiri*, *ugba*, etc. These indigenous products are packaged with indigenous packaging

ing materials such as leaves, earthen wares etc.

In recent times, pre-processing treatment of traditional packaging materials (e.g. leaves) has not been explored to reduce the microbial load in traditionally packaged foods before use. Also since the advent of modern packaging materials, they have not been utilized in the area of packaging locally processed food condiment such as *ogiri*, *ugba*, etc. *Ogiri* is a fermented soup condiment processed from melon seed popularly known as *egusi* or from castor bean and wrapped in leaves by the processor who are mainly from the rural areas. Odibo *et al.* (1989) reported that *ogiri* has a flavoring agent whose characteristics and organoleptic properties depend on microbial activities from the traditional methods of preparation which is as a result of uncontrolled solid substrate fermentation. Steinkraus (1997) reported that fermented foods have a very good safety record even in the developing world, where foods are manufactured by people without training in microbiology or chemistry. But fermentation process itself cannot solve the problem of cross contamination by undesirable micro-organism in a packaged food. Packaging should provide adequate protection against such contamination.

However, *ogiri*, a preferred relish is locally packaged in leaves with little or no treatment and sometimes covered with cement papers. This may have contributed to the fast spoilage and even maggot infestation of the product. The use of treated and untreated local packaging material and modern packaging materials in the packaging of locally prepared *ogiri* will be explored in this study to ascertain the chemical and microbiological effects of packaging methods.

This study is aimed at determining the effect of both treated and untreated local packaging material and modern packages on the quality of *ogiri* produced from melon seed (*Citrullus vulgaris L.*) locally called *egusi*. Also, it is expected that the microorganisms involved in the fermentation of the melon seed wrapped in different packaging materials will be isolated and identified.

Success has not yet been made in the industrialization of *ogiri* production. Consequently, the traditional method has remained the applicable method of production with resultant cross-contamination issues and short shelf life of the final product, which usually bear high moisture content in a paste-like texture. Most of

the reasons for this hindrance to large scale production are related to low acceptability, offensive aroma, poor aesthetic quality and fear of microbial cross-contamination – all of which can be evaluated and controlled through improved packaging and strategic microbial studies. The findings of this research will significantly aid the solution to the problems of packaging encountered in *ogiri* production which has delayed its acceptability and industrialization to a large extent. Also, a better choice of packaging material for *ogiri* which plays the role of preserving the condiment could reduce microbial load and contamination and improve the chemical properties that enhance its flavour.

MATERIALS AND METHODS

Materials

Plant material

Melon seeds (*Citrullus vulgaris L. series*) were purchased from a local seller at Abeokuta, Ogun State, Nigeria. Some packaging materials such as aluminum foil, polyethylene were purchased from a store at Ihiagwa, Owerri west L.G.A Imo State (Nigeria).

Chemicals

Chemicals such as formaldehyde, potassium oxalate, n-hexane were purchased from store at Douglas and Mbaise road in Owerri while others such as sodium hydroxide, sulphuric acid, phenolphthalein, ethanol were obtained from the laboratory of the department of Food Science and Technology, Federal University of Technology, Owerri, Imo State. All chemicals and reagents were of analytical standards.

Equipment

Equipments used were mostly sourced from the department of Food Science and Technology and they were as follows: Muffle furnace (Carbolite Bamford, Sheffield, England, 530 2AU), Desiccator, Hot air oven (Genlab, model –Mino/50, Serial No-10co76), Analytical sensitive balance (item No: AR3130, OHAUS corp, China), Incubator (Uniscop SM9098, SURGUFIELD MEDICALS, ENGLAND), among others.

METHODS

Full Paper

Preparation of raw sample

Undehulled and dehulled melon seeds were subjected to cooking for 4-hours and subjected to chemical analysis in comparison with raw melon seed. After cooking, the undehulled and dehulled seeds were milled separately.

