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Effect of Different Packaging Material on the Quality of Fermented Melon Seeds

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Abstract. Effect of different packaging material on the quality of fermented melon seeds was investigated. Melon (Citrullus vulagaris) seeds were boiled and separately packaged in glass jar, aluminum foil, uma leaves (Thaumatococcus daniellii), nylon/polyethylene and plastic materials. The packaged melon seeds (egusi) were each allowed to ferment for five days to produce five samples of the ogiri-egusi. The samples were subjected to proximate, mineral, vitamin, sensory and microbial analysis. The values of the proximate composition ranged from 39.43 - 41.13%, 24.12 - 29.53%, 20.03 - 25.71%, 1.24 -1.70%, 0.25 - 0.37% and 2.67 - 14.46% for moisture, sprotein, fat, ash, fibre and carbohydrate content respectively. The mineral analysis showed magnesium (1.27 - 1.90)mg/100g), sodium (1.76 - 2.05 mg/100g), potassium (1.13 - 1.73 mg/100g) and calcium (1.74 - 1.87 mg/100g). The values obtained for all the minerals in the samples had significant (p<0.05) differences. Evaluation of the ogiri-egusi samples revealed vitamin A (19.88 -22.08µg/100g), vitamin B1 (0.01 - 0.04mg/100g), vitamin B2 (0.01 - 0.05mg/100g) and vitamin B3 (0.04 - 0.11 mg/100g) respectively. The viable microbial count on the samples indicated baccilus species, pseudomonas, proteus and lactobacillus being dominant while enterobacter was not detected. The packaging materials significantly influenced the qualities of the samples, uma leaves supported fermentation rate and microbial growth more than other packaging materials. Glass and foil seem to prolong the shelf life more than uma leaves in terms of texture which depicts delayed fermentation rate. Samples A had the least microbial load while all the isolated and identified bacteria were more dominant in sample C. Generally, all the packaging materials did not impart objectionable qualities to the ogiri.

Keywords: melon seed, fermentation, packaging, shelf-life

Introduction

Food packaging is an integral part of food processing, which entails the use of some materials in the wrapping of foods (Ihekoronye & Ngoddy, 1985; Ayo, 2003). The success of most preservation methods depends on appropriate packaging. However, faulty packaging will undo all that a good processor has attempted to accomplish by the most meticulous manufacturing process. The earliest forms of packaging pre date written history and were found in nature, and progressed as man-made advancement in the production of food. Leaves, goatskin, woods, leathers, etc. were traditional packaging materials used by the early man. The beginning of modern packaging originated from the industrial revolution which changed the structure of society and concentrated large numbers of people in towns and cities, so altering their habits and creating a great demand for similar qualities of various products in large replication (Peter-Ikechukwu et al., 2014). Today, packaging materials such as glass, metals (like tinplates, tin-free steel and aluminum), plastics (like homogenous films, coated films, cellophanes, etc.), papers (e.g. paper boards and fibre boards) and laminates are now being widely used (Ahenainen, 2003). Although the early man developed his own packaging using leaves for wrapping purposes, and the skins of goat as the first flexible packaging material in the transport of water and wine (Awa & Okaka, 2005), the use of leaves is still being widely practiced, especially in the packaging of indigenous fermented products like

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Ogiri, Ugba, Iru, etc. Ogiri' is a food condiment produced from the fermentation of melon seeds (Citrullus vulgaris), fluted pumpkin (Telferia occidentallis), castor oil seeds (Ricimus communis) and African oil bean seeds (Pentaclethra macrophylla). These raw materials are used to create the different varieties of "ogiri" such "ogiri-egusi," "ogiri-ugba", "ogiri-isi" and "ogiri-mkpuru ugu" (Achi, 2005). Ogiri-egusi has gray colour with porous structure and sharp smell. It is adjudged to be an indigenous fermented soup condiment which is used as flavouring agent whose character and organoleptic properties depend on microbial activities (Nwosu and Ojimelukwe, 2000). The production of 'ogiri' has been limited to household level and only women are involved in its production. "Ogiri" is use as a flavouring agent in soups, sauces and vegetable dishes especially by the Ibos (Cooper, 2007) and also serve as a nutritious non-meat protein substitute (Achi, 2005). The nutrient qualities of ogiri-egusi as influenced by different packaging materials are not widely known to its manufacturers and consumers. The nutrient qualities of ogiri-egusi as influenced by different packaging materials are not widely known to its manufacturers and consumers. This research will help to proffer solution to the problem of packaging encountered in ogiri production which has delayed its industrialization to a large extent. This in turn will help to improve the acceptance of the product at both local and probably international market.

Materials and Methods

Material Procurement

The raw castor oil seed and packaging materials (foil, glass bottle, plastic container, nylon and *uma* leaf) were obtained from Ore-Akokwa in Ideato South L.G.A. of Imo State.

Sample Preparation

Castor oil seeds (*Ricinus communis*) was dehulled and then sorted to remove bad seeds and unwanted materials. The cotyledons was wrapped in blanched banana leaves and boiled for 8h to soften it. Then, it was left to ferment at the ambient temperature $(32-35^{\circ}C)$ for 4 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 then wrapped with different packaging materials which comprises of aluminum foil, glass bottle, plastic, nylon and blanched *uma* leaf. The samples were left to ferment for another 5 days (secondary fermentation).

Proximate Analyses

The proximate analysis was carried out according to the standard procedures of AOAC (2010).

Moisture Content Determination

Two grams of each sample was weighed out into the crucible. The sample was put into a moisture extraction oven at 105° C and heated for 3 hours. The dried sample was put into desiccators, and allowed to cool and reweighed. The process was repeated until a constant weight was obtained. The drying and weight of moisture lost was determined and expressed as a percentage. It is calculated as shown below.

% moisture =
$$\frac{W_2 - W_3}{W_2 - W_1} \times \frac{100}{1}$$

Where W_1 = weight of empty can, W_2 = weight of dish + undried sample, W_3 = weight of dish + dried sample

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Figure 1: Production of Ogiri

Protein Determination

The micro Kjeldahl method described by AOAC (2010) was used. Two grams of each of the samples was mixed with 10ml of concentrated H_2SO_4 in a heating tube. One table of selenium catalyst was added to the tube and mixture heated inside a fume cardboard. The digest was transferred into distilled water, Ten millimetre portion of the digest mixed with equal volume of 45% NaOH solution and poured into a kjeldahl distillation apparatus. The mixture was distilled and the distillate collected into 4% boric acid solution containing 3 drops of methyl red indicator. A total of 50ml distillate was collected and titrated as well. The sample was duplicated and the average value taken, the nitrogen content was calculated and multiplied with 6.25 to obtain the crude protein content. The percentage nitrogen (%N) was given by:

% N =
$$\frac{(100 \text{ x N x14 x VF}) \text{ T}}{100 \text{ x V2}}$$

Where: N = Normality of the titrate (0.1N), VF = total volume of digest =100ml, T = Titration value, Va = Aliquot volume distilled. % Protein = %N x 6.25

Ash Content Determination

A measured weight two gram of each sample was put in a previous weighed crucible, heated in a moisture extraction oven for 3h at 100C before being transferred to a muffle furnace set at 550° C and allowed to burn until the sample becomes white and free of carbon. The sample in the crucible was carefully removed from the furnace (take care not to allow air blow the ash away and cooled in desicators). It was pre weighed. The weight of ash was obtained and in percentage using the formula:

% Ash =
$$\frac{\text{weight of ash}}{\text{weight of sample}} \times \frac{100}{1}$$

Crude Fibre Determination

Crude fibre was determined using the method of AOAC (2010). About five gram of each sample was weighed into a 50ml, Erlen meyer flask was added. It was brought to boil and refluxed. For exactly 40 minutes counting from the starting of boiling. The flask was removed from the heater, cooled a little then filtered through a 15.0cm number 4 whatmann paper, the residue was washed with hot water, stirred once with spatula and transferred to a desiccator and was weighed as W_1 . It was burnt in a muffle furnace at 500⁰C for 6h and allowed to cool and reweighed as W_2 .

% crude fibre =
$$\frac{W_1 - W_2}{W_0} \times \frac{100}{1}$$

Where: W_1 = weight of crucible + fiber + ash, W_2 = weight of crucible + ash, W_0 = dry weight of the samples

Determination of Fat Content

The soxlet extraction method as described by AOAC (2010) was used in determining fat content of the samples. About two grams of the sample was weighed. Also the weight of the flat bottomed flask was taken with extractor mounted on it. The thimble was held half way into the extractor and the weighed sample. Extraction was carried out at temperature range of $40-60^{\circ}$ C. The thimble was plugged with cotton wool. At completion of the extraction, the solvent was removed by evaporation on a water bathe and the remaining part in the flask was dried at 80° C for 30 minutes in the air oven to dry the fat and then it is cooled in a desiccator. The flask was reweighed and percentage of fat calculated as

% fat =
$$\frac{\text{Weight loss}}{\text{Weight of samples}} \times \frac{100}{1}$$

Determination of Carbohydrates

The carbohydrate was calculated as

Carbohydrate = 100 - (M + A + P + F + CF)Where: P= protein, F= fat content, CF= crude fibre, A= ash, M= moisture

Determination of Mineral Compositions

Five grams (5g) of each sample was heated gently over a Bunsen burner flame until most of the organic matter is destroyed. This is further heated strongly in a muffle furnace for several hours until white-grey ash is obtained. The ash material is cooled. About 20ml of distilled water and 10ml of the dilute hydrochloric acid was added to the ashed material. This mixture was boiled, filtered into a 250ml volume trick flask, cooled and made up to volume. Minerals content of each sample was analysed using spectrophotometric method. Samples was analysed for potassium (K), calcium (Ca), iron (Fe), magnesium (Mg), and phosphorus (P). The concentration of each mineral was calculated as follows

<u>Absorbance of sample \times Concentration of standard solution \times Dilution factor</u>

Absorbance of standard solution × Sample Volume

Determination of Vitamins

Determination of vitamin B1 (Thiamine)

Thiamin content was determined using the spectrophotometric method described by AOAC (2010). A weighed portion of each ogiri sample (2g) was extracted by homogenizing it in 50ml molar ethanol sodium hydroxide solution. The homogenate is filtered to obtain the extract in 100ml flask. Ten (10) milliliters of the filtrate is pipetted and the colour developed by addition of 10ml of potassium dichromate and the wavelength is read at 360nm with Gen-

Way spectrophotometer at 360nm. A standard solution of thiamine was prepared and treated the same way as the sample. The thiamine content was calculated as shown;

Thiamine (mg/100g) =
$$\frac{100}{W} \times \frac{AU}{AS} \times C$$

Where: W = Weight of ugba sample analysed, AU = Absorbance of the sample, AS =Absorbance of the standard, C = Concentration of the standard

Determination of riboflavin (vitamin B2) content

Riboflavin content of ugba was determined by the method described by AOAC (2010). Two (2) grams of ogiri sample were extracted with 100ml of 50% ethanol solution shaken for one hour. The suspension is filtered into 100ml flask. A measured volume (10ml) of 30% hydrogen peroxide was added and allowed to stand for about 30 minutes. A 2ml of 41% sodium sulphide solution was added which will result to formation of a yellowish pale colour. The absorbance was measured in a Gen-Way spectrophotometer at 500nm wavelength. Riboflavin (Vit. B₂) was calculated in mg/100g using the formula below.

Riboflavin (mg/100g) =
$$\frac{AV}{AS} \times \frac{C}{1} \times \frac{100}{1}$$

Where: AV = Absorbance of ugba sample, AS = Absorbance of standard, C = Concentrationof Standard vitamin

Determination of niacin (vitamin B3)

The method described by AOAC (2010) was used to determine vitamin B3 content of the samples. Five (5) grams of the sample were treated with 50ml of 1N sulphuric acid and shaken for 30minutes. Three (3) drops of ammonia solution were added to the sample and filtered. Ten (10) milliliters of the suspension were pipeted into a 50ml volumetric flask and 5ml of 0.02NH₂SO₄ and absorbance was measured in the spectrophotometer at 47nm wavelength. This is used to plot the calibration curve. The thiamine riboflavin and niacin content can be calculated thus;

% thiamin, riboflavin or niacin =
$$\frac{AU}{AS} \ge C \frac{100}{1} \ge \frac{V_f}{V_a}$$

Where: AU = Absorbance of test sample, AS = Absorbance of the standard, C =Concentration of the standard, V_f = Volume of total extract, W = Weight of sample, V_a = Volume of extract analysed

Determination of vitamin A

This was determined using the method described by Capel and Dorrell (2005). The sample was weighed (2g) into a flat bottom reflux flask and 10ml of distilled water was added followed by careful shaking. This was followed by the addition of 25ml of alcoholic potassium hydroxide solution and the attachment of reflux condenser. The mixture was heated in boiling water bath for 1 hour with frequent shaking and rapidly cooked with 30ml of distilled water added. The hydrolysate obtained was transferred into a separating funnel and the solution was extracted three times with 250ml quantities of chloroform. Two grams of Anhydrous Sodium tetraoxosulphate (Na₂SO₄) was added to the extract to remove any traces of water. The mixture was filtered into a 100ml volumetric flask and made up to mark with chloroform.

Standard solutions (within the range of 0 to 50 micron gram/ml) prepared was determined with reference to their absorbance from which average gradients was taken to calculate Vitamin A (Beta-Carotene in micron gram/100 gram). Absorbance of each sample and standards was read on the spectrophotometer (Spectronic 21D, Milton Roy Model) at a wavelength of 328nm.

Calculation; Vitamin A (micron gram/100g) = $[(Absorbance \times dilution Factor) / (weight of fa$ sample)] \times 100/1

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Microbiological Analysis

One millilitre of each sample was put in 9ml of sterile distilled water in sterile test tubes, shaken and then serially diluted. From the appropriate dilution, 0.1ml was inoculated separately onto Nutrient agar and MacConkey agar plates and spread evenly using sterile bent glass rod. Each experiment were carried out in triplicates to get a mean standard value of the colony forming units (cfu/ml) on the plates. The inoculated Nutrient agar and MacConkey agar plates were incubated at 30°C and 35°C for 24 hours. After the period of incubation, the colonies on the plates were counted and recorded as colony forming unit per millilitre (cfu/ml) and coli form respectively. Each of the bacteria colonies on the agar plates were subcultured and the pure culture obtained. Isolates were identified by carrying out tests which include Gram staining, spore staining and biochemical tests such as catalase, coagulase, oxidase, citrate utilization, indole, methyl red, urease, Voges Proskauer and sugar fermentation.

Sensory Evaluation of the Ogiri Samples

The sensory evaluation was carried out using the multiple comparison tests. The samples were served to 30 semi trained panelists made up of staff of Imo State University, Owerri and some married women who are familiar with the sensory attributes (taste, aroma, flavour, colour, mouth feel, etc.). They were respectively provided with water to rinse their mouth after testing each sample and were instructed to carry out the evaluation for appearance, taste, aroma, mouthfeel and general acceptability using a hedonic scale. Where: 1 = dislike extremely, 2 = dislike very much, 3 = dislike moderately, 4 = dislike slightly, 5 = neither like nor dislike, 6 = like slightly, 7 = like moderately, 8 = like very much, 9 = like extremely.

Statistical Analysis

Data generated in this research was subjected to analysis of variance (ANOVA) as described by Ihekoronye and Ngoddy (1985). Tukey's test was used to separate means and the differences between the means were considered to be significant at P<0.05.

Results and Discussion

Proximate Composition

The proximate parameters evaluated were moisture content (MC), protein, fat, ash, fibre and carbohydrate (CHO). The statistical analysis revealed significant differences (p<0.05) across all these parameters in the ogiri samples.

Moisture Content of the Ogiri Samples

The percentage moisture content of the ogiri-egusi wrapped/packaged with different materials ranged from 39.43% to 41.13%. The highest moisture content was obtained in ogiri-egusi packaged in uma leaves (41.13%) and the least obtained in glass packaged ogiri-egusi (sample A) with significant differences (p<0.05) existing across all the samples. The values presented in Table 1 revealed that the moisture content of all the samples packaged with modern packaging materials (sample A, B, D and E) had lesser moisture content when compared to the sample packaged in conventional packaging materials (sample C). The trend of this results corresponds to the findings of Osobie *et al.* (2014) were lower moisture content recorded by sample a (Glass) could be attributed to non-porous nature of glass which did not create any means for moisture migration. High moisture content in food has been shown to encourage microbial growth (Sanni & Oladapo, 2018). Lower residual moisture content in food is advantageous in that microbial proliferation is reduced and storage life may be

prolonged if stored in appropriate packaging materials under good environmental conditions (Okoye & Ebujie, 2018). Thus, glass may be an appropriate packaging material to be used in elongating the shelf lives of the fermented melon seed product.

