Microbiological and nutritional quality of retail and laboratory “Ikpan” (mushroom -melon cake); a local snack

B Adebayo-Tayo, E Friday, B Adebayo-Tayo

Citation

B Adebayo-Tayo, E Friday, B Adebayo-Tayo. *Microbiological and nutritional quality of retail and laboratory “Ikpan” (mushroom -melon cake); a local snack*. The Internet Journal of Nutrition and Wellness. 2008 Volume 8 Number 2.

Abstract

Microbiological and nutritional quality of “Ikpan” (mushroom-melon cake), a traditional snack locally made from fermented sclerotium of P. tuber-regium and melon was investigated. The identified bacteria isolates from retail “Ikpan” was Streptococcus sp., Staphylococcus aureus, Bacillus subtilis, Bacillus sp., Micrococcus sp., Proteus sp. and Salmonella sp. The fungi isolates were A. glaucus, Aspergillus flavus, P. expansium, Aspergillus niger, Eutorium spp. and Absidia sp. Of the six species, A. glaucus were the dormant species. The crude protein ranged from 20.94 – 24.28% in which samples from Ikot Ekpene had the highest. Laboratory samples had the least protein. The crude fat, crude fibre ranged from 3.89- 4.56% and 0.73 -.33%. Statistical analysis of sensory evaluation results showed a preference for the laboratory samples in terms of appearance, texture, flavour and palatability while retailed samples from Ikot Ekpene, and Uyo matched each other in terms of taste. The result shows that combination of melon and sclerotium of P.tuber-regium for production of cake or traditional snacks resulted in rich snacks that can enhance the nutritional status of the consumers.

INTRODUCTION

“Ikpan” (mushroom-melon cake) is a traditional snack locally made from fermented sclerotium of P. tuber-regium and melon. It originated from “Ikon”festival which is celebrated by Annang tribe in Akwa Ibom state, south southern Nigeria. The cakes are used during the festival to feed the gods and as celebrative cake. With the advent of Christianity the cake are been used during Christmas and New Year season. Ikon is widely consumed and used as traditional religious practices as well as celebrative cake by Ibos of the Imo and Anambra states of Nigeria. Mushrooms are widely consumed in Nigeria particularly in the rural areas (Oso, 1977). Pleurotus tuber-regium is a tropical sclerotial mushroom which is very common in Nigeria. The mushroom produces a sclerotium or the tuber as well as a mushroom. Both the sclerotium and the mushroom are edible. The sclerotium which is dark brown on the outside and white on the inside is spherical to ovoid and can be quite large. Pleurotus tuber-regium is useful as food and as medicinal. The slerotium which is hard can be peeled and ground for use in vegetable soup and may be dried for future use. The dried sclerotium is used as soup thickner by the Ibibio in the south eastern Nigeria. The dried sclerotium is called “Isuo” in Ibibio

Citrullus lanatus (egusi melon) is a member of the Cucurbitaceae family originated from West Africa. The juicy flesh is pale yellow or green, and also tastes bitter. Citrullus lanatus is a creeping annual herb with hairy stems, forked tendrils and three-lobed hairy leaves. Melon various species of Curcubitaceae seeds are used as main food ingridients and are enjoyed by many people in Africa. In Nigeria the sweet melon is mainly cultivated for its seeds, the oriental melon with elongated fruits is often consumed as vegetables while imature water melons are eaten raw and are occasionally used in preserves. The seeds can be roasted and eaten since they contain edible oil (Oyenuga, 1968, Purseglove, 1968, Tindall, 1983). Melon commonly known as “egunsi” in Yoruba and as “Ikon” in Ibibio is used as food ingridients, soup thickner and the melon seed can be fermented to produce “ogiri” (Oke 1965, Oyenuga and Feliga 1975). The potential of Curcubaceae seeds as sources of protein has been demostrated by Akpapunam and Markakis (1981) and it has a role to play in improving human diet.

This study was carried out to determine the miocrobial and nutritional quality of retailed and laboratory prepared snacks from mushroom and melon seeds.

MATERIALS AND METHODS

COLLECTION OF “IKPAN” SAMPLES

The Ikpan samples were purchase from four different

locations in Akwa Ibom state. The samples were collected using a sterile container and then transported to the laboratory for analysis.

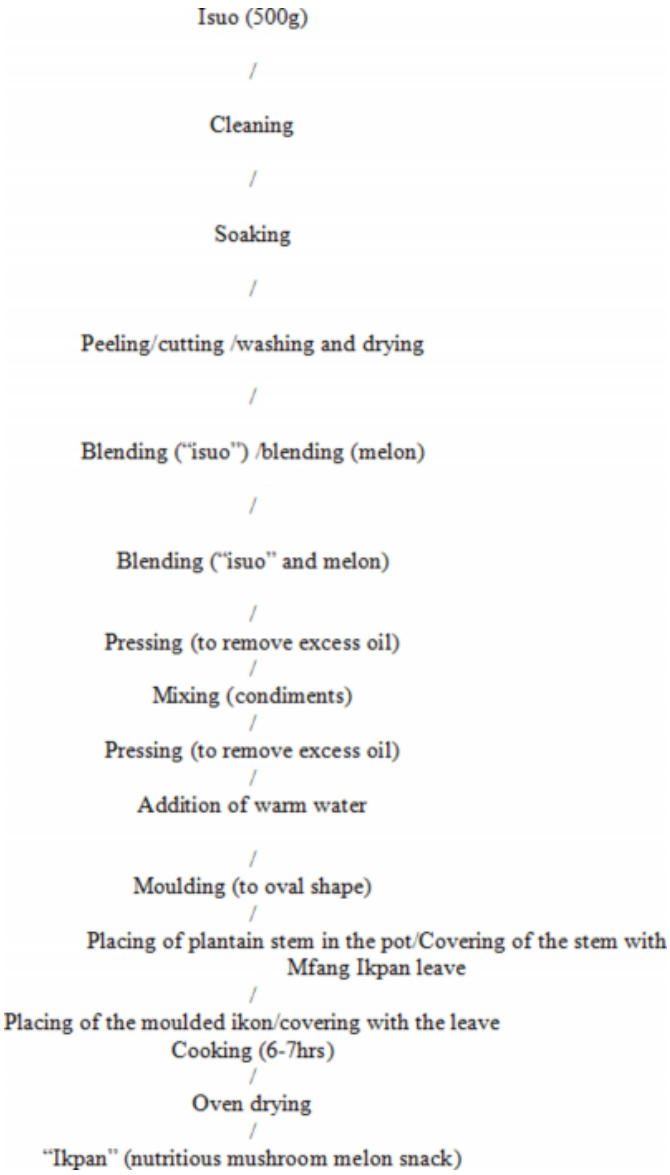
LABORATORY PRODUCTION OF "IKPAN"

Egusi melon seeds used for this study were obtained from a local market in Uyo, Akwa Ibom State, Nigeria and were identified as Citrullus Ianatus by a taxonomist in the Department of Botany, Faculty of Science, University of Uyo, Akwa Ibom, Nigeria. Seeds were screened to remove bad ones, shelled manually and further screened. The seed were put in an air-tight container and stored in desiccators for further analysis.

Five hundred grams of "Isuo" (dried sclerotium of Pleurotus tuberegium) was washed and soaked in water for 1 day, it was removed, peeled, washed and cut into pieces and was ovendried. The dried fermented "Isuo" and 200gm of melon (Citrullus Ianatus) was ground using mechanical blender. The ground "isuo" and melon were then mixed together. Excess oil was removed by pressing after which the condiments were added (ground onions, pepper, star magi, and salt) and mixed together. The mixture was mixed to a desire consistency by adding warm water. The mixtures were moulded into oval shape, water was brought to boil while the moulding was on and plantain stem are cut and placed on the bottom of the pot. "Mfang Ikpan" leaf (colour enhancer) was placed carefully on the stem and moulded cake was placed carefully on the leave. The boiled water was added into the pot and the leave was spread on the melon cake. The pot was covered and allowed to cook for 6-8hrs but water was added at intervals to avoid dryness. The cooked cakes which are brownish in colour was removed and ovendried and stored in a clean container for futher analysis.

Figure 1

Fig.1. Flow chart for production of "Ikpan" (Mushroom melon cake)



MICROBIOLOGICAL AND PHYSICOCHEMICAL ANALYSES

One gram of retail"Ikpan" samplewas weighed into 90ml sterile 0.1% peptone water as described in the Bacteriological Analytical Manual (FDA, 1991). Ten fold dilutions of each of the samples were made and 0.1ml of the diluents were pour plated in triplicate plates on Nutrient agar for total bacteria counts, MacConkey agar (oxoid) for coliform count, Salmonella/Shigella agar for Salmonella/Shigella counts, chocolate agar for Staphyloccoci count, thioxycolate citrate bile salt agar for vibrio count and Sabourad dextrose agar with chloramphenicol (250mg/100ml) for fungal counts. All

plates were incubated for 48hrs at 30C except Sabouraud Dextrose agar that were incubated at 26C for 6 days.

Colonies were selected randomly, bacteria cultures were characterized and identified using various morphological and biological test such as gram stain, spore stain, motility, catalase, coagulase, indole, MR –VP, urease, citrate, Oxidase and sugar fermentation. Pure cultures of each isolate were obtained by streaking the specific colonies on suitable media and incubated appropriately; these were maintained in an agar slant in McCartney bottles.

The identification of the microbial isolates was based on classification Scheme proposed by Harrigan and McCance (1976), Buchanan and Gibbson (1974) and Collin and Lyne (1984). The identification was based essentially on morphological and biochemical reactions. The associated fungi were then identified with reference to Frazier and Westholf (1998).

The proximate analysis of the samples for moisture, total ash and crude fibre were carried out in triplicate using methods described (AOAC, 1990). The nitrogen was determined by micro Kjeldahl method described by Pearson (1976) and the nitrogen content was converted to protein by multiplying by a factor of 6.25. Determination of crude fat/lipid content of the samples was done using Soxhlet (Cehmglass) type of the direct solvent extraction using petroleum ether (boiling range 60-80 ° C) as solvent. At the end of the extraction, the solvent was evaporated and the flask dried in the oven (at 60 ° C). Total carbohydrate content was estimated by 'difference'. All the proximate values were reported in percentage (%).

SENSORY EVALUATION OF THE "IKPAN" SAMPLE

Coded samples of "Ikpan" were served to 10 semi trained panelists. The panelists were asked to rate the samples for appearance, flavor, texture and overall acceptability. The ratings were presented on a 9-point Hedonic scale ranging from 9 = "like extremely" to 1 = "dislike extremely". Result obtained was subjected to analysis of variance using one – way ANOVA. Differences between means were separated using Duncan's Multiple Range Test (Steel and Torrie, 1980, Duncan, 1955).

RESULTS AND DISCUSSION

The microbial counts of retailed and laboratory "Ikpan" samples are shown in Table 1.

Figure 2

Table1: Total count of microbial groups in the"Ikpan" samples (cfu/g)

Source	Sample	Total bacteria count ($x10^3$ cfu/g	x 10^2 Coliform cont $x10^2$cfu/g	*Salmonella/Shigella* count $x10^1$cfu/g	Vibrio count	Fungal count x 10^2	Staphylococci countx10^2
Ikot Ekpene park	IK_1	6.3	N.G	3.2	NG	3.6	1.8
	IK_2	6.0	N.G	3.0		3.4	1.0
Uyo main park	UYO_1	5.2	N.G	2.8	NG	4.3	1.0
	UYO_2	5.0	N.G	2.3		6.1	NG
Itam market park	ITA1	4.7	NG	2.6	NG	5.4	$<10^2$
	ITA_2	3.2	NG	2.4		4.9	
Laboratory sample	LAB_1	1.2	NG	NG	NG	2.8	NG
	LAB_2	1.0	NG	NG	NG	1.3	NG

The result showed that virtually all the retailed and laboratory "Ikpan" samples did not contain vibrio and coliform. The total bacteria count ranged from 1.0 - 6.2 $x10^2$ cfu/g in which the lowest count was obtained from samples prepared in the laboratory and samples from Ikot Ekpene market had the highest. The Staphylococci count ranged from 4.8 -5.0$x10^2$cfu/g in which samples from Itam had the highest while there was no observable growth in laboratory samples. Salmonella/Shigella count ranged from 2.6 – 3.2 x 10^1 cfu/g in which the highest was obtained from Ikot Ekpene samples. The fungal count ranged from 1.6 x 10^2 – 2.1 x 10^2 cfu/g.

The microorganism associated with "Ikpan" samples from different location is shown in Table 2.

Figure 3

Table 2: Microorganism associated with Retailed and Laboratory "Ikpan"

location	Sample	*S.aureus*	*Proteus sp.*	*Micro cocci sp*	*B. subtilis*	*B.sp*	*Salmonella sp.*	*Streptococcus sp*	*Aspergillis flavus*	*A - glaucus*	*Aspergillus niger*	*P-expansum*	*Absidia sp.*	*Eukrium sp.*	*Monilla sp*
Ikot Ekpene park	IK_1	-	-	-	+	-	-	-	+	+	+	-	-	-	+
	IK2	-	-	+	+	+	-	-	-	+	+	-	-	-	-
Uyo main park	UY O1	+	+	-	+	-	-	+	+	+	-	-	-	-	-
	$UY O_2$	+	-	-	+	-	+	-	-	+	+	+	-	-	-
Itam park	ITA_1	+	+	-	+	+	-	-	+	+	-	-	+	-	-
	ITA_2	-	+	-	+	-	-	-	-	+	-	-	-	+	+
Laboratory samples	LAB 2	-	-	-	+	-	-	-	-	-	-	-	-	-	-
	LAB 2	-	-	-	+	-	-	-	-	+	-	-	-	-	-

A- Ikot Ekpene sample, B- Itam sample, C- Uyo sample and D- Laboratory sample,

The associated bacterial were Streptococcus sp., Staphylococcus aureus, Bacillus subtilis, Bacillus sp., Micrococcus sp., Proteus sp. and Salmonella sp. The fungi isolates were A. glaucus, Aspergillus flavus, P. expansium, Aspergillus niger, Eutorium spp. and Absidia sp. Of the six species, A. glaucus were the dormant species

The frequency of occurrence of bacteria and fungi isolates is shown in figure 1 and 2.

Figure 4

Figure 1: Frequency of occurrence (%) of bacterial associated with Retailed and Laboratory "Ikpan".

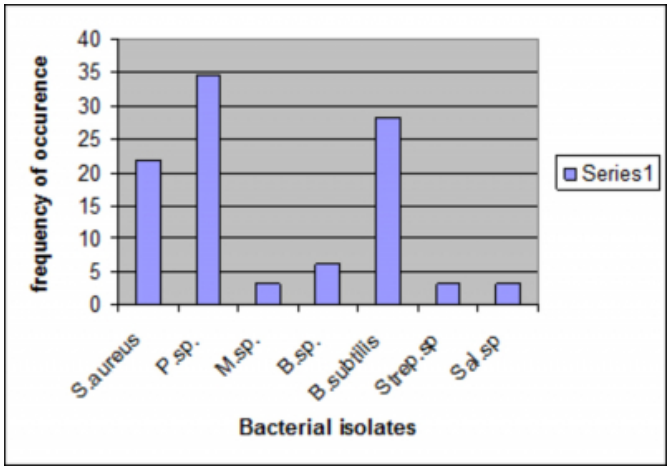
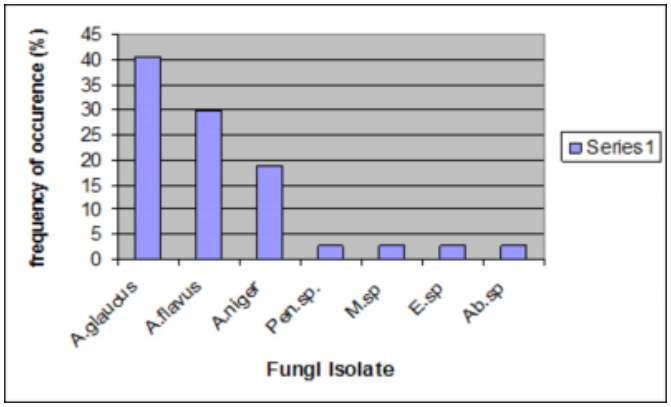


Figure 5

Figure 2: Frequency of occurrence (%) of fungi associated with Retailed and Laboratory "Ikpan".



Staphylococuss and Micrococcus species were possible contaminants from handlers. Staphylococcus aureus, a mesophile have been implicated in food poisoning outbreak of some food material. Odunfa (1988) reported that Staphylococcus aureus levels of 10^8ml are considered potential hazardous to consumers. The presence of Staphylococcus aureus is an indication of contamination by food handlers. 80% of them are being harbored by man as normal micro flora

Generally "Ikpan" are displayed in wired basket for prospective consumers and in the process exposed to microbial contamination. Most of the organisms isolated have health implications for man except Micrococcus sp. which have not been associated with human infections. It has occasionally been isolated from human clinical specimen where it visually represents contaminants from the skin or mucous membrane surfaces or from environment. (Koreman et al., 1992).

The occurrences of Bacillus sp. can be said to be as a result of prevalence of their spores in environment (Jay, 1978). Bacillus species are spore formers whose spores could survive high temperatures of processing.

Bacillus subtilis has been isolated from other food condiments such as Iru (Parkia biglobosa) and Ogiri (Citrullus Ianatus)

Occurrences of Steptococcus sp. in retailed "Ikpan" may be as a result of bad habit of the handlers of the "Ikpan", such as sneezing and coughing without covering their mouth during production and handling (Hobbs and Gilbert, 1978).

The fungi found associated with "Ikpan" mainly species of Aspergillus. This could be attributed to the prevalence of their spores in the atmosphere. This organism was easily trapped during handling of "Ikpan". Since most fungal spores are found in the air, the spores must have contaminated the "Ikpan" sample during processing and handling. The liberated spore can easily settle on food and ceilings of room and then germinated (Okhuoya and Ayanlola (1986). Dongo and Ayodele (1997) have shown that Aspergillus occurred highest in the number of colonies identified from air spora of some localities.

From the result of this study, it has been made clear that most of the toxigenic fungi isolated from the Ikpan samples may be as a result of contamination of the melon and dried sclerotium and from improper handling during processing.

The proximate composition of "Ikpan were showed in Table 3.

The crude protein ranged from 20.94 – 24.28% in which samples from Ikot Ekpene had the highest while laboratory samples had the least protein. It was observed from these results that the protein value compares favourably with those

of protein rich foods such as soybean, cowpeas, pigeon peas and pumpkin with protein contents ranging between 23.1 and 33.0% (Olaofe et al., 1994). This protein value also falls within the recommended daily allowance for children (23.0 – 36.0 g) (NRC, 1989). Many plant proteins usually in the form of protein extracts or seed flours are being investigated and tested for new products such as low cost fabricated foods which are nutritious, attractive and acceptable to consumers just like conventional foods from meat, fish and dairy products (Lawhom and Cater, 1971; Lin et al., 1974; McWalters et al., 1976).

Figure 6

Table 3: Proximate composition of retailed and laboratory "Ikpan"

Proximate composition (%)	A	B	C	D
Protein	24.28	21.34	21.52	20.94
Fat	3.56	3.89	3.95	4.56
Carbonhydrate	66.58	66.38	68.03	62.45
Fiber	0.87	0.92	0.77	1.13
Ash	7.45	7.47	6.34	7.28
Moisture	60.10	59.15	53.98	61.38

A- Ikot Ekpene sample, B- Itam sample, C- Uyo sample and D- Laboratory sample,

Seeds have nutritive and calorific values, which make them necessary in diets. Research attention that are geared towards increasing utilization of plant protein sources for food use includes pumpkin (Olaofe et al., 1994), peanut (Khan et al., 1975), pigeon pea (Oshodi and Ekperigin, 1989), African yam bean (Adeyeye et al., 1994), and akee apple (Akintayo et al., 2002). Ojieh et al (2008) reported similar result from Citrullus lanatus.

The crude fat ranged from 3.89- 4.56%. The crude fat content obtained in this study is lower than that obtained from melon reported by other workers (Ige et al., 1984, Fagbemi and Oshodi, 1991) from variety of melon oil seeds which ranged between 47.9 and 51.1%. The crude fat content of 45.7% was reported by Ojieh et al (2008) from Citrullus lanatus (egusi melon). The lower crude fat content may be as a result of pressing during processing of Ikpan. Despite the fact that melon could be regarded as an oil seed but combination of melon and sclerotium of P.tuberegium together with the method of production, Ikpan can be regarded as an excellent diet for human consumption.

The crude fibre ranged from 0.73-1.33%. The crude fibre content of Ikpan obtained in this study is low compared to those of melon (12.0%) (Ojieh et al (2008) and legumes (5.0 - 6.0%) (Aremu et al., 2006). The result shows that combination of melon and sclerotium of P.tuber-regium for production of cake or traditional snacks resulted in rich snacks that can enhance the nutritional status of the consumers.

As observed from our result, Ikpan is high in carbohydrate (62.45 – 68.56%) compared to other legumes which have as high as 20.0-60.0% carbohydrate content (Arkroyed and Doughty, 1964) and in contrast to egusi melon which is low in carbohydrate (10.6%)( )

The ash content of Ikpan samples ranged from 6.34 – 7.47% which is higher than the one reported by Ojieh et al (2008) obtained from egunsi melon (3.7%). The range obtained in this study is above the range of 1.5 – 2.5% recommended for seeds and tubers for animal feed formulation by Pomeranz and Clifton (1981). On this basis, mushroom –melon cake could be considered not to be suitable for animal feeds.

Table 4 shows the sensory attribute of the samples. Statistical analysis of sensory evaluation results showed a preference for the laboratory samples in terms of appearance, texture, flavour and palatability while retailed samples from Ikot Ekpene, and Uyo matched each other in terms of taste. Uyo match Itam samples only in texture.

Figure 7

Table 4: Sensory scores of retailed and laboratory "Ikpan"

Sample	Appearance	Texture	Flavor	Taste	palatability
A	7.55^{b}	6.11^{b}	6.88^{c}	7.40^{a}	7.22^{b}
B	6.55^{c}	6.00^{c}	6.80^{d}	7.41^{a}	7.10^{c}
C	7.50^{b}	6.00^{c}	7.30^{b}	7.00^{b}	7.00^{d}
D	7.77^{a}	7.77^{a}	7.44^{a}	6.80^{c}	8.55^{a}

A- Ikot Ekpene sample, B- Itam sample, C- Uyo sample and D- Laboratory sample,

Conclusively, the present study revealed that retailed Ikpan from different location in Akwa Ibom though nutritionally rich contain some microorganisms which may be hazardous to the consumers, prolong intake of which can constitute a health risk and significantly reduce net population growth rate. The study on the whole evidenced the microbial,

nutritional and sensory status of Ikpan from Akwa Ibom state, However it did bring out the probable hazard associated with the consumption of contaminated Ikpan and equally revelealed that Ikpan is an excellent dietary snack which is very rich in required nutrient for human growth and development. It is recommended that Government should organize seminar for the producers, retailers and consumers and enlighting or educate them on the need to improve their personnal hygiene and proper preservation and handling after processing and how to display their food after processing by using glass showcase instead of metal basket. The consumers should be enlighting not buy or eat cake that are displayed inside a metal basket or has been exposed for too long.

It can thus be concluded that nutritionally rich diet could be prepared from combination of melon and sclerotium of P.tuber-regium for snacks production. Moreover the cake will be suitable in the total amelioration of protein energy malnutrition (PEM) in the developing countries

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Author Information

Bukola C. Adebayo-Tayo, Ph.D.
Department of Botany & Microbiology, Faculty of Science, University of Ibadan

E. E. Friday, BSc.
Department of Microbiology, University of Uyo

B.C. Adebayo-Tayo
Department of Botany and Microbiology, University of Ibadan