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Effect of processing on the micronutrient and toxicants level in mackerel fish sold by street food vendors in Port Harcourt

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***Abstract**: This study investigates the level of some vitamins as well as heavy metals and poly aromatic hydrocarbons (PAH) in processed fish sold by street food vendors in Port Harcourt, Nigeria. The Atlantic Horse Mackerel fish (Trachurus trachurus) was processed in three different ways namely; Washed Fresh Fish (WFF), Roasted Fish without Spice (RFWoS) and Roasted Fish with Spice (RFWS). Ultraviolet spectrophotometer, gas chromatography (GC) and atomic absorption spectrophotometer (AAS) were used to evaluate the vitamins, poly aromatic hydrocarbons and heavy metals content respectively. The results showed that the vitamins content ranged from 11.68 - 14.69 mg/l; 4.78 - 10.32 mg/l and 0.63 - 0.98 mg/l for Vitamin B_6, Vitamin A and Vitamin E respectively. Vitamin B_6 and Vitamin E contents of RFWS decreased compared to the washed fresh fish (WFF). The total concentration of PAHs in RFWS (0.0734 ppm) was significantly ($p \leq 0.005$) higher compared to the WFF sample (0.0263 ppm). Flourene (0.0162 - 0.044 ppm), Benzo (K) fluoranthene (0.001 - 0.090 ppm) and other PAHs were present in varying concentration in all three samples studied. Heavy metals such as Zn (0.044 - 0.100 µg/ml), Cd (0.006 - 0.091 µg/ml) and Cu (0.045 - 0.087µg/ml) were higher in RFWoS and RFWS compared to the WFF sample where most of these metals were either not present or present but not in significant quantity. This study therefore shows that processing decreased the vitamins B_6 and E content of the fish while the toxic substances level increased. This is possibly due to the smoke, environmental exposure and the additives which served as spices. Prolonged consumption of these processed Mackerel fish may have negative health consequences.*

***Keywords:** Mackerel fish, Vitamins, PAHs, Heavy metals, Food vendors.*

I. Introduction

Atlantic Horse Mackerel or European Horse Mackerel (*Trachurus trachurus*) is specie of jack mackerel in the family of carangidae [1] It can be found in the North-eastern Atlantic from Iceland to Senegal including the Mediterranean and the black sea [2]. The Atlantic mackerel is rich in proteins, vitamins and minerals with little or no saturated fat [3,4]. It is an important commercial fish widely consumed, after processing, in Nigeria and West Africa sub region. The processing methods include; boiling, roasting, grilling, baking, pre-treatment with additives like salt and spices (garlic-*Allum sativum*, onion- *Allum cepa*, curry and knorr cube). The effect of different processing methods have been previously reported [4,5,6,7]. Most of these researchers reported loss of micronutrient during processing.

Processed street foods have been associated with toxic environmental substances such as poly aromatic hydrocarbons (PAHs) and heavy metals. The mechanism by which PAHs are found in food is based on incomplete combustion or thermal decomposition of the organic materials [8]. PAHs are produced in food when the fat from products such as fish, meat or chicken drips as oil unto hot coals, gauze or stones resulting in smoke which later is absorbed into the food sample [9]. Food samples can also be contaminated with toxic substance via environmental exposure, mainly during processing of coal tar, crude oil, natural gas, exhaust from automobiles etc. [10,11,12]. Kinze *et al*., [13] observed that some cooking methods such as roasting, barbecuing and smoking increases the level of PAHs in foods; while steaming and boiling barely introduces PAHs. It has also been reported that PAHs are produced in roasted foods based on the distance of food from the heat source [14] duration of roasting [15] and temperature used [16]. PAHs formed in processed foods are very well known eco-toxicants that are harmful to human and animal health as they bind to cellular macromolecules, including DNA, thereby causing errors in DNA replication leading to mutation that initiates carcinogenesis [9,17,18]. A number of PAHs have been found to have carcinogenic effects; while some others may act as synergists [9].

Processing methods can also influence the presence of heavy metals in foods. Heavy metals can be assimilated by humans through processed food, drinking water and air [19]. As a result of environmental pollution, some of the toxicants leach into underground water [20,21,22] which are often exploited for domestic use.