Sample	Parameters (%)					
	MC	Protein	Fat	Ash	Fibre	Carbohydrate
А	39.43° <u>+</u> 0.03	24.12 ^e +0.05	20.03 ^d +0.03	1.70 ^a <u>+</u> 0.02	$0.26^{c} \pm 0.02$	14.46 ^a +0.04
В	$40.02^{d} \pm 0.04$	28.15 ^b +0.02	25.71ª <u>+</u> 0.07	1.48 ^b +0.03	0.37 ^a +0.04	4.27 ^d +0.01
С	41.13 ^a +0.02	29.53 ^a +0.05	25.10 ^b +0.04	$1.24^{c} \pm 0.01$	$0.32^{b} \pm 0.02$	2.67 ^e +0.06
D	40.70 ^b +0.01	$26.22^{d} \pm 0.06$	21.26° <u>+</u> 0.03	1.46 ^b +0.02	0.27 ^c +0.01	11.09 ^b +0.08
Е	40.28° <u>+</u> 0.09	26.38° <u>+</u> 0.05	21.20° <u>+</u> 0.07	$1.44^{b} \pm 0.02$	$0.25^{\circ} \pm 0.03$	10.45° <u>+</u> 0.03
LSD	0.04	0.11	0.13	0.08	0.05	0.23

Table 1: Proximate composition of ogiri – egusi packaged in different packaging
materials

Note: All values are means of triplicate determinations in Percentage (%) \pm SD. Means on the same column with different superscripts are significantly (p<0.05) different from one another. Key: A - Ogiri packaged with glass, B - Ogiri packaged aluminium Foil, C - Ogiri packaged uma leaves, D - Ogiri packaged nylon, E - Ogiri packaged plastic, LSD - Least significant difference

Protein Content of Ogiri Samples

The results (Table 1) above show that the percentage protein content of the ogiri-egusi samples ranged from 24.12 ± 0.05 to $29.53 \pm 0.05\%$. The results showed significant (p<0.05) differences amidst all the samples. It was observed that sample wrapped in uma leaves had the highest protein content of $29.53 \pm 0.05\%$ than the other samples packaged in modern packaging material (glass, foil, nylon and plastic). This result is in complete agreement with the report of (Osobie *et al.*, 2020) on ogiri-egusi wrapped in dried banana leaves which retained more protein than other samples wrapped in aluminum foil, black polyethylene and transparent polyethylene. The higher protein contained in sample wrapped conventionally (sample C) could be attributed to the porous nature of leaves which created an aerobic environment for fermentation and microbial growth (Oboh, 2006). The primary functions of protein are to build body tissues, regulate functions such as muscle, contraction and blood pressure, synthesize enzymes and some hormones (such as insulin, that regulate communication among organs and cells) and other complex substances that govern body processes (Fashakin, 2008).

Fat Content of Ogiri Samples

The fat content of the ogiri-egusi samples ranged from 20.03% to 25.71%. The sample wrapped with aluminum foil (sample B) had the highest fat content $(25.71 \pm 0.07\%)$ while sample packed in glass (sample A) had the least value (20.03 ± 0.03) for fat. There were no significant (p>0.05) differences between samples packed in Nylon (sample D) and plastic (sample E) while samples A, B and C were significantly (p<0.05) different from one another. Iru *et al.* (2020) reported a fat content in the range of 20.78 ± 0.86 to $24.29 \pm 0.80\%$ for ogiriegusi wrapped in aluminum foil, dried banana leaves, paper, black cellophane and transparent cellophane. The values were found to be closely related to the values obtained from this present study. The sample packed in glass (sample A) beside its low moisture content may have a longer shelf life because Ojewumi (2016) reported that low fat content in fermented products could contribute to extension of shelf stability due to its resistance to rancidity.

Ash Content of Ogiri Samples

The ash content of the samples was within 1.24% to 1.70% with least ash content recorded by ogiri-egusi wrapped with leaves (sample C). There were no significant difference

(p>0.05) between the mean ash content obtained in ogiri-egusi samples packaged with aluminum foil, nylon and plastic. Ire *et al.* (2020) reported a higher ash content (3.09%) for ogiri-egusi wrapped in leaves and therefore is not in agreement to the findings of this present work. The ash content of food is a reflection of the mineral content present in the food.

Fibre Content of Ogiri Samples

The values obtained from the determination of fibre content of ogiri-egusi packed in different packaging materials ranged from 0.25% to 0.37% with the highest value obtained by sample wrapped in aluminum foil (sample B) and the least mean value obtained by sample packed in plastic container (sample E). The fibre content of samples packaged in glass, nylon and plastic container were found to be non-significantly (p>0.05) different from each other. The values obtained from this research work corresponds to values (0.25 ± 0.05 to $0.34 \pm 0.02\%$) recorded by Osobie *et al.* (2014) but were found to be low when compared with 1.95 ± 0.36 to $3.87 \pm 0.24\%$ reported by Ire *et al.* (2020) for ogiri-egusi packaged in different packaging materials. Fibre is needed to assist in digestion and keep the gastrointestinal tract healthy, and to also help keep the blood sugar stable (Usman, 2012). However, the low crude fibre content of ogiri-egusi cannot be seen as disadvantageous since the product is only but a food condiment.

Carbohydrate Content of Ogiri Samples

The percentage carbohydrate content of the ogiri-egusi samples which ranged from 2.67 ± 0.06 to $14.46 \pm 0.04\%$ had the least value with ogiri-egusi wrapped in Uma leaves (sample C) and sample A, with the highest percentage of carbohydrate content ($14.46 \pm 0.04\%$). The least significant difference of 0.23 was obtained for the carbohydrate content of the ogiri samples which indicated significant differences (p<0.05) across all the samples. The reduction or variation in carbohydrate content of the samples could be traced to fermentation by the micro-organisms. According to Ihekoronye and Ngoddy (1985) the hydrophilic molecules in carbohydrate take up moisture in proportion to the relative humidity of the environment. So the high moisture content of sample wrapped in leaves (sample C) could be attributed to this. This characteristic behavior encourages moisture uptake and apparent reduction in percentage of carbohydrate (Akinoso & Raji, 2011) and perhaps could be the reason why sample wrapped in uma leaves had the least percentage carbohydrate content.