Traditional production of fermented melon seed (*Ogiri*)

Undehulled melon seeds were washed properly and severally and boiled for 4 hours, cooled and dehulled. The cotyledons were then soaked in water overnight and wrapped tightly in layers of blanched plantain leaves and then perforated with glass rod. The wrapped cotyledon was thereafter boiled for 2 hours, placed on a wire mesh over flame in order to reduce the moisture content for 1 hour. The wrapped cotyledon was then left to ferment at the prevailing ambient temperature (28°C) for 3 days (primary fermentation). At the end of the fermentation period, the seeds were pounded in a scientific mortar with pestle into a paste. The paste was subsequently distributed into the various packaging material which comprise unblanched leaf, blanched leaf, dried leaf, aluminum foil, transparent polyethylene and black polyethylene and left to ferment for 3 days (secondary fermentation).

Isolation of microorganism

Fermented samples of melon seeds were taken aseptically from traditionally fermented melon. 10 grammes of the sample was weighed and thoroughly mashed with laboratory pestle and mortar and mixed with 90ml of peptone water as diluent in a conical flask and the content was thoroughly shaken. Subsequent serial dilutions (10^{-2} , 10^{-3} , 10^{-4} , 10^{-5} , 10^{-6} , 10^{-7} , 10^{-8} , and 10^{-9}) were made from this solution by adding serially 1ml of solution from preceding concentration to 9ml of the diluent, using sterile pipette in McCartney bottle, the samples were screened for the presence of bacteria and fungi. Nutrient Agar was inoculated with 10^{-7} for more dilution of the sample (s) for fungi. The NA plates were incubated at 35°C for 24h and PDA plates were incubated at 25°C for 48 – 72h. The media used were prepared and incubated according to the labeled manufacturer instructions. The colonies were enumerated and expressed as colony forming unit per gramme (cfu/g).

Identification and characterization of isolate

Identification of isolated cultures was carried out based on the method described by Azu (2004). It was based on morphological, cultural and biochemical characteristics. The cultural and morphological characteristics were studied after incubation at 37°C for

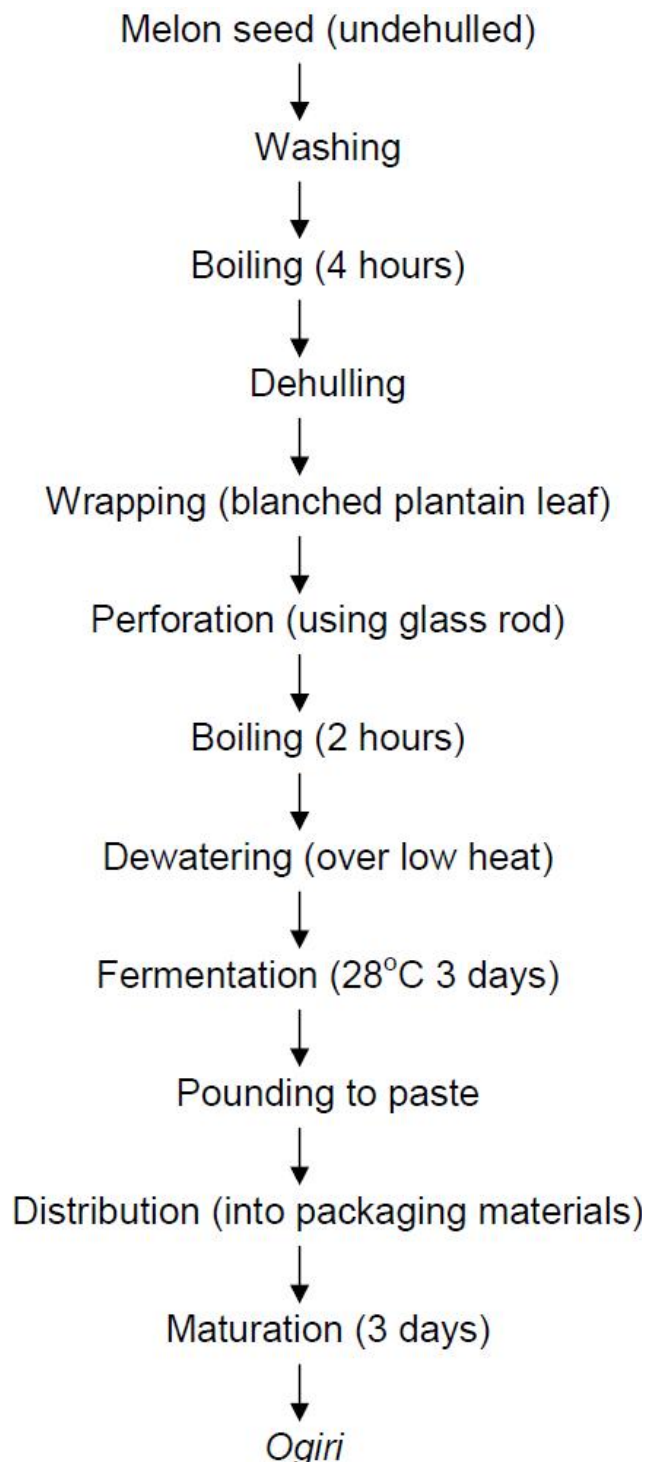


Figure 1 : Flow chart of production of *Ogiri*

24hours. Features recorded for cultural characteristics include shape, elevation, margin, color, surface, and opacity (optical characteristics). These were also carried out with the aid of microscope. After the cultural and morphological examination of the cultures, all isolates were subjected to the following biochemical tests: catalase test, oxidase test, indole test, citrate test and starch hydrolysis test.

Gram stain test

A smear was made by suspending a twenty-four hours old culture in small drops of distilled water at one end of a clean and grease-free slide. The smear was heat-fixed by passing it over a Bunsen flame for about three times, then flooded with crystal violet for one minute, thereafter washed under gentle running tap water and flooded with Lugol's iodine solution for 30 minutes. It was blotted dry, decolorized with 95% ethanol until the solvent flowed colourless from the slide. Subsequently, it was washed with water and flooded with safranin for two minutes. It was also blotted dry and afterwards examined under the microscope using oil immersion objective (x 100).

Motility test

A semi solid Nutrient Agar medium was prepared by dissolving 30 grammes (instead of 28g as specified by the manufacturer) of the nutrient agar powder in 1 litre of distilled water, dispensed in test tubes, sterilized, then cooled to 47°C and allowed to solidify before use. A twenty-four hour old pure culture was collected using a sterile inoculating needle and was stabbed in a straight manner into the semi-solid agar tubes respectively and these were incubated at 37°C for 24hours.

Catalase test

This test aids in the determination of microorganism that contain catalase enzyme capable of releasing oxygen gas when mixed with hydrogen peroxide. A drop of 3% peroxide was placed on a clean and grease-free slide. A loopful of twenty-four hour old culture was placed on the drop and was examined for the appearance of bubbles. The evolution of bubbles indicated a positive reaction.

Oxidase test

A twenty-four old culture was smeared onto a Whiteman filter paper impregnated with 1% solution of *tetramethyl-p-phenylenediamine hydrochloride*. The appearance of a light pink colour that immediately changes to purple indicated the presence of the cytochrome enzyme, "oxidase."

Indole test

This test was used determine bacteria capable of breaking down the amino acid, tryptophan to indole. 3% peptone water was prepared dispensed in test tube and sterilized at 121°C for 15 minutes. The medium was allowed to cool and was inoculated with a twenty-four old hour culture and incubated at 37°C for 2 – 3days. The presence of indole after incubation was detected by adding a few drops of Kovac's reagent (5g p-dimethylaminobenzaldehyde, 57g amyl alcohol and 25ml Conc. HCl) to the culture. Ring-like coloration on top of the tube indicated a positive result.

Citrate test

Koser's citrate medium was prepared according to manufacturer's instruction and dispensed into a test tube. The test tube was sterilized by autoclaving. The medium was inoculated from peptone water culture. The medium was incubated at optimum growth temperature for up to 7 days. Growth in the medium involves citrate utilization which is shown through turbidity in the medium.

Coagulase test

In 18 – 24hours, nutrient agar culture was used for the test. A slide was marked with a grease pencil into two sections. A loopful of peptone water was dropped on each until a homogeneous suspension was obtained, then a drop of the culture was added to the suspension and stirred for 5 seconds. The reaction was observed at 37°C. A positive result was indicated by clumping which was not re-emulsified and the second suspension served as control.

Starch hydrolysis test

Fresh cultures (24-hours old) were inoculated on starch agar plates by single line inoculation and plates incubated for five days at 30°C. After incubation, the plates were flooded with Lugol's iodine. Cultures which produced a colorless clear zone around the line of

Full Paper

streaking were recorded as positive while those that produced blue coloration throughout the medium and around the growth line were recorded as negative.

CHEMICAL ANALYSES

Chemical analysis was carried out as described by Han *et al.* (2001).

Titrateable acidity determination

The titrateable acidity of the inoculated seeds (expressed as percentage lactic acid) for each fermentation process was measured on inoculation and at 24-hours interval, for primary and secondary fermentation. 5 grammes of the mashed seed was dissolved in 100ml distilled water, 2 – 5 drops of phenolphthalein was added and carefully neutralized with 0.1N Sodium hydroxide (NaOH) and titrated with aqueous 0.1N Sodium hydroxide (NaOH). The solution was constantly swirled until a pink colour persisted for 15 seconds. The titre value was recorded and the titrateable acidity was calculated as % lactic acid.

$$\% \text{ TTA (\% Lactic Acid)} = \frac{\text{Titre value} \times 0.09 \times N \times 100}{Wt}$$

Where: N = Normality of NaOH; 0.09 = Equivalent weight of predominant acid; Wt = Weight of sample used

Amino nitrogen determination

The amino nitrogen of the mashed seeds for each fermentation process was measured at 24-hours interval, for primary and secondary fermentation. 2 grammes of the mashed sample was placed in a conical flask, and then 0.5 ml of phenolphthalein (0.5%) and 0.4ml of neutral saturated potassium oxalate was added. The mixture was kept to stand for few minutes and this was neutralized with 0.1N NaOH to a standard pink colour. 2ml of 40% formaldehyde solution was added and allowed to stand for few minutes (until mixture was colourless), then titration with 0.1N NaOH to pink colour. The titre value obtained is designated 'V'.

$$\% \text{ Amino Nitrogen} = \frac{N \text{ NaOH} \times V \text{ NaOH} \times 100}{\text{Mass of sample}}$$

Where: N = Normality; V = Titre value

Temperature and pH determination

Temperature was measured with a thermometer and

pH was measured with universal pH paper.

ANALYSIS OF DATA

The data obtained from this study were statistically analyzed using Analysis of Various (ANOVA). The means were separated using Fischer's least significant difference (LSD).

RESULTS AND DISCUSSION

Microbial assessments

Total viable count for primary fermentation and secondary fermentation

The result of the total viable count on fermented melon seed is shown in TABLE 1. The total viable count increased exponentially from 0hour-72hours both for bacteria and fungi. This indicates that the organisms were at their exponential phase of growth (Azu, 2004) and that the fermenting melon seed was a suitable substrate for the microorganism to grow. Azu (2004) also described total viable count as the total number of living cells in a substrate. At 0-hour, the fermented melon had 8.5×10^5 cfu/g of bacteria count which increased to an uncountable number (blurred) while there was no fungi count at 0-hour.