Additives (spices) such as knorr cube, garlic, onion and curry which are added to foods to improve flavour and taste have been shown to contain some mineral elements and PAHs. [17,23,24]. Abdullahi *et al*., [25] reported that the type of soil used for cultivation, fertilizers and source of water used for irrigation may introduce these toxic substances into some of these additives. Darko *et al.*, [24] also reported that aerial depositions during drying or during commercial milling can introduce heavy metals into the spices due to wear and tear of the machinery. This work therefore, is aimed at investigating the effect of processing (roasting with some additives/spices) on the vitamins content as well as heavy metals and poly aromatic hydrocarbon concentrations in the mackerel fish.

II. Materials And Methods

2.1 Sample Preparation

Mackerel fish (*Trachurus trachurus*) with length, 22 – 70 cm, and weight 200 – 450 g, was obtained from street food vendors located in mile 1 Diobu, Port Harcourt, Nigeria. The fish samples were divided into three different sets. The first set was cleaned and washed (Washed fresh fish-WFF), the second set was cleaned, washed and roasted without the addition of spices (Roasted fish without spices-RFWoS) while the third set was cleaned, washed and spiced with additives such as onion, garlic, ginger, knorr cube, salt and palm oil before roasting (Roasted fish with spice-RFWS). Roasting, which involves the use of charcoal as a source of heat, was gradually carried out for about 40 min with the fish samples being placed on wire gauze over the burning charcoal at various distances to prevent immediate charring. The roasted fish was collected after about 1 – 2 hours exposure still placed on the gauze but farther away from the burning coals together with the other two sets, they were wrapped in separate paper foil and taken to the laboratory for analysis. Prior to analysis, samples were oven dried at 50^0C for 18 hours, ground into powder using laboratory blender and stored in an air tight container until ready for analysis.

2.2 Vitamin Analysis

2.2.1 Vitamin B_6

Modified method by Kaman *et al*., [26], using high performance liquid chromatography (HPLC) was applied for the determination of vitamin B_6. Each of the dry powdered samples was weighed (5g) into a 250ml conical flask; followed by the addition of 30ml of 0.1M HCl. The flask was closed with cotton and wrapped with aluminium foil before placing it in an autoclave for about 35 minutes. pH of the autoclave sample was adjusted to 6.5 with sodium acetate and to 4.5 using HCl. The volume was made up with distilled water and filtered using normal filter paper. The filtered sample was further centrifuged for 10 minutes at 6000 rpm to eliminate the presence of turbidity. The sample was finally placed in the HPLC column for Vitamin B_6 estimation.

2.2.2 Vitamins A and E

The fat soluble Vitamins A and E were determined using HPLC as described by Manz and Philip[27]. The samples were homogenized and later saponified using 50% ethanolic potassium hydroxide and 50% methanolic ascorbic acid in addition to 50% potassium hydroxide for Vitamin A and E respectively. Saponified samples were extracted three times using 70 ml diethyl ether and separated using separating funnel. Each of the three extracts for Vitamin A and Vitamin E was combined and washed with distilled water before being concentrated and made to volume with diethyl ether in a 250 ml volumetric flask. Both vitamins were determined separately using HPLC at different conditions.

2.3 Heavy Metals

The ground fish samples (5g each) were digested using 9.0 ml concentrated HCl and 3.0 ml concentrated HNO_3 in a 25 ml volumetric flask. The digest was made up to mark with deionised water. A blank digest was carried out in the same way and the metals were determined using atomic absorption spectroscopy (AAS) against aqueous standards. The metal concentration was expressed in ppm [28].

2.4 Poly Aromatic Hydrocarbon (PAHs)

The amount of PAHs present in each of the fish sample was determined by weighing 2.0 g of the sample into an extraction unit containing 10.0 g of anhydrous sodium sulphate (Na_2SO_4) salt to absorb moisture. The extraction was carried out using 10 ml dichloromethane (CH_2Cl_2). The dichloromethane extract was cleaned up by passing through a column packed with anhydrous Na_2SO_4 salt. The resulting extract was concentrated on a rotary evaporator to give an oily residue. This oily residue was dissolved with dichloromethane and 1.0 µl was injected into the gas chromatography column for analysis. The GC used was coupled with flame ionization detector (GC –FID). The identification of PAHs was based on comparison of the retention times of the peak with those obtained from standard sample mixture of PAHs.

The amount of PAH from the processed fish sample was extrapolated from external calibration curves prepared from PAH standards.

2.5 Statistical analysis

SPSS version 17.0 was used for statistical analysis. Means were separated using analysis of variance (ANOVA) and Duncan multiple range test; Wahua [29].

III. Results And Discussion

3.1 Vitamins

Results of the Vitamins determined in the fish samples (Vitamin B_6, A and E) are shown in Table 1. Vitamin B_6 had mean values of 14.69, 10.35 and 11.68mg/l; Vitamin A had mean values of 0.632, 0.776 and 0.975 mg/l and Vitamin E had mean values of 10.28, 4.78 and 4.82mg/l for WFF, RFWoS and RFWS respectively.

Table 1 Vitamin Content of Processed Mackerel Fish (mg/ml)

Vitamins	WFF	RFWoS	RFWS
B_6	14.69 ± 0.03^a	10.35 ± 0.89^b	11.68 ± 1.02^b
A	0.632 ± 0.00^a	0.776 ± 0.01^a	0.975 ± 0.03^b
E	10.28 ± 1.00^a	4.781 ± 0.33^b	0.815 ± 0.00^c

Results are means of triplicate determination.
Means within the same row not followed by the same superscript differ significantly ($p\leq0.005$).

Different processing methods affect the vitamin content of a food sample. For instance, water soluble vitamins (Vitamin B_6) are susceptible to destruction at high temperatures in the presence of oxygen whereas fat soluble vitamins (Vitamin A and Vitamin E) are not easily destroyed by heat [30]. Results in Table 1 show that roasting decreased Vitamin B_6 content of the roasted fish. The relative degradation was found to be 41.92% for RFWoS and 25.83% for RFWS. This result corroborates the findings of Ersoy and Ozeren[31]. They reported that Vitamin B_6 content of baked and grilled African cat fish decreased significantly ($p\leq0.005$).

They also reported that solubility in water, exposure to air, exposure to sunlight leading to oxidation, exposure to alkaline solutions as well as lose due to storage, are some other factors in addition to heat which can destroy vitamins and that Vitamin E is more affected by such exposure compared to Vitamin A.

Table 1 shows that Vitamin A content increased while Vitamin E content decreased following processing. This increase in Vitamin A content is contrary to the report of Ersoy and Ozeren[31] who reported a decrease in Vitamin A content of fin-fish (pacific cod) as a result of processing. The decrease in Vitamin E content corroborates with the findings of Francois *et al*., [32] who reported that the Vitamin E content of roasted *curcurbita* seed decreased from 107.0 mg/100g to 68.0 mg/100g.

In this study, Vitamin E content was further reduced in the spiced processed sample compared to the processed samples without spices. Components of the spices may have interacted with the Vitamin E thereby accelerating its degradation during the roasting process. However, Ersoy and Ozeren[31] reported that baking, grilling, microwaving and frying processes increased the vitamin E content of African Cat fish compared to the raw fish. These variations support the report of Greenfield and Southgate [33] where they indicated that vitamin content can vary in different parts of the same tissue and among animals collected at different times and locations.

Vitamin A content in the spiced fish (RFWS) was higher compared to the other samples. This could be attributed to the fact that some of the spices serve as rich sources of vitamins [23,24]

3.2 Heavy Metals

The concentrations of heavy metals in the processed fish are presented in Table 2 below.

Among all the heavy metals analyzed, Cadmium(0.091µg/ml) and Zinc (0.101µg/ml) were more predominant in RFWS. Copper was found in RFWoS (0.045µg/ml) and RFWS (0.087µg/ml) while Lead was present only in RFWS (0.002µg/ml). The concentrations of Zinc present in RFWS and RFWoS were significantly ($p\leq0.05$) higher than that reported for WFF. A total of five out of ten heavy metals analysed were found to be present in detectable quantities.

Studies have shown that heavy metals are toxic, however, trace metals are said to be beneficial to health in certain quantities [6]. Exposing these processed fish samples to metals especially, cadmium, lead and nickel could be hazardous to health. Acute lead exposure can affect cardiac function especially the mechanical and electrical activity of the heart via the alteration of vascular-smooth muscle function. Lead can also affect the intercellular messenger protein kinase C and its role in smooth muscle contraction [6,34]. Effect of cadmium exposure is seen in organs like kidney, placenta, lungs brain and bones. Accumulation of cadmium has some irreversible effects on human health [10]. Cadmium has been reported to increase oxidative stress and lipid peroxidation [34].