Mineral Composition of Ogiri-Egusi Samples

The mineral composition of ogiri-egusi packaged in glass, aluminum foil, uma leaves, nylon and plastic containers are shown in Table 2 below. The minerals that were analysed includes; magnesium, sodium, potassium and calcium.

Magnesium Content of Ogiri Samples

The magnesium content of the ogiri-egusi samples ranged from 1.27 ± 0.02 to $1.90 \pm 0.03 \text{ mg}/100 \text{ g}$. The results depicted in Table 2 revealed that the magnesium content of samples packaged in aluminum foil (sample B) and uma leaves (sample C) were found non-significantly (p>0.05) different from each other but significantly (p<0.05) different from samples packaged in glass (sample A), nylon (sample D) and plastic container (sample) respectively. The magnesium content of these samples were found to be low when compared to $78.4 \pm 3.7 \text{ mg/kg}$ recorded by Oboh (2006) for condiment produced from melon seed. But higher than $0.31 \pm 0.01 \text{ mg}/100 \text{ g}$ reported by Okafor *et al.* (2015). Magnesium is an activator of many enzyme system and maintains the electrical potential in the nerves (Adeyeye & Agesin, 2007). It works with calcium to assist in muscle contraction, blood clotting and the regulation of blood pressure and lung function (Swaminathan, 2003).

Sodium Content of Ogiri Samples

The ogiri-egusi samples have sodium content within the range of 1.76 ± 0.05 to 2.05 ± 0.06 mg/100g with the least value recorded by sample wrapped in aluminum foil (sample B) and sample packaged in plastic (sample E) recorded the highest sodium content of 2.05 mg/100g. In spite of the least value obtained by sample B, there were no significant differences (p.0.05) between it and samples A, C and D respectively. It was observed that the sodium content obtained from these samples are low when compared to 45.90 ± 2.70 mg/100g reported by Oboh (2006). However, sodium is normally consumed in food as salt and it is essential in the regulation of osmotic pressure of the body fluid. It also aids in the transportation of CO₂ in the blood. Furthermore, sodium is one mineral whose intake is considered a factor in the etiology of hypertension, hence its low intake is encouraged (Okoka, 2005). This therefore means that the low content of sodium in the ogiri-egusi samples could justify their being considered as healthy foods.

Sample	Parameters (mg/100g)					
	Magnesium	Sodium	Potassium	Calcium		
А	1.27 ^c <u>+</u> 0.02	1.85 ^b +0.04	1.43 ^b +0.04	1.82 ^a +0.03		
В	1.90 ^a +0.03	1.76 ^b +0.05	$1.13^{c} \pm 0.02$	1.76 ^b +0.03		
С	1.86 ^a +0.03	1.79 ^b +0.04	1.13 ^c +0.06	1.74 ^b +0.02		
D	$1.40^{b} \pm 0.01$	1.86 ^b +0.03	1.71 ^a +0.02	1.81 ^a +0.03		
Е	$1.37^{bc} \pm 0.04$	$2.05^{a} \pm 0.06$	1.73 ^a +0.03	1.87^{a} +0.02		
LSD	0.13	0.11	0.15	0.08		

Table 2: Mineral composition of ogiri – egusi packaged in different packaging materials

Note: Means on the same column with different superscripts are significantly (p>0.05) different from each another. Key: A - Ogiri packaged with glass, B - Ogiri packaged aluminium foil, C - Ogiri packaged uma leaves, D - Ogiri packaged nylon, E - Ogiri packaged plastic, LSD - Least significant difference

Potassium Content of Ogiri Samples

The potassium contained in the ogiri-egusi sample packaged in different packaging materials were found to be in the range of 1.13 ± 0.02 to 1.73 ± 0.03 mg/100g. There were no significant (p>0.05) difference between samples B and C and samples D and E respectively. The potassium contained in all the samples were found to be higher than 0.85 ± 0.0 mg/100g reported by Okafor *et al.* (2015) on ogiri-egusi. The samples may be considered very fair sources of potassium. Potassium is primarily an intercellular cation, mostly this cation is bound to protein and with sodium influences osmotic pressure and contribute to normal pH equilibrium (Adeyeye & Agesin, 2007).

Calcium Content of Ogiri Samples

The calcium content of the ogiri-egusi samples ranges from 1.74 ± 0.02 to 1.87 ± 0.02 mg/100g with the least value obtained from sample wrapped in uma leaves (sample C), while sample packaged in plastic container (sample E) had the highest mean value for calcium. These were no significant difference (p>0.05) between samples A, D and E and samples B and C respectively. It was observed that sample packaged in plastic container (sample E) retained more of sodium, potassium and calcium than the other samples packaged in glass, aluminum foil, uma leaves and nylon (samples A, B, C and D) respectively. The calcium content obtained from this present study were found to be higher than 0.58 \pm 0.00 mg/100g reported by Okafor *et al.* (2015) but lower than 11.60 \pm 1.20 mg/Kg reported by Oboh (2006). Calcium is an essential micronutrient and its deficiency is more prevalent than any other mineral (Norman & Joseph, 2007). Calcium deficiencies results chiefly in bone and teeth diseases like rickets and osteoporosis. Therefore the consumption of these ogiri samples may be of help.

Vitamin Composition of the Ogiri-Egusi Samples

The results of the vitamin content of the ogiri-egusi samples packaged with different packaging materials is shown in Table 3 below.

