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AMINO ACID PROFILE AND POLYCYCLIC AROMATIC HYDROCARBONS (PAHS) OF SMOKED FARMED *CLARIAS GARIEPINUS* (BURCHELL, 1822) RAISED UNDER DIFFERENT CULTURE SYSTEMS IN IBADAN, NIGERIA

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ABSTRACT

Fish food quality and safety is critical to consumers due to its public health implication. To exploit huge export opportunities for smoked farmed catfish, there is need for strict adherence to international quality and safety standards. This paper examined amino acid profile and polycyclic aromatic hydrocarbons (PAHs) of smoked farmed catfish *Clarias gariepinus* raised under different culture systems with a view to determining its quality and safety. Six farms with the two most common fish culture systems (3 concrete tanks (CTs) and 3 earthen ponds (EPs) were purposively selected based on frequency of harvest and yield. Fish samples (500±10g) obtained from these farms after 4 months of culture were processed, smoked, packaged and stored for 36 weeks. Amino acid profile in farmed fresh and smoked *C. gariepinus* was determined at 12, 24 and 36 weeks of storage and polycyclic aromatic hydrocarbons (PAH) was determined at 0 and 16 weeks of storage. These were compared with traditional smoked wild catfish obtained from local fish processor (LFP). The amino acids with highest concentrations (mg/g) found in this study were leucine (22.16 - 31.61 and 13.89 – 29.64), lysine (16.31 – 20.19 and 9.86 – 18.08), arginine (15.16 – 12.29 and 8.97 - 15.86), valine (15.96 – 21.35 and 9.68 - 19.36) and asparagine (19.66 – 21.61 and 12.36 mg/g – 20.71mg/g) for fresh and smoked catfish respectively. Levels of other amino acids ranged from 1.5mg/g to 9.98mg/g in smoked fish and 2.95mg/g to 12.21mg/g in fresh fish. The mean total poly aromatic hydrocarbons in smoked catfish at 16 weeks of storage were 0.039±0.004µg/kg, 0.034±0.005 µg/kg and 0.053±0.005 µg/kg for EP, CT and LFP, respectively. Therefore smoked farmed catfish raised under different culture systems still contain essential amino acids and Benzo(a)pyrene which is a carcinogen was not at detectable level in the samples.

Keywords: Benzo(a)pyrene, essential amino acids, catfish, smoking, hydrocarbons

INTRODUCTION

Fish is a major source of food for human populations, providing a significant portion of the protein intake, in the diets of a large

number of people, especially in developing countries (Da Silva, 2002). This protein is relatively of high digestibility, biological and growth promoting value for human consumption. According to Osibona *et al*., (2009), fish proteins comprise all the ten essential amino acids which are: lysine, leucine, arginine, methionine, alanine, histidine, phenylalanine, isoleucine, threonine and tryptophan in desirable quantities for human consumption. This accounts for the high biological value of fish flesh, which also provides minerals, iodine, vitamins and fat. Fish cooks easily, offers palatable taste and flavour and is easily digestible. Fish is consumed either as a preparation from freshly caught fish or from those that have been preserved in some form (Da Silva, 2002). Fish, however, is more susceptible to spoilage than certain other animal protein foods; such as meat and egg. As part of the natural process by which organic matter is broken down and returned to the nitrogen cycle, fish is rapidly invaded, digested and spoiled by the microorganisms which are abundant on the skin, gills and intestines. To prevent spoilage of fish, some form of preservation is necessary. Eyo (2001) stated that, preservation is a means of keeping the fish, after landing in a condition wholesome and fit for human consumption, for a short period of few days or for a longer period of over a few months. During the period of preservation, the fish must be kept as 'fresh' as possible, with minimum reduction in flavour, taste, odour, form, nutritive value, weight and digestibility of flesh. This preservation should cover the entire period from the time of capture of the fish to its sale at the retailer's counter (Eyo, 2001).

Amino acids are important biomolecules that serve as building blocks of proteins and are intermediates in various metabolic pathways. Mohanty *et al*., (2014), stated that amino acids serve as precursors for synthesis of a wide range of biologically important substances including nucleotides, peptide hormones, and neurotransmitters. Amino acids are mainly obtained from proteins in diet and the quality of dietary protein is assessed from essential to non-essential amino acid ratio. High quality proteins are readily digestible and contain the dietary essential amino acids (EAA) in quantities that correspond to human requirements (Usydus *et al*., 2009). According to Mohanty *et al.,* (2014) inadequate uptake of quality proteins and calories in diet leads to protein-energy malnutrition (PEM) (or protein-calorie malnutrition, PCM) which is the most lethal form of malnutrition/hunger. Kwashiorkor and marasmus, the extreme conditions of PCM mostly observed in children, are caused by chronic deficiency of protein and energy, respectively.

Polycyclic aromatic hydrocarbons (PAH) are formed when complex organic substances are exposed to high temperature or pressure or by the incomplete combustion of woods, coal or oil. They can be found in complex mixtures throughout the environment (Easton *et al*., 2002 and Ikechukwu *et al*., 2012). Foods can be contaminated by PAHs from environmental sources, industrial food processing and during home food preparation. This important class of carcinogens had been studied by many authors and several hundreds of PAHs have been identified. Sixteen of these PAHs have been considered to be more harmful than others (Ikechukwu *et al*, 2012; Wretling *et al*., 2010; Anyakora and Coker, 2007 and Chimezie and Hebert, 2006). The aim of this paper is to examine the amino acid profile and polycyclic aromatic hydrocarbon of smoked farmed *Clarias gariepinus* raised under different culture systems.

MATERIALS AND METHOD

Collection and processing of Samples

Fifty kilogram of fresh samples of *Clarias gariepinus* were collected from the six commercial fish farms in Ibadan. The fish was transported to the processing laboratory live in a plastic bucket with water. The distance of the fish farms to the processing laboratory is about two kilometres. At the processing laboratory the samples were placed in ice for five hours and the rapid change in temperature led to the death of the fish; and ice also preserved it. The preserved fish were subjected to some pre-processing operations and then smoked under Good Manufacturing Practices (GMP). The smoking time and temperature were monitored during the smoking operations and the smