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Mycoflora and Nutritional Analysis of Smoked Dried Crayfish (*Penaeus monodon* – Prawns) During Storage

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Authors' contributions

This work was carried out in collaboration among all authors. Author EDF designed the study and wrote the protocol, Author OAP performed the statistical analysis. Authors EDF, OAP and AOD wrote the first draft of the manuscript. Author AOD managed the analyses of the study. Authors OAP and AOD managed the literature searches. All authors read and approved the final manuscript.

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ABSTRACT

This study was carried out to investigate the mycoflora and nutritional composition of smoked dried crayfish *Penaeus monodon* (prawns) during storage for twenty-four weeks. The mycoflora were isolated at four weeks interval using direct plating and dilution methods on Potato Dextrose Agar (PDA), Saboraud Dextrose Agar (SDA) and Malt Extract Agar (MEA). The fungi isolated using direct plating methods and dilution methods were *Aspergillus niger, Aspergillus flavus, Aspergillus fumigates, Rhizopus* sp*., Phytophthora siskiyouensis, Penicillum* sp. and *Mucor* sp. The result of proximate analysis (g/100 g) of smoked dried crayfish *Penaeus monodon* (prawns) showed a decrease in ash content (12.53-10.86), fat (14.95-12.30), crude fibre (1.60-1.29) while moisture content (3.10-3.71), crude protein (66.34-66.84) and carbohydrate (1.66-5.00) increased respectively. The result of mineral analysis (mg/100 g) of smoked dried crayfish *Penaeus monodon*

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(prawns) showed a decrease in Sodium (110.90-104.9), Potassium (107.30-94.96), Calcium (120.61-98.66), Magnesium (137.50-120.22), Zinc (2.15-1.87), Iron (12.33-10.17), Copper (0.16-0.22), Manganese (0.40-0.25), cadmium (0.42-0.13) and Phosphorous (485.00-460.76) respectively. This study showed that the smoked dried crayfish products were invaded by fungi which could be due to display of the products in open trays without coverage for sale, most of the times which were not hygienic. This, in turn, allows the dust and fungal spores to settle on the products leading to fungal contamination, production of toxins and spoilage. Stored smoked dried crayfish (prawns) sellers should be enlightened on good hygienic practices.

Keywords: Mycoflora; storage; proximate; minerals; Penaeus monodon (Prawns).

1. INTRODUCTION

The Asian tiger shrimp, Penaeus monodon, is a widespread penaeid shrimp species that is native to the Indo-West Pacific with a range comprising southern Japan, Korea, China, Taiwan, the Philippines, Vietnam, Cambodia, Malaysia, Singapore, Indonesia, Papua New Guinea, Australia, Thailand, Myanmar, Bangladesh, Sri Lanka, India, Pakistan, Tanzania, Madagascar, South Africa, and the Red Sea of Yemen [1]. Crayfish is used to a large extent in local food preparations in Nigeria, the nutritional benefits of crayfish were reported by Ibironke, et al. [2] as used in complementary food formulations. Crayfish are classified as an animal polypeptide consisting of about 36-45% protein [3]. Like most seafood, it contributes immensely to the nutrition of consumers [4]. The protein is relatively cheaper than other animal protein and possesses high nutritional value Many Nigerian riverine regions sources their livelihood from the marketing of seafoods such as smoked-dried crayfish or dried fish. The commodity is processed, stored and packaged in woven polythene or hessian bags or woven baskets and transported in dugout wooden boats from processing centres in creeks to onshore markets [5].

Preservation of crayfish is either by salting, freezing, canning, sun-drying or smoke-drying. Sun and smoke-drying are common preservation methods because of their relatively lower costs. The use of smoke in local fish preservation was earlier reported by Eyo [6]. However, there is a gap of knowledge on the impact of traditional smoke drying and handling of crayfish in Nigeria. Crayfish has the potential for export to market in countries with a high population of people from the producing countries. Such cross-country trade demands high consideration for quality and safety [7]. Information on the duration of effectiveness of smoke drying on crayfish quality is scarce; hence this study is aimed at evaluating the changes in mineral composition, proximate analysis and the mycoflora of smoked dried crayfish *Penaeus monodon* (Prawns) under storage for twenty-four weeks.

2. MATERIALS AND METHODS

2.1 Collection of Samples

Crayfish samples namely Prawns (*Penaeus monodon*) were purchased at Igbokoda, Ilaje Local Government Market, Ondo State, Nigeria. The dried crayfish samples were stored in a sterile airtight container, clearly-labelled and kept in a well-ventilated laboratory for 6 months in the Department of Microbiology, Ekiti State University, Ado Ekiti, Nigeria. The samples were identified at the Department of Zoology, Ekiti State University, Ado Ekiti.

2.2 Isolation of Mycoflora from the Stored Smoked Dried Crayfish *Penaeus monodon* (Prawns)

The mycoflora associated with smoked dried crayfish during storage were isolated using the following methods:

2.2.1 Direct plating method

The sundried crayfish samples were examined randomly for the presence of moulds according to the method of Amusa [8]. The surfaces of the two samples were sterilized separately with ethanol and washed in two changes of sterile distilled water. Using a sterile spatula, the sterilized samples were each aseptically placed on PDA, SDA, MEA plates and incubated at 28°C for 2-5days. The hyphae tips of each fungal growth were successively sub-cultured on freshly prepared Potato Dextrose Agar (PDA), Saboraud Dextrose Agar (SDA) and Malt Extract Agar (MEA) plates until axenic colonies were obtained for each sample [9]. The cultures were examined under the microscope to

determine the hyphae, sporangium and other fruiting bodies of the fungi present.

2.2.2 Dilution plate method

The sundried crayfish samples were examined randomly for the presence of moulds according to the method of Amusa [8]. The surfaces of the samples were sterilized separately with ethanol and washed in two changes of sterile distilled water. Using a sterile spatula, 10 grams of the sample was ground into a fine powder, and one gram was weighed. This was dissolved in 9ml of distilled water. One ml each of the standardized sample was pipette into 9ml of sterile distilled water in a test tube and serially diluted in a series of test tubes containing sterile distilled water. One ml each of aliquots of 10^{-2} and 10^{-3} was introduced into molten Potato Dextrose Agar (PDA), Saboraud Dextrose Agar (SDA) and Malt Extract Agar (MEA) plates in duplicates for each isolate. The fungal colonies were observed every 24hours until they started to merge. Successful hyphae transfer of the culture were made until pure cultures were obtained. The cultures were examined under the microscope to determine the fungi present.

2.3 Identification of Mycoflora

The isolated fungi were identified by their cultural and morphological features. The isolates were examined under bright daylight for colour of the culture and further examination was carried out using needle mount preparation method as described by Tuite [10], Crowley, et al. [11] and Fagbohun, et al. [9] and slide culture technique method as described by Fagbohun, et al. [9].

2.4 Nutrient Analysis

2.4.1 Proximate analysis

The proximate analyses for the stored dried smoked crayfish were determined according to the methods of Pearson [12] and AOAC. [11] for ash, crude fibre, moisture and fat. The nitrogen was determined by Micro-Kjeldahl method as described by Pearson [12] and the percentage nitrogen was converted to crude protein by multiplying 6.25. The carbohydrate content was estimated by the difference in the value obtained when all the chemical composition values were subtracted from 100%. All determinations were in triplicates and the values of each constituent were expressed in percentage.

2.4.2 Mineral analysis

The minerals of the crayfish samples were analyzed using the solution obtained by dry ashing the sample at 550°C and dissolving it in 10% HCL (25ml) and 5% lanthanum chloride (2ml), boiling, filtering and making up to standard volume with deionized water. Mn, Cu, Co, Zn, Fe, Mg, Na, and Ca were determined with a Buck Atomic Absorption Spectrometer (Buck Scientific, Model 200A/200, Inc. East Norwalk, Connecticut, U.S.A). Sodium was measured with a Corning 405 flame photometer (Corning Halstead, Essex, UK, Model 405) (AOAC) [13]. The detection limits had precisely been determined using the methods of Varian Techtron [14] as Mn 0.01, Cu 0.005, Co 0.05, Zn 0.005, Fe 0.02, Mg 0.002, Ca 0.04, Na 0.001, ppm (all for aqueous solutions). The optimum analytical range was 0.5 to 10 absorbance units with a coefficient of variation of 0.05-0.40%. Phosphovanadomolybdate method using a spectronic 20 colourimeter (Galenkamp, London, UK) (AOAC) [13]. All chemicals were BDH analytical grade