Vitamin A Content

The vitamin A content of the fermented melon (ogiri-egusi) seeds are presented in $19.88 \pm 0.08 \mu g/100 g$, $20.11 \pm 0.05 \mu g/100 g$, $22.08 \pm 0.41 \mu g/100 g$, Table 3 as follows: 20.09±0.11µg/100g and 20.02±µg/100g for ogiri-egusi samples packaged in glass jar (Sample A), aluminum foil (Sample B), uma leaves (Sample C), nylon (Sample D) and plastic container (Sample E) respectively. The ogiri-egusi sample wrapped in uma leaves (Sample C) had the highest vitamin A content (22.08µg/100g) and were found to exhibit significant difference (p<0.05) from other samples (A, B, D and E) which on the other hand exhibited no significant differences (p>0.05) from each other. Okwunodulu et al. (2020) reported vitamin A content in the range of 11.63µg/100g to 16.03µ/100g which were found to be lower than values obtained from this present work. Okwunodulu et al. (2020) fermented blends of castor oil seeds and melon seeds in different ratios and observed that vitamin A content of the ogiri-egusi increased with increase in melon seeds. Thus, this could be the reason for high vitamin A content in this present study. The highest value for vitamin A obtained by sample C could be attributed to liberation of Vitamin A as fermentation progresses (Bradford, 2015). Vitamin A is a fat soluble vitamin and a powerful antioxidant. It plays a critical role in maintaining healthy vision, neurological function, healthy skin and support immune function. It is involved in reducing inflammation through fighting free radical damage (Bradford, 2015).

Vitamin B1 Content of Ogiri Samples

The values obtained for the thiamin content of the ogiri-egusi samples ranged from 0.01 \pm 0.001 to 0.04 \pm 0.0012 mg/100g. From the results presented in Table 3, it was observed that ogiri-egusi sample wrapped with leaves (sample C) had the highest thiamin content (0.04 \pm 0.012 mg/100g) and was significantly (p<0.05) different from the other samples. The higher thiamin present in sample C may be attributed to the leaching out of vitamin B1 from uma leaves into the fermented melon seed paste as the vitamin is water soluble and the moisture content of sample C is high and could have initiated the leaching out of the vitamin B1. Ileola *et al.* (2020) reported a vitamin B1 content value of 0.13 mg/100g for fermented melon seed and is higher than the values obtained from this present study. Thiamin plays an important role in the utilization of carbohydrate for supply of energy, where it functions as the coenzyme thiamin pyrophosphate, or cocarboxylase, in the oxidation of glucose (Norman & Joseph, 2007). Absence of vitamin B1 results in a specific deficiency disease called beriberi.

Samples	Parameters (mg/100g)					
	Thiamine (Vit. B1)	Riboflavin (Vit. B2)	Niacin Vit. A(µg)			
А	0.01° <u>+</u> 0.001	0.01° <u>+</u> 0.001	$0.04^{b}\pm0.0219.88\pm0.08$			
В	0.02 ^b <u>+</u> 0.01	0.03 ^b +0.01	0.06 ^b <u>+</u> 0.03 20.11 <u>+</u> 0.05			
С	0.04ª <u>+</u> 0.012	0.05ª <u>+</u> 0.02	0.11 ^a <u>+</u> 0.04 22.08 <u>+</u> 0.41			
D	0.02 ^b <u>+</u> 0.01	0.03 ^b +0.001	0.06 ^b +0.03 20.09+0.11			
E	$0.02^{b} \pm 0.01$	$0.02^{bc} \pm 0.01$	$0.05^{b} \pm 0.02 \ 20.02 \pm 0.3$			
LSD	0.01	0.02	0.04			

 Table 3: Vitamin composition of ogiri-egusi packaged in different packaging materials

Note: Means on the same column with same superscripts are insignificantly different (p>0.05) from one another. Key: A - Ogiri packed in glass container, B - Ogiri wrapped with aluminium foil, C - Ogiri wrapped with uma leaves, D - Ogiri wrapped with nylon, E - Ogiri packed in plastic container, LSD - Least significant difference

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Vitamin B2 Content of Ogiri Samples

The riboflavin content of the ogiri-egusi obtained from this research work fell within 0.01 to 0.05 mg/100g. There were no significant difference (p>0.05) in the vitamin B2 content of samples B, D and E and amidst samples A and E. Sample wrapped in uma leaves was observed to have the highest vitamin B2 content and could as well be attributed to the leaching out of vitamin B2 from the leaves of Uma plant into the fermented melon seed paste. Ileola *et al.* (2020) reported a vitamin B2 content ($1.14 \pm 0.69 \text{ mg}/100\text{g}$) that is higher than those obtained from this study. The utilization of vitamins during fermentation could also have influenced the result of this research. The United State recommended dietary allowance (USRDA) for vitamin B2 is 1.70 mg/100g (Usman, 2012). Riboflavin helps in the oxidation processes of living cells and is essential for cellular growth and tissue maintenance (Norman & Joseph, 2007).

Vitamin B3 Content of Ogiri Samples

The niacin content of the ogiri-egusi samples were as followed; 0.04 mg/100g, 0.06 mg/100g, 0.011mg/100g, 0.06 mg/100g and 0.05mg/100g for samples A, B, C, D and E respectively. Sample wrapped in uma leaves (sample C) had the highest (0.11mg/100g) vitamin B3 content and is significantly (p<0.05) different from other samples (samples A, B, D and E) which did not have any significant difference (p>0.05) amongst one another. A niacin content of 1.62mg/100g for fermented melon seed was reported by Ileola *et al.* (2020) and is not in agreement with the lower values obtained from this research work. Vitamin B3 is important as it helps the body to convert food into glucose, used to produce energy, maintain normal skin and mucous membranes as well as aid the function of the nervous system (Asdrubal, 2012).