The presence of these metals in food may be attributed to metal bioaccumulation in the food chain. In addition, roasting with wood charcoal generates ash (which may contain these metals) that may volatilize and get absorbed in the fish samples. Smoke from the burning charcoal and heavy metal contaminated air may contain volatilized heavy metals which adsorb unto the fish being processed as well.

Zinc is a nutritional essential element for a healthy life; for instances, Zinc is important for proper immune function. Zinc supports normal growth and development and is an essential element required especially for children [35]. Delayed neurological and behavioural development in children occurs as a result of deficiency in zinc [36]. It is also important in the synthesis of DNA [37]. These metals are important cofactors in enzymatic reactions. Interestingly, levels of these five metals identified in the samples analyzed are generally below the permissible levels set by World Health Organization [38].

Table 2 Heavy Metals Contents of Processed Mackerel Fish (µg/ml)

Heavy Metals	WFF	RFWoS	RFWS
Lead	N.D	N.D	0.020
Cadmium	0.006	0.063	0.091
Selenium	N.D	N.D	N.D
Chromium	N.D	N.D	N.D
Mercury	N.D	N.D	N.D
Nickel	N.D	0.001	0.001
Zinc	0.044	0.095	0.101
Copper	N.D	0.045	0.087
Cobalt	N.D	N.D	N.D
Thallium	N.D	N.D	N.D

ND= Not Detected

The spiced sample (RFWS) had higher heavy metal contents than the other samples analyzed. This supports the reports of Darko *et.al.*,[24] which indicated that spices contain heavy metals in varying concentrations; some of which may naturally be present or absorbed during its processing. The heavy metals in the spices become absorbed when applied to the fish sample as seen in RFWS.

3.3 Poly Aromatic Hydrocarbon

The results of analysis of PAH from the various fish samples are presented in Figure 1 below.

A total number of 9, 10 and 12 PAHs were detected for WFF, RFWoS and RFWS respectively. Roasting significantly ($p \leq 0.05$) affected PAH values. The maximum PAH detected with the highest concentration were Benzo(k)fluoranthene (0.090ppm), 2-Methylnaphthalene (0.023 ppm) and Fluorene (0.016 ppm) for RFWS, RFWoS and WFF respectively; whereas the minimum concentration recorded for each of the sample was 0.001ppm. The total amount of PAHs in the entire processed fish sample was 0.30ppm out of which 45.33% were of low molecular mass; while 55.66% were of high molecular mass. The high molecular mass PAHs are considered as being relatively stable [39] and are classified as probable human carcinogens except for chrysene which happens to be among the PAHs considered as low molecular mass though with four aromatic rings. RFWS had more of the high molecular mass PAHs (62.56%) compared to RFWoS and WFF. The non-detection of some PAHs especially of the low molecular weight may be due to instrument sensitivity or their stability [9]. It is also possible that most of these PAHs may have been destroyed by thermal decomposition [8]. Our findings are in agreement with other researchers [5,6,9,12,40].