3. RESULTS AND DISCUSSION

The results of the proximate analysis of smoked dried crayfish *Penaeus monodon* (prawn) during twenty-four weeks storage is shown in Table 1.

The results of the mineral analysis of smoked dried crayfish *Penaeus monodon* (prawn) during twenty-four weeks storage is shown in Table 2.

The results of the mycoflora isolated from smoked dried crayfish *Penaeus monodon* (prawn) during twenty-four weeks storage is shown in Table 3.

3.1 Proximate Analysis

The proximate analysis of smoked dried crayfish *Penaeus monodon* (prawns) stored for 24 weeks showed a decrease in ash content (12.51-10.86 g/100 g), fat (14.95-12.30 g/100 g), crude fibre (1.60-1.29 g/100 g) and an increase in moisture content (3.10-3.71 g/100 g), crude protein (66.34-66.84 g/100 g) and carbohydrate (1.66-5.00 g/100 g). This result is similar to the findings of Fagbohun and Oluwaniyi [15] who reported a decrease in ash content of *Oryza sativa* “rice” from (4.51-4.25) mg/100 mg but the result is different from the findings of Faleye and Fagbohun [16] who reported an increase in ash content of “Tinco” dried meat from (2.05-3.07) mg/100 g. Ash content in food gives the number of mineral elements present in a sample and a

decrease in ash content indicates loss of nutrients as the storage progressed [17]. However, this result's ash content is low when compared to the standard of RDA for ash which is 48 g for children, 63 g for males, 50 g for females [18]. It could be deduced that smoked dried crayfish (prawn) is not a good source of ash. Increase in the carbohydrate content of this study is in agreement with the findings of Faleye and Fagbohun [16]. The conditions that favour fungal activity could lead to an increase in carbohydrate content of the stored product [8]. Therefore, smoked dried crayfish (prawn) is not a good source of carbohydrate. The significant increase in the moisture content of smoked dried crayfish, *Penaeus monodon* (prawn) during storage is similar to the work of Fagbohun, et al. [19], who reported an increase in the moisture content of stored sun-dried chips from (6.80-8.34) mg\100 g. Moisture content is a widely used parameter in the processing and testing of food. It is an index of water activity of many foods and determines the shelf life or keeping quality of the food Adepoju, et al. [20]. The increase in the moisture content may be due to an increase in the atmospheric humidity which favours the growth and multiplication of fungi [17]. The increase in the protein content in this study is in agreement with the findings of Oladejo and Adebayo-Tayo [19] who reported an increase in crude protein (21.68-54.16) mg/100 g of "Banda" dried meat during storage. However, the result of this study shows that protein content in smoked dried crayfish (prawn) is high compared to the RDA standard. RDA for protein is 56g/day for men and 46g/day for women. Therefore, smoked dried crayfish (prawn) is a good source of protein.

3.2 Mineral Analysis

The mineral analysis of smoked dried crayfish (prawns) stored for 24 weeks showed a decrease in Sodium (110.90-104.99 mg/100 g), Potassium (107.30-94.96 mg/100 g), Calcium (120.61-98.66 mg/100 g), Magnesium (137.50-120.22 mg/100 g), Zinc (2.15-1.87 mg/100 g), Iron (12.33-10.17 mg/100 g), Copper (0.16-0.22 mg/100 g), Manganese (0.40-0.25 mg/100 g), cadmium (0.42-0.13 mg/100 g) and Phosphorous (485.00-460.76 mg/100 g). The result of this work is in agreement with that of Fagbohun, et al. [19], who reported a decrease in Calcium from (0.59-0.24) mg/100 g present in stored melon seeds. Calcium plays a part in muscle contraction and relaxation, blood clotting, synaptic transmission and absorption of vitamin B_{12} [21]. However, the result of this study showed that the calcium content in smoked dried crayfish (prawn) was low when compared with the RDA value for calcium which ranges from 600-1400 g [22].Therefore, smoked dried crayfish (prawn) is not a good source of calcium. The significant decrease in iron (Fe) from (12.33-10.71) mg/100 g during six months storage is in agreement with the work of Fagbohun, et al. [19] who recorded a decrease in iron content of stored melon seeds from (1.11-1.10) mg/100 g. Iron is also very important in the formation of haemoglobin in red blood cells and deficiency of iron leads to anaemia [22] However, the iron content of smoked dried crayfish (prawn) in this study is higher than RDA standard of 8 g [23]. Therefore, smoked dried crayfish (prawn) is a good source of iron. The decrease in value of Phosphorus in smoked dried crayfish (prawn)is similar to that of Fagbohun, et al. [17] who reported a decrease in phosphorus content of stored sun-dried soya beans during 20 weeks of storage (586.12-560.03) mg/100 g. The reduction may be due to deterioration caused by the fungi since fungi require some essential nutrient for growth and survival. Phosphorus is needed for DNA and RNA synthesis. However, the result of this study shows that phosphorus content is low compared to the RDA standard of 700 mg. Therefore, smoked dried crayfish (prawn) is not a good source of phosphorus. The significant decrease in the sodium content observed in this study agrees with the work of Fagbohun, et al. [19] who recorded a decrease in the sodium content of melon seed from (2.71-2.47) mg/100 g. Sodium is used in the transmission of nerves impulse and maintenance of the osmotic balance of the cell [22]. However, the result of this study shows that sodium is low when compared to the RDA standard which is 1500 mg [24]. Therefore, smoked dried crayfish (prawn) is not a good source of sodium. In this study, the amount of potassium content significantly decreased from during 24 weeks storage which is similar to the work done by Mensah, et al. [21] who reported a decrease in potassium content of "Kale" (7.03-4.08) mg/100 g. However, the result of this work is in contrast to the findings of Atanda, et al. [25] who reported an increase in potassium (57.01-60.3) mg/100 g of dry pepper and suggested that it might increase the risk of disease and mineral consumption. Thus, the potassium content of stored smoked dried crayfish (prawn) in this study was low when compared to the RDA standard which is 4700 mg [26]. Therefore, smoked dried crayfish (prawn) is not a good source of potassium.

Table 1. Results of Proximate analysis of smoked dried crayfish *Penaeus monodon* (prawn) during 24 weeks of storage (g/100g)

Weeks of Storage	Ash	MC	CP	FAT	CF	CHO
Fresh	12.51±0.03^{F}	3.10±0.23^{A}	66.34±0.03^{A}	14.95±0.07^{E}	1.60±0.28^{B}	1.66±0.17^{B}
4	12.49±0.21^{EF}	3.13±0.01^{A}	66.35±0.04^{A}	14.90±0.03^{E}	1.55±0.07^{AB}	1.56±0.02^{B}
8	12.45±0.01^{DE}	3.86±0.02^{C}	67.23±0.02^{C}	14.81±0.01^{D}	1.52±0.01^{AB}	0.14±0.04^{A}
12	12.41±0.01^{D}	3.97±0.01^{D}	67.34±0.01^{E}	14.75±0.01^{D}	1.49±0.02^{AB}	0.50±0.00^{A}
16	11.59± 0.01^{C}	3.99±0.01^{D}	67.29±0.01^{D}	13.90±0.02^{C}	1.39±0.01^{AB}	1.85±0.05^{C}
20	11.34±0.02^{B}	3.90±0.04^{C}	67.70±0.01^{F}	13.49±0.01^{B}	1.36±0.01^{AB}	2.24±0.08^{D}
24	10.86±0.01^{A}	3.71±0.01^{B}	66.84±0.01^{B}	12.30±0.03^{A}	1.29±0.01^{A}	5.00±0.02^{E}

MC: Moisture content, CP: Crude protein, CF: Crude Fiber, CHO: Carbohydrate. Means for each treatment with the same alphabet in each row are not significantly different at 5% level of significance (p< 0.05), while different alphabets in each row are significantly different at 5% level

Table 2. Results of Mineral analysis of smoked dried crayfish *Penaeus monodon* (prawn) during twenty-four weeks storage (mg/100g)

Weeks of storage	Na	K	Ca	Mg	Zn	Fe	CU	Mn	CD	P
Fresh	110.90±0.14^{C}	107.30±0.99^{E}	120.61±0.01^{F}	137.50±0.14^{E}	2.15±0.07^{D}	12.33±0.16^{C}	0.16±0.03^{E}	0.40±0.00^{C}	0.42±0.29^{B}	485±7.07^{D}
4	109.89±1.61^{BC}	106.63±0.12^{E}	120.53±0.01^{E}	137.14±0.35^{E}	2.12±0.01^{CDE}	12.22±0.02^{C}	0.14±0.01^{C}	0.39±0.02^{C}	0.29±0.03^{A}	486.77±0.02^{D}
8	115.22±0.01^{C}	107.12±0.01^{E}	121.23±0.01^{G}	137.37±0.30^{E}	2.22±0.01^{E}	12.52±0.54^{b}	0.12±0.01^{C}	0.44±0.01^{C}	0.24±0.02^{A}	486.84±0.06^{D}
12	112.97±0.03^{C}	98.66±0.01^{D}	119.28±0.04^{D}	132.74±0.09^{D}	2.19±0.02^{D}	11.59±0.02^{B}	0.09±0.01^{B}	0.41±0.01^{C}	0.19±0.01^{A}	475.89±0.01^{C}
16	110.19±0.03^{BC}	97.69±0.01^{C}	110.32±0.02^{C}	130.68±0.04^{C}	2.08±0.04^{BC}	11.49±0.01^{B}	0.04±0.01^{A}	0.33±0.04^{B}	0.15±0.01^{A}	470.21±0.10^{BC}
20	98.71± 0.21^{A}	88.74±0.01^{A}	96.84±0.02^{A}	128.86±0.01^{B}	2.01±0.01^{B}	10.65±0.06^{A}	0.03±0.01^{A}	0.26±0.01^{A}	0.11±0.02^{A}	466.87±0.02^{AB}
24	104.99± 7.23^{AB}	94.96±0.25^{B}	98.66±0.01^{B}	120.22±0.09^{A}	1.87±0.01^{A}	10.71±0.02^{A}	0.02±0.01^{A}	0.25±0.02^{A}	0.13±0.02^{A}	460.76±0.01^{A}

Na: Sodium, K: Potassium, Ca: Calcium, Mg: Magnesium, Zn: Zinc, Fe: Iron, Cu: Copper, Mn: Manganese, CD: Cadmium, P: Phosphorus. Means for each treatment with the same alphabet in each row are not significantly different at 5% level of significance (p< 0.05), while different alphabets in each row are significantly different at 5% level

Table 3. Mycoflora isolated from smoked dried crayfish *Penaeus monodon* (prawn) during twenty-four weeks storage (mg/100 g)

Mycoflora	Week of storage													
	0 1	0 2	4 1	4 2	8 1	8 2	12 1	12 2	16 1	16 2	20 1	20 2	24 1	24 2
A	+	+	+	+	+	+	+	+	+	-	-	-	-	-
B	-	-	+	+	+	+	-	-	-	-	-	-	-	-
C	-	-	-	-	+	+	+	+	+	+	-	-	-	-
D	-	+	+	+	-	-	-	-	-	-	-	-	-	-
E	-	-	-	-	-	-	+	+	+	+	+	+	-	-
F	-	-	-	-	-	-	-	-	+	+	-	-	-	-
G	-	-	-	-	-	-	-	-	-	-	+	+	+	+