Microbial Count of Ogiri-Egusi Samples

The bacterial isolates identified in the ogiri-egusi samples were Bacillus species, Pseudomonas species, Proteus species and Lactobacillus species. While Enterobacter was not found present in the substrate, just as reported by Ukaoma *et al.* (2018).

Bacillus Species in the Ogiri Samples

The total viable count of *Bacillus* species present in the ogiri-egusi samples packaged in different materials and fermented for a period of 24, 72 and 120 hours (5 days) ranged from 9.25 x 10^6 cfu/g to 16.34 x 10^6 cfu/g; 9.71 x 10^6 cfu/g to 19.27 x 10^6 cfu/g and 10.04 x 10^6 cfu/g to 21.47 x 10^6 cfu/g respectively. *B*. species was found to be most predominant in ogiri-egusi sample wrapped with uma leaves (sample C) while sample packaged in glass jar (sample A) had the least total viable count of *B*. species throughout the fermentation period. The high total viable count recorded by sample wrapped in uma leaves (sample C) could be attributed to the porous nature of the leave, as well as high moisture content of the sample while materials like glass, nylon and foil are not (or poorly) permeable to both oxygen and moisture. It was observed that progression in the fermentation period brought about increase in the total viable count of the *B*. species. The result of this study justifies the assertion of Ogbuonye (2018) that *B*. species was the predominant bacteria involved in the fermentation of ogiri.

Table 4: Microbial count of ogiri – egusi samples							
Sampla	Parameters (CFu/g)						
Sample	Bacillus spp	Pseudomonae spp	Proteus spp	Enterebacter	Lactobacillus		
DAY 1							
А	9.25x10 ⁶	1.87×10^{6}	2.47×10^{6}	-	2.74×10^{6}		
В	11.13×10^{6}	2.34×10^{6}	3.14×10^{6}	-	3.80×10^{6}		
С	16.39x10 ⁶	3.95x10 ⁶	4.61×10^{6}	-	5.33x10 ⁶		
D	11.25×10^{6}	2.38×10^{6}	3.23x10 ⁶	-	3.82x10 ⁶		
E	11.07×10^{6}	2.29×10^{6}	3.06x10 ⁶	-	3.57×10^{6}		
DAY 3							
А	9.71x10 ⁶	2.20×10^{6}	2.77×10^{6}	-	3.15x10 ⁶		
В	13.29×10^{6}	3.41×10^{6}	4.54×10^{6}	-	4.61×10^{6}		
С	19.27×10^{6}	5.74×10^{6}	6.37x10 ⁶	-	7.28x10 ⁶		
D	13.47×10^{6}	3.47×10^{6}	4.64×10^{6}	-	4.56x10 ⁶		
E	13.13x10 ⁶	3.31×10^{6}	4.42×10^{6}	-	4.3010^{6}		
DAY 5							
А	10.04×10^{6}	2.42×10^{6}	3.03×10^{6}	-	3.70×10^{6}		
В	15.19x10 ⁶	4.25×10^{6}	5.48x10 ⁶	-	5.34×10^{6}		
С	21.47×10^{6}	6.86x10 ⁶	8.02×10^{6}	-	9.70×10^{6}		
D	15.24x10 ⁶	4.38×10^{6}	5.13x10 ⁶	-	5.41x10 ⁶		
Е	15.14×10^{6}	4.24×10^{6}	5.03×10^{6}	-	5.25x10 ⁶		

Note: A = Ogiri-Egusi packaged in glass, B = Ogiri-Egusi packaged with aluminium foil, C = Ogiri-Egusi packaged with uma leaves, D = Ogiri-Egusi packaged with nylon, E = Ogiri-Egusi packaged with plastic

Pseudomonas Species of Ogiri Samples

The total viable count of *Pseudomonas* species present in the fermented melon seeds paste (ogiri-egusi) samples ranges from 1.87×10^6 cfu/g to 3.95×10^6 cfu/g; 2.20×10^6 cfu/g to 5.74×10^6 cfu/g and 2.42×10^6 cfu/g to 6.86×10^6 cfu/g for ogiri samples fermented for 24, 48 and 120 hours respectively. The total viable count of *P*. species present in sample packaged in glass jar (sample A) were found to be low in comparison to the other samples and this could be attributed to the non-permeable nature of the glass jar that created no room for more moisture and oxygen migrating into the sample. Thus, sample packed in glass jar, could be more shelf stable than those packaged in aluminum foil, uma leaves, nylon and plastic container. Pseudomonas is aerobic and thrives well in the presence of oxygen and water (Ukaoma *et al.*, 2018).

Proteus Species of Ogiri Samples

The *proteus* species viable count over a period of 24, 48 and 120 hours fermentation ranges from 2.47 x 10^6 cfu/g to 4.61 x 10^6 cfu/g; 2.77 x 10^6 cfu/g to 6.37 x 10^6 cfu/g and 3.03 x 10^6 cfu/g to 8.02 x 10^6 cfu/g respectively. The *P*. species count was found too high in sample wrapped in uwa leaves while sample packed in glass jar had the least total viable count from day 1 to day 5 fermentation periods. The results as shown in Table 4 revealed that the total viable count increased with increase in fermentation time. The detection and isolation of *P*. species in this study agrees with Ukaoma *et al.* (2018) that reported the presence of *P*. species in ogiri produced from egusi and its absence in castor oil seed ogiri.