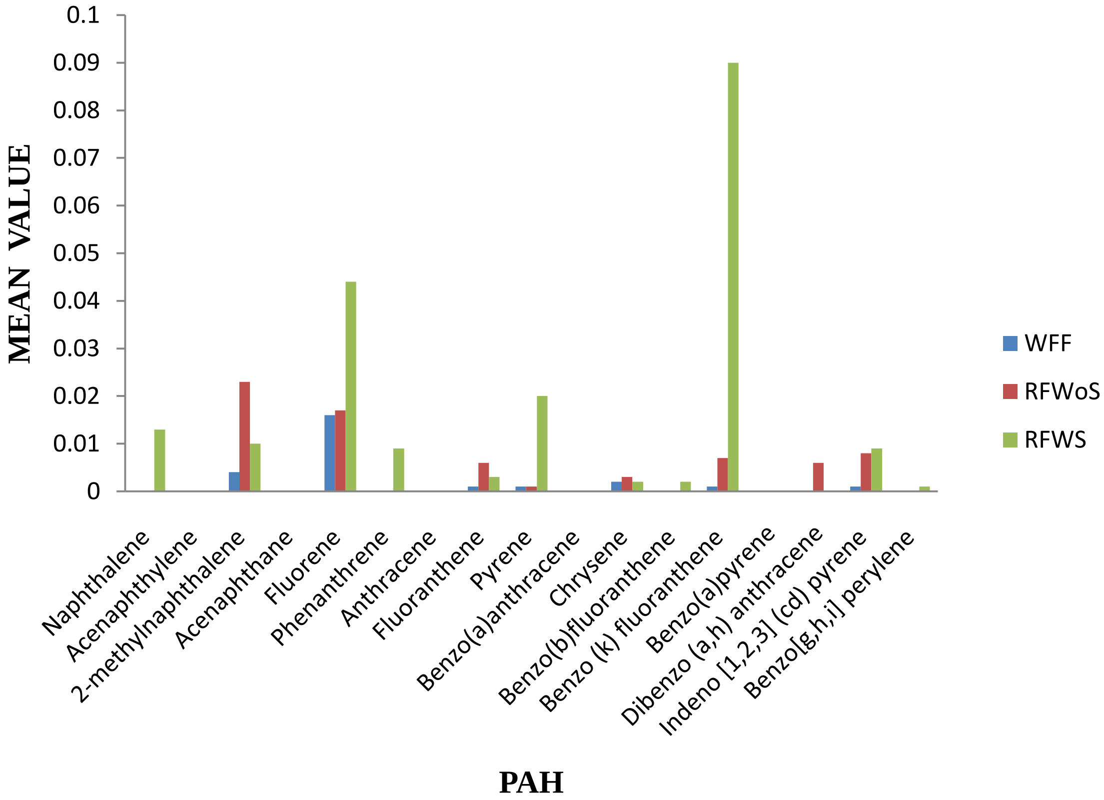

Figure 1: Mean values of the individual PAHs in the processed fish sample

The high PAH content in RFWS shows that most additives contain some amounts of PAHs. Schlemitz and Pfannhauser [41] determined seven nitro-PAHs that are associated with mutagenic activities from tea and spices. This is contrary to the report by ElBadry [42], who reported that treatment of chicken with either spices mixture or garlic paste prior to cooking, caused a reduction in total and carcinogenic PAH levels. He attributed this observation to some inhibitory effect possibly due to some antioxidant properties present in garlic. Besides, some of the variation observed could also be attributed to the interaction between PAHs showing a synergetic effect.

IV. Conclusion

This study reveals that depending on the type of processing and additives applied, Vitamins and toxicants levels are reduced or increased in processed Mackerel fish.

Roasting decreased Vitamin B_6 content by 41.92% in the roasted fish without spices (RFWoS) and 25.83%, in the roasted fish with spices (RFWS). Vitamin A content increased while Vitamin E content decreased following the roasting processing process.

Environmental exposure, food processing technique and the presence of different additives could be major sources of heavy metals and poly aromatic hydrocarbons in street foods. Five heavy metals (cadmium, lead, nickel, copper and zinc) were detected in the processed Mackerel fish. The spiced sample (RFWS) had higher heavy metal contents than the other samples analysed. However the levels of these heavy metals were generally below the World Health Organization permissible limits.

Poly aromatic hydrocarbons (PAHs) were detected at various levels in the processed Mackerel fish samples. The total amount of PAHs in the entire processed fish sample was 0.30 ppm out of which 45.33% were of low molecular weight; while 55.66% were of high molecular weight. The roasted fish with spices (RFWS) had more of the high molecular mass PAHs (62.56%) compared to the other samples.

The decrease in Vitamin B and E content and increase in the level of toxicants following the processing of Mackerel fish by street food vendors may induce negative health consequences for regular consumers.

References

[1]. Hogan, M.C. (2011). Sea of the Hebrides. Eds. P. Sandry & C.T. Cleveland. Encyclopedia of earth national Council for Science and the Environment. Washington DC.

[2]. Rainer, F. & Daniel P. (2006). "In Fish Base Trachurus trachuru."

[3]. Dias, M.G., Sanchez, M.V. Bartolo, H., & Oliveira, L. (2003) Vitamin content of fish and fish products consumed in Portugal. Electronic Journal of Environmental, Agricultural and Food Chemistry, 2(4),510-513.