A: Aspergillus niger, B: Aspergillus fumigatus, C: Aspergillus flavus, D: Rhizopus sp., E: Phytophthora siskiyouensis, F: Penicillium sp., G: Mucor sp. 1: Dilution method, 2: Direct plating method, (+): isolated, (-): not isolated

3.3 Mycoflora of Smoked Dried Crayfish *Penaeus monodon* (Prawn)

A total number of seven fungal species belonging to five different genera were isolated, namely *Aspergillus niger, Aspergillus fumigates, Aspergillus flavus, Rhizopus* sp*., Phytophthora siskiyouensis, Penicillum* sp*.,* and *Mucor* sp. from the smoked dried crayfish samples. This is similar to the work of Adebayo-Tayo, et al. [27] who reported the isolation of *Aspergillus flavus, Aspergillus tereus, Aspergillus fumigatus, Abisidia sp., Rhizopus sp., Aspergillus niger, Mucor* sp*., Cladosporum* sp*., Penicillium viridatus, Candida tropicalis* and *Fusarium moniliformis* from selected smoked fish from different markets sites in Uyo, Akwa Ibom state. Similarly, Hassan [26] also reported the isolation of several species of fungi belonging to the genera *Penicillium, Aspergillus, Fusarium, Rhizopus* and *Mucor* from smoked dried fish sold at the market place in Giza Governorate, Egypt. The occurrence of these fungal contaminations in smoked dried crayfish samples could be attributed to improper sanitation from the catching stage all through the processing stage [28,29] which could lead to vulnerability of the fish to fungal contamination.

4. CONCLUSION

This study showed that fungi had been implicated as contaminants of stored smoke-dried crayfish (prawns) during the twenty-four weeks of storage which led to a reduction in some of the nutritional contents of the prawns. Good sanitation and hygiene practices must be followed during processing procedures and handling to minimize contamination of the prawns by fungal spores which will germinate during storage of the smoked dried crayfish (prawns). Stored smoked dried crayfish (prawn) sellers, especially the illiterates should be enlightened about good hygienic practices during storage. Also, the results of this study showed that stored smoked dried crayfish (prawns) contained some essential elements like calcium, iron, the crude protein which is good for body cells and tissue replenishment.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. Fuller PL, Knott DM, Kingsley-Smith PR, Morris JA, Buckel CA, Hunter ME, Hartman LD. Invasion of Asian tiger shrimp, Penaeus monodon Fabricius, 1798, in the western north Atlantic and Gulf of Mexico. Aquatic Invasions. 2014;9(1):59-70.
2. Ibironke SI, Fashakin JB, Badmus OA. Nutritional evaluation of contemporary food developed from plant and animal protein sources. Nutrition of Food Science. 2013;42(2):111 – 120.
3. Ibironke SI, Joseph BF, Morakinyo MI. Nutritional quality of animal polypeptide (Crayfish) formulated into contemporary foods. American Journal of Food and Nutritional. 2014;2(3):39 – 42.
4. Israel Dorothy U, Inana Mandu E, Adindu Matthew N, Akande Samuel A. Mycoflora, proteolytic potential and quality implication of dried crayfish at Uyo Urban market, Uyo

Nigeria. Food Science and Quality Management. 2016;47:48 – 54.