Lactobacillus in Ogiri Samples

The results obtained revealed the total viable count of *Lactobacillus* present in the ogiri-egusi samples to be in the range of 2.75×10^6 cfu/g to 5.33×10^6 cfu/g; 3.15×10^6 cfu/g to 7.28×10^6 cfu/g and 3.70×10^6 cfu/g to 9.70×10^6 cfu/g for a fermentation period of 24, 48 and 120 hours respectively. *Lactobacillus* based on the figures presented in Table 4 were found to be more predominant in sample packaged with uma leaves (sample C) followed by

samples packaged in nylon (sample D), aluminum foil (sample B) and plastic container (sample E) while sample packaged in glass jar (sample A) had the least total viable count of *Lactobacillus*. Ukaoma *et al.* (2018) reported also the presence of *Lactobacillus* in ogiri produced from fermented melon seeds. Lactobacillus is dominant in fermentation of food within pH range of 3.5 - 4.5). It is also aerobic and therefore very active in the presence of oxygen and moisture.

Sensory Evaluation of Ogiri Samples

Taste of ogiri samples

The sensory score for taste of the ogiri samples ranges from 5.58 to 6.78. The highest score was recorded in sample B with value (6.78) followed by samples C with value (6.59), sample D (6.40), E with value (6.30) while sample A was rated the least (5.58). From the scores, the panelists liked the ogiri samples B, C, D and E slightly. There was no significant (p>0.05) difference among the ogiri samples C, D and E as well as between samples B and C but sample A differed significantly from the rest of the samples.

Aroma of ogiri samples

The scores for aroma of the ogiri samples showed that the scores varied from 6.22 to 7.13. The scores of the ogiri samples include 6.22, 7.10, 7.13, 6.88 and 6.79 for samples A, B, C, D and E respectively. The assessment showed that samples B (foil) and C (leaf) were liked moderately. The ogiri sample wrapped with leaf (sample C) scored the highest value of (7.13) while the lowest score (6.22) was recorded in the ogiri sample that was stored in a glass container (sample A). Statistically, there were no significant (p>0.05) difference among the ogiri samples except sample A. Aroma is one of the sensory attributes that make a product to be liked or disliked. Aroma is a very important food quality that "speaks" for the good quality of the food even before it is being consumed. In other words, it is a strong force that attracts consumers to the food (Hanani *et al.*, 2014).

Samples	Aroma	Taste	Appearance	Mouthfeel	Overall Acceptability
Α	5.58 ^c	6.22 ^b	7.11 ^a	6.58 ^b	6.00 ^d
В	6.78 ^a	7.10 ^a	6.90 ^a	7.00 ^a	7.11 ^a
С	6.59 ^{ab}	7.13 ^a	6.88 ^a	7.00 ^a	7.23 ^a
D	6.40 ^b	6.88 ^a	7.05 ^a	6.66 ^b	6.50 ^c
Е	6.30 ^b	6.79 ^a	7.10 ^a	6.59 ^b	6.78 ^b
LSD	0.33	0.40	0.27	0.30	0.28

 Table 5: Sensory evaluation of ogiri samples

Note: A = Ogiri sample packaged with glass, B = Ogiri sample packaged with foil, C = Ogiri sample packaged with leaf, D = Ogiri sample packaged with nylon, E = Ogiri sample packaged with plastic

Appearance of ogiri samples

From Table 5 the scores of the appearance of the ogiri samples fell within 6.88 to 7.11. The results indicate that there were no significant (p>0.05) difference among all the ogiri samples despite the variations in the scores. This implies that probably the packaging materials did not cause any difference in the appearance of the samples. The panelist saw the sample stored in a glass (sample A) appeasing hence it was scored the highest (7.11) while sample C (sample wrapped with leaf) was scored the lowest (6.88). Appearance is an important sensory attribute which consumers' preferences may depend on.

Mouthfeel of ogiri samples

The mouth feel of the ogiri samples ranged from 6.58 to 7.00. The sensory scores as presented in Table 5 showed that samples B and C had the highest score (7.00) in terms of mouth feel, followed by sample D (sample rapped with nylon), sample E (sample stored in plastic container) with scores of 6.66 and 6.59 respectively. While the least score (6.58) was

recorded in the sample stored in a glass container (sample A). The result indicates that the panelists liked samples B and C moderately.

Overall acceptability of ogiri samples

The overall acceptance score of the ogiri samples are 6.00, 7.11, 7.23, 6.50 and 6.78 for samples A, B, C, D and E respectively. The overall acceptability scores of the ogiri samples ranges from 6.00 to 7.23. The scores showed that sample C (sample wrapped with leaves) which had the highest score (7.23) was liked moderately. There were no significant (p>0.05) differences between ogiri samples B and C but the rest of the samples differed significantly (p<0.05) from each other. Sample A was given the least score of 6.00 which indicates it was liked slightly. Overall acceptability has to do with the consumer's acceptance of the product.

Conclusion

This study showed the variations in proximate, mineral and vitamin composition of the ogiri samples which are attributable to the different packaging materials. Sample packaged in *uma* leaves comparatively had higher nutrients (protein, fat, vit. A, Vit. B2, etc.) than those packaged in plastic, foil, glass and nylon. All fermentation microbes were found in the samples but the traditional *uma* leave supported the growth of microbes/fermentation more than others. The sensory evaluation revealed that the packaging materials did not cause any objectional quality in the ogiri but it is remarkable that the sample in *uma* leave deteriorated more than those in plastic, foil, glass and nylon which implies that glass, foil, plastic or nylon material can be used where delay in fermentation or extension of shelf life is needed. Aside preservation of the ogiri sample, glass and foil also proffer better quality of ogiri in terms of aroma, appearance and general acceptability. This obviously suggests that all the packaging materials in this study could serve well in the production and packaging of ogiri-egusi.

Recommendation

The use of glass, foil, plastic and nylon materials should be encouraged as well as the usual traditional *uma* leave. These results should therefore be disseminated to domestic and commercial producers of ogiri-egusi. Attempts should be made to produce the ogiri-egusi in dry form and study its nutritional, microbial and sensory qualities as well as the acceptability in comparison with the usual paste form.

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