[4]. Cenzon, M. (2012). Health benefits of Mackerel. Nutrition and Supplements.

[5]. Amos-Tautua, B.M.W, Inengite, A.K., Abasi, C.Y. & Amirize, G.C. (2013). Evaluation of Polycyclic aromatic hydrocarbons and some heavy metals in roasted food snacks in Amassoma, Niger delta, Nigeria. African Journal of Environmental Science and Technology, 7(10), 961-966.

[6]. Ujowundu, C.O., Iheakweazu, K.L., Alisi, C.S. & Ujowundu, F.V. & Igwe, C.U. (2014). Procarcinogens: Polycyclic aromatic hydrocarbons and heavy metal content in some locally processed foods in south eastern Nigeria. British Journal of Applied Science and Technology, 4(1), 249-260.

[7]. Ersoy, B., Yanar, Y., Kucukgulmez, A., Celik, M. (2006). Effect of four cooking methods on heavy metal concentrations of sea bass fillets (Dicentrachus labrax linne,1785). Food Chemistry 99(4), 748-751.

[8]. WHO (2006). World Health Organization. Polycyclic aromatic hydrocarbons. WHO Food additives series 55: Safety evaluation of certain contaminants in Food. International Programme of Chemical Safety (IPCS), Geneva, 563-743.

[9]. Agbozu, I.E. (2014), Polycyclic aromatic hydrocarbon composition and source in food snacks in University community, Nigeria. Research Journal of Chemical and Environmental Sciences, 2(3), 26-31.

[10]. ATSDR (1995). Agency for Toxic Substance and Disease Registry, Public health statement for polycyclic aromatic hydrocarbons (PAHs). US Department of Health and Human Services, Public Services, Atlanta.

[11]. Masih, A., Saini, R.& Taneja, A. (2008). Contamination and exposure profiles of priority polycyclic aromatic hydrocarbons (PAHs) in ground water in a semi-arid region in India. International Journal of Water 4,(1-2), 136-147. Doi:10.1504/IJW. 2008.018152.

[12]. Ogbuagu, D.H., & Ayoade, A.A. (2012). Presence and levels of common polynuclear aromatic hydrocarbon (PAHs) in staple foods of Nigeria. Foods and Public Health, 2(1),50-54.

[13]. Knize, M.G., Salmon, C.P., Pais, P. and Felton, J.S. (1999). Food heating and the formation of heterocyclic aromatic amine and PAH mutagens/carcinogens. In: L.S. Jackson, M.G. Knize & J.N. Norgan (eds). Impact of processing on Food safety, New York: Kluwer Academic.

[14]. Phillips, D.H. (1999). Polycyclic aromatic hydrocarbon in diet. Mutation Research, 443,139-147.

[15]. Nawrot, P.S., Vavasour, E.J. & Grant, D.L., (1999). Food irradiation, heat treatment and related processing Techniques: Safety evaluation, In: K. Vander Heijden, M. Younes, L. Fishbein & E. Miller (eds). International Food Safety Hand Book, New York: Marcel Dekker, 306-308.

[16]. WHO, (1998). World Health Organization. Selected non-heterocyclic PAHs: Environmental Health Criteria, 202 Geneva;

[17]. Badry, N.E. (2010). Effect of Household cooking methods of some food additive, on polycyclic aromatic hydrocarbons (PAHs) formation in chicken meat. World Applied Sciences Journal, 9(9), 963-974.

[18]. Giovasnnvcci, E., Rimm, E.B., Stampfer, M.J. Cloditz, G.A., Ascherio, A.& Willet, W.C. (1994). Intake of fat meat and fiber in relation to risk of colon cancer in men. Cancer Research, 54,2390-2397.

[19]. Lenntech (2008). Water Treatment and Air purification. Water treatment published by Lenntech, Rotterdamseweg, Netherland,

[20]. Ansari, T.M.,Ikram, N., Najam-ul-Haq, M., Fayyaz, O. Ghafoor, I. & Khalid, N. (2004), Essential trace metal (Zu, Mn, Cu, and Fe) levels in plants of medicinal importance. Journal of Biological Science, 4, 95-99.

[21]. Duribe, J.O., Ogwuegbu, M.O.C., Egwurugwu, J.N. (2007) Heavy metal pollution and human biotoxic effects. International Journal of Physical Science, 2(5), 112-118.