The result of the total viable count on fermented melon seed packed in various packaging materials for secondary fermentation at 24hours and 72 hours is shown in TABLE 2. An increase was observed in the total viable count for both bacteria and fungi. The sample packaged in Aluminum foil had the least bacteria count of 9.6×10^6 cfu/g after 72 hours while unblanched leaf was the highest in bacteria count (2.44×10^7 cfu/g). Sample packaged in dried leaf had the least fungi count of 1.18×10^4 cfu/g at 72hours, followed by aluminum foil, while unblanched leaf had the highest fungi count of 2.00×10^4 cfu/g. The least bacteria count observed in aluminum foil could be because the aluminum foil has a characteristics of being non-porous thereby preventing the entrance of external growth factor such as moisture, air for microbial growth^[11]. Dried leaf showing the least count for fungi could be inferred to the drying operation carried out on the leaf which reduced the leaf's moisture content making it unavail-

able to the substrate for further microbial activity.

Cultural, morphological and microscopic characteristics of bacteria and fungi detected

The result in TABLE 3 shows the cultural and morphological characteristics of bacteria detected during the first 48 hours of fermentation. Two types of colonies were observed for the bacteria on the nutrient agar plate. The colonies had length 1-4mm. Azu (2004) reports that the usefulness of using size as a distinguishing feature is however limited by the fact that the sizes change with changes in prevailing condition. Colonies growing in conditions of over-crowding tends to be considerably smaller than those of the same organism growing on places containing only a few colonies. So, it could be assumed that the length of the organism in the agar plate was small to the over-crowdiness in the agar plate. The organism on the agar plate showed regular and irregular shape, they were creamy in colour with an entire and serrated margin. The bacteria have low convex and flat elevation with surface appearance which was either

moist and shiny or dull and dry. The characteristics showed a continuous and consistent succession of microorganism with similarities.

The result in TABLE 4 shows the cultural and microscopic characteristics of fungi isolated during the first 48 hours of the fermentation. No growth on the potato dextrose agar plate on the 0-hour.

However *Sacharomyces cerevisiae* (yeast) was identified after 24hours of fermentation. This is an organism known to ferment carbohydrates, breaking them down to sugar in cereal based product mainly; its presence therefore has to do with the carbohydrate content

TABLE 1 : Total viable count on fermented melon (*Ogiri*) seed (Bara L. Series) for primary fermentation at 28°C

Fermentation period (Hours)	Bacterial count	Fungi count
0 hour	8.5×10^5 cfu/g	-
24 hours	2.2×10^6 cfu/g	2.56×10^3 cfu/g
48 hours	3.68×10^6 cfu/g	2.85×10^3 cfu/g
72 hours	Blurred	1.4×10^4 cfu/g

TABLE 2 : Total viable count on fermented melon (*Ogiri*) seed (Bara L. Series) for secondary fermentation at 28°C

Packaging Materials	Bacteria Count		Fungi Count	
	24 hours	72 hours	24 hour	72 hours
Unblanched leaf	2.12×10^5 cfu/g	2.44×10^7 cfu/g	Not visible	2.00×10^4 cfu/g
Blanched leaf	1.42×10^6 cfu/g	1.81×10^7 cfu/g	1.4×10^3 cfu/g	1.62×10^4 cfu/g
Dried leaf	1.18×10^6 cfu/g	1.43×10^7 cfu/g	3.7×10^2 cfu/g	1.18×10^4 cfu/g
Aluminum foil	7.9×10^5 cfu/g	9.6×10^6 cfu/g	2.38×10^3 cfu/g	1.43×10^4 cfu/g
Polyethylene (Transparent)	1.19×10^6 cfu/g	2.08×10^7 cfu/g	Blurred	1.78×10^4 cfu/g
Polyethylene (Black)	1.16×10^6 cfu/g	1.56×10^7 cfu/g	Blurred	1.58×10^4 cfu/g

TABLE 3 : Cultural and morphological characteristics of bacteria detected

Sample/ Hours	Number/ type of colonies	Size(mm)	Shape	Colour	Elevation	Margin	Surface
0 hour	1	2-3mm	Regular	Creamy	Low convex	Entire	Moist and shiny
	2s	2mm	Irregular	Creamy	Flat	Serrated	Dull and Dry
24 hours	1	2-4mm	Irregular	Creamy	Flat	Serrated	Dull and Dry
	2	2mm	Regular	Creamy	Flat	Entire	Moist and shiny
48 hours	1	1-2mm	Irregular	Creamy	Flat	Serrated	Dull and dry
	2	1mm	Regular	Creamy	Low convex	Entire	Moist and shiny

TABLE 4 : Microscopic characteristics of fungi isolated

Hours	Number/Type of colony	Colonial characteristics	Microscopic appearance	Identity of isolates
0 hour	-	NO GROWTH	-	-
24 hours	1	Tiny creamy circular colonies	Gram positive large Spherical budding cells	<i>Sacharomyces Cerevisiae</i>
48 hours	1	Tiny creamy circular colonies	Gram positive large Spherical budding cells	<i>Sacharomyces Cerevisiae</i>

Full Paper

TABLE 5 : Microscopic and biochemical characteristics of bacteria isolated

Hour	Type of colony	Microscopic characteristics	Catalase	Oxidase	Coagulase	Indole	Citrate	Motility	Sugar Fermentation					Most Probable Organism Identified
									Gluc	Sucr	Lact	Malt	Mann	
0	1	Gram Positive spherical chain	+	-	-	-	+	-	+	+	+	+	+	Enterococcus
Hour	2	Gram +ve rod short chain	+	-	-	-	+	-	+	+	+	+	+	Bacillus spp
24	1	Gram +ve rod short chain	+	-	-	-	+	-	+	+	+	+	+	Bacillus spp
Hours	2	Gram positive spherical chain	+	-	-	-	+	-	+	+	+	+	+	Enterococcus Faecalis
48	1	Gram +ve rod short chain	+	-	-	-	+	-	+	+	+	+	+	Bacillus spp
Hours	2	Gram +ve rod spherical chain	+	-	-	-	+	-	+	+	+	+	+	Enterococcus faecalis

Where: +ve = Positive; Gluc = Glucose; Sucr = Sucrose; Lact = Lactose; Malt = Maltose; Mann = Mannitol

of melon seed. Traditional fermented castor oil seeds (*Ogiri-isi*) have been reported to be contaminated by fungi (Iwuoha and Eke, 1996) possibly from the processing environment. It could be deduced that the presence of fungi during fermentation was due to contamination since it is not noted in previous research work on fermenting melon.

Microscopic and biochemical characteristics of bacteria isolated

The result in TABLE 5 shows the biochemical test conducted on the bacteria colonies during the fermentation period. *Bacillus species* and *Enterococcus faecalis* were the most predominant organism identified from the biochemical test. Osho *et al.* (2009) reported that quite a number of *Bacillus species* have been isolated from various fermented food condiments, although yeast and other bacteria are also seen; only part of them can be considered to play a substantial role in fermentation process. Iwuoha and Eke (1996) also reported that the combination of *Bacillus species* and *Alcaligene species* in *Ogiri-egusi* was capable of producing the quality characteristics of a good *Ogiri-egusi*. From this report, it could be deduced that *Bacillus spp.* is the main fermenting organism in melon seed which can also be confirmed from the result of the biochemical test conducted on fermented melon seed (Bara L. series). It can also be said that the fermentation of melon seed is not effected by *Bacillus species alone* especially with dependence on natural inoculum. There is evidence that a combination of *Bacillus species* and

TABLE 6: Result of chemical analysis on fermented melon primary fermentation

Parameters	24 hours	48hours	72 hours
Temperature	36°C	28°C	28°C
pH	6	7	8
Total Titratable Acidity	1.02%	0.96%	0.36%
Amino Nitrogen	5.15%	6%	7.35%

other bacteria which in this study is *Enterococcus faecalis* actually effected natural fermentation of melon seed.

CHEMICAL ASSESSMENT

Chemical assessment after primary fermentation of melon seed

The result in TABLE 6 shows the chemical parameters that were monitored during the primary fermentation of melon seed (Bara L. series). Temperature decrease was observed from 36°C (24 hours) to 28°C (48 hours to 72 hours) during fermentation. The initial temperature was as a result of the drop in the cooking temperature of the melon seeds which finally reduced to the prevailing ambient temperature of 28°C as at the time of fermentation. Also an increase in pH of the fermented seed was observed during fermentation (6 – 8). Odunfa (1981) reported that pH of *Ogiri-egusi* increase from 6.5 to 8.1 and fermentation temperature from 29°C – 30°C. The pH level was measured with universal pH paper which was not able to give the exact value of the pH except for approximated value. The

TABLE 7 : Chemical assessment at secondary fermentation (at 28°C)

Packaging Material	pH		TTA		Amino Nitrogen	
	24 hrs	72 hrs	24 hrs	72hrs	24 hrs	72hrs
Unblanched leaf	7	8	1.03%	0.19%	25.25%	33.25%
Blanched leaf	7	8	1.32%	0.31%	23.55%	32.75%
Dried leaf	7	8	1.07%	0.32%	26.25%	34.25%
Aluminum foil	7	8	0.96%	0.31%	18.25%	32.75%
Transparent Polyethylene	7	8	0.87%	0.20%	28.25%	36.05%
Black polyethylene	7	8	1.54%	0.18%	25.75%	34.25%

rise in pH during production of alkaline fermented food is due to the ability of the dominant microorganism *Bacillus Species* to hydrolyze protein into amino acid and ammonia. The higher pH values of fermented legumes compared to other materials under similar condition have been attributed to their higher protein content^[2]. Therefore, the increase in the pH of fermented melon (Bara L. series) would be attributed to its high protein content and the *Bacillus Species* that hydrolyzed the protein to amino acid and ammonia. TABLE 6, also shows a decrease in titratable acidity from 1.02% to 0.36%. Titratable acidity is counted as lactic acid content which is produced by lactic acid bacteria or enzyme from the raw material^[10]. The result of the total titratable acidity obtained shows the percentage of lactic acid that was produced during fermentation of melon seeds. Lactic acid if produced in good quantity in food would prevent the activities of some micro-organisms which could cause spoilage in food. In alkaline fermented food, lactic acid production would not be much benefit due to the desired alkaline nature of the product expected at the end. Increase in pH leads to decrease in total titratable acidity and increase in the rate of alkaline fermentation^[10].

There was an increase in the amino nitrogen produced during fermentation of melon seed (TABLE 6) as the period of fermentation increased. Amino nitrogen can be used as a good index of yeast growth^[7] and as an indicator for autolysis and microbial degradation of food protein^[8] as well as an indicator for increase in PH value^[5]. The increase in pH of the fermenting melon seed provided a conducive environment for *Saccharomyces cerevisiae* which was identified microbiologically as one of

Chemical assessment of the secondary fermentation of melon seed

The result in TABLE 7 shows the chemical parameters that were examined during the secondary fermentation when the primary fermented melon seed paste was packed into various packaging materials (leaves, aluminum foil, transparent and black polyethylene). The prevailing temperature was constant at 28°C and pH was within the range of 7–8. The total titratable acidity at this stage varied among the packaging materials. The result showed that samples packaged in black polyethylene, unblanched leaf and transparent polyethylene had low total titratable acidity of 0.18%, 0.19% and 0.2% respectively at 72 hours while dried leaf has the highest total titratable acidity of 0.32%. Also the result shows a drastic increase in the amino nitrogen. Aluminum foil and blanched leaf had the least value (32.7% respectively) while the transparent polyethylene was the highest (36%) at 72hours. Due to the ammonium ion produced from the amino content, fermented melon seed was observed to have a lightly putrid aroma which is also a known characteristic of fermented melon seed (*Ogiri*). The increase in amino nitrogen for the transparent-polyethylene-packaged could be attributed to the passage of light rays through the packaging material when compared to others. It is known that light favours some of the chemical reactions involved in the breakdown of food nutrients and therefore it is assumed that there was more breakdown and production of amino nitrogen in transparent polyethylene compared to others.

CONCLUSION

From the result obtained at the end of this work, changing the traditional packaging method of *Ogiri* discourages excessive microbial load, and pre-treated traditional leave packaging has lower microbial load than

Full Paper

the untreated (unblanched). Also, modern packaging materials maintains and improves the chemical composition of *Ogiri* such as pH and amino nitrogen, which are greatly responsible for the characteristic flavor of the condiment. Therefore, *Ogiri* can be packaged in modern packaging materials such as aluminum foil or polyethylene for enhancement of the microbial and aesthetic qualities which will improve its utilization in the country and beyond.

RECOMMENDATION

Due to the inherent problem associated with *Ogiri* utilization in the area of ease of spoilage, it is recommended that further research be embarked on to produce dried *Ogiri* packaged with different packaging materials from the primary fermentation stage in order to check the quality of the dry product in flavor performance as well as study the effects of the packaging material with the aim of increasing its marketability in developing and developed countries.

REFERENCES

- [1] A.I.Ihekoronye, P.O.Ngoddy; Integrated Food Science and Technology for the Tropics, Macmillan, London (1985).
- [2] A.I.Sanni, G.S.Agenor, E.Sakyi-Davion, S.Sefa-Dehah; Aerobic Spore Forming Bacteria and Chemical Composition of Some Nigerian Fermented Soup Conditions Soup Condiments, Plant Foods Human Nutr., **55**,111-118 (2000).
- [3] A.Osho, O.O.Mabekoje, O.O.Bello; Comparative Study on the Microbial Load of Garri, Elusbo-Isu and Iru in Nigeria. Afr. J. Food Sci., **4**(10), 646-649 (2009).
- [4] Association of Official Analytical Chemicals (AOAC), Official Methods of Analysis Washington, DC (2000).
- [5] B.Z.Han, R.R.Beumer, F.M.Rombouts, M.J.R.Nout; Microbiological Safety and Quality of Commercial Sufu – a Chinese Fermented Soybean Food, Food Control, **12**, 541-547 (2001).
- [6] C.I.Iwuoha, O.S.Eke; Nigerian Indigenous Fermented Foods their Traditional Process Operation, Inherent Problems, Improvements and Current Status. Food Research International, **29**, 527 – 540 (1996).
- [7] C.Lekkas, A.A.Stewart, A.E.Hill, B.Taid, J.Hodgson; Elucidation of the Role of Nitrogenous Wort Components in Yeast Fermented. J. Inst. Brew., **113**, 3-8 (2007).
- [8] F.G.Ijong, Y.Olita; Microflora and Chemical Assessment of an Indonesia Traditional Fermented Fish Sauce Bakasang, J. Fac. Appl. Biol. Sci., **34**, 95-100 (1995).
- [9] K.H.Steinkarus; Classification of Fermented Foods: Worldwide Review of Household Fermentation Technique, Food Control, **8**, 311-3 (1997).
- [10] L.D.Vugst; Technology Aspect Related to the Application of Functional Starter Cultures, Food Techno. Biotechnol., **38**,105-112 (2002).
- [11] N.N.Potter, J.H.Hotchkiss; Food Sciences, 5th Ed., Chapman and Hall, New York, U.S.A (1995).
- [12] O.Azu; Laboratory Guide for Microbiology, Crystal Publishers, Okigwe, Imo State, Nigeria (2004).
- [13] S.A.Odunfa; A Note on the Microorganisms Associated with the Fermentation of African Locust Bean (*Parkia filicoideea*) During Iru Production, J. Plant Food, **3**, 245-250 (1981).