5. Abou-Zaid AM, Mohammed ASE. Production and quality evaluation of nutrition, of high quality biscuits and potato puree tablets supplemented with crayfish (*Procombarus clarkia*) protein products. Journal of Applied Science Research. 2014;10(7):43-53.
6. Eyo AA. Fish Processing Technology in the Tropics, University of Ibadan Press, Ibadan Nigeria. 2000;165-168.
7. Israel DU, Inana ME, Adindu MN, Akande SA. Mycoflora, proteolytic potential and quality implication of dried crayfish at Uyo Urban Market, Uyo Nigeria. Food Science and Quality Management. 2016;47.
   Available:www.iiste.org
8. Amusa NA, Kehinde IA, Ashaye OA. Biodeterioration of the Africa star apple (*Artocarpus communis*) in storage and its effects on the nutrient composition. African Journal of Biotechnology. 2001;1(2):57-60.
9. Fagbohun ED, Anibijuwon I, Egbebi O, Lawal OU. Fungi associated with spoilage of dried cocoa beans during storage in Ekiti State, Nigeria. Journal of Microbial Biotechnology and Food Science. 2011;1(2):204-214.
10. Tuite J. Fungi isolated from unstored corn seed in Indian in 1956 - 1988. Plants Diseases Report. 1961;45:212-215.
11. Crowley N, Bradley JM, Darrell JH. Practical Bacteriology. Butterworth and Co., Ltd. London. 1969;164-168
12. Pearson DH. Chemical Analysis of Foods. Churchhill: London. 1976;335-336.
13. AOAC. Official Method of Analysis. 14th Ed. Association of Official Analytical Chemist, Washington DC; 2005.
14. Techtron V. Basic Atomic Absorption Spectroscopy: A Modern Introduction. Domican Press, Victoria: Australia. 1975;104–106.
15. Fagbohun ED, Oluwaniyi TT. Mycoflora, proximate composition and nutritional changes during the storage of *Oryza sativa.* Food Science and Quality Management. 2015;40:108-116.
16. Faleye OS, Fagbohun ED. Effects of storage on the proximate, mineral composition and mycoflora of “tinco” dried meat sold in Oshodi market, Lagos State, Nigeria. Global Journal of Bio-Science and Technology. 2012;1(1):54-58.
17. Burnett JH. Fundamentals of Mycology 2nd Edition, Edward Amold Publishers, Ltd. 2005; 673.
18. Onwuka GI. Food Analysis and Instrumentation; Theory and Practice. Naphthalic prints, Surulere, Lagos, Nigeria. 2005;219- 230.
19. Fagbohun ED, Lawal OU, Hassan OA. Effect of storage on the chemical composition and of melon seeds (*Citrullus vulgaris*). International Research Journal of Microbiology. 2011;2(8):310-.314.
20. Adepoju OT, Onasanya LO, Udoh CH. Comparative studies of nutrient composition of cocoyam (*Colocassia esculenta*) leaf with some green leafy vegetables. Nigerian Journal of Nutritional Science. 2006;27:40-43.
21. Mensah JK, Okoli RI, Ohaju-Oboto JO, Eifediyi K. Phytochemical, nutritional and medicinal properties of some leafy vegetable consumed by Edo People of Nigeria. African Journal Biotechnology. 2008 ;7:2304-2308.
22. Bolt GH, Bruggenwert WG. Solid Chemistry, Basic Elements. Elsevier Scientific Publishing Co., New York. 1978;145.
23. Dietary Reference Intake (DRIs). Recommended intake for individuals. Food and Nutritional Board, Institute of Medicine, National Academy Press: Washington DC. 2002;1322.
24. Dietary Reference Intake (DRIs). Recommended intakes for individuals. Food and Nutrition Board, Institute of Medicine, National Academy Press, Washington DC. 2004;45-78.
25. Atanda OO, Akano DA, Afolabi JF. Mycofloral of dry “tatase” pepper (*Capsicum annum* L.). Stored for sale in Ibadan markets. Lett. in Appl. Microbiol. 1990;10:35-37.
26. Dietary Reference Intake (DRIs). Recommended intake for individuals. Food and Nutritional Board, Institute of Medicine, National Academy Press: Washington DC. 2004;45-78.
27. Adebayo-Tayo BC, Onilude AA, Ukpe GP. Mycoflora of smoke-dried fishes sold in Uyo, Eastern Nigeria. World

Journal of Agricultural Sciences. 2008; 4(3): 346-350.

28. Olajuyigbe OO, Akande GR, Ezekiel CN, Ezekiel MO. Aflatoxigenic moulds and aflatoxin contamination of retailed fishery products in Lagos markets. Mycotoxicology. 2014;1:57-63.

29. Hassan AA, Hassan MA, El Shafei HM, El Ahl RM, Abd El-Dayem RH. Detection of aflatoxigenic moulds isolated from fish and their products and its public health significance. Nature and Science. 2011; 9(9):106-114.

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