[22]. Opuene ,K. & Agbozu, I.E. (2008). Relationships between heavy metals in shrimp (macrobrachium felicinum) and metal levels in the water column and sediments of taylor creek. International Journal of Environmental Research 2(4), 343-348.

[23]. Ogunka-Nnoka, C.U., & Jaja (2012). Micronutrient contents of under utilized spices common in Nigeria. Global Journal of Science Frontier Research Chemistry, 12(4), 13-16.

[24]. Darko. B., Ayim, I., & Voegborlo, R.B.(2004). Heavy metal content in mixed and unmixed seasonings on the Ghanaian market. African Journal of Food Science, 8(1), 14-19.

[25]. Abdullahi, M.S., Uzairu, A., & Okunola, O.Y. (2008). Determination of some trace metal levels in onion leaves from Irrigated farmlands on the bank of River Challawa, Nigeria. African Journal of Biotechnology 7(10), 1526-1529.

[26]. Kaman, Y.F., Wanthesen, J.J. & Labuza, T. P. (1980). Technique for measuring thiamine and riboflavin in fortified foods. Journal of Food Science, 45, 1497-1499.

[27]. Manz, U. Philip, K. O (1981) Method for the routine determination of tocopherols in animal feed and human food-stuffs with the aid of HPLC. International Journal of Vitamin and Nutrition Research 51, 342-348.

[28]. AOAC (1995). Official methods of Analysis (16th ed.) Washington, D.C: AOAC International.

[29]. Wahua, T.A. T. (1999). Applied Statistics for Scientific Studies. African link press Aba, Abia State, Nigeria, 68-76.

[30]. Lund, D.B. (1973). Effect of heat processing. Food Technology, 27, 16-18.

[31]. Ersoy, B., & Ozeren, A. (2009). The Effect of cooking methods on mineral and vitamin contents of African catfish. Food Chemistry, 115,419-422.

[32]. Francois, G. Nathalie, B., Jean-Pierre, V., Daniel., P. & Didier, M. (2006). Effect of roasting on tocopherols of gourd seeds (*Curcubita pepo*). Grasas Y. Aceites, 57(4), 409-416.

[33]. Greenfield, H., & Southgate., D.A.T. (2003). Food composition data: production management and use (2nd ed). Rome: FAO.

[34]. Alissa, E.M. & Fern, G.A. (2011). Heavy metal poisoning and cardiovascular disease. Journal of toxicology, 1-21.

[35]. Colak, H., Soylak, M., & Turkoglu, O. (2005). Determination of trace metal content of various herbal and fruits teas produced and marketed from Turkey. Trace Element Electrolytes, 22,192-195.

[36]. Caulfield, L. E.,, Zavaleta, N., Shankaar, A. H., & Merialdi, M. (1998). Potential contribution of maternal zinc supplementation during pregnancy to maternal and child survival. American Journal of Clinical Nutrition, 68, 499S-508S.

[37]. USDA (2003). United State Department of Agriculture National Nutrient Data Basin: Heavy Metals and Standards, Report no.6.

[38]. Brain, A.F., & Allen, G.C. (1993). Food Science, Nutrition and Health. Edward Arnold Plc. Great Britain, 226-241.

[39]. Igwo-Eakpe, M.N., Gbenle, O.G., Ilori, M.O., Okpuzor, J., & Osuutoki, A.A. (2010). mixture of high molecular weight polycyclic aromatic hydrocarbons: biodegratdation by tropical bacteria and via co-metabolism with phenanthrene. Research Journal of Environmental Sciences, 4;317-326.

[40]. Olabemiwo, O.M., Alade, A.O., Tella, A.C & Adediran, G.O. (2011). Assessment of polycyclic aromatic hydrocarbons content in smoked C. gariepinmus and T. Guineensis fishs species available in Western Nigeria. L. JBAS. IJENS, 11, 135-140.

[41]. Schlemitz, S & Pfannhauser, W. (1996). Analysis of nitro-PAHs in food matrices by-on-line reduction and high performance liquid chromatography. Food Additives and Contaminants 13(8) 969-977.

[42]. Elbadry N. (2010). Effect of household cooking methods and some food additives on polycyclic aromatic hydrocarbons formation in chicken meat. Journal of World Applied Science, 9((9), 963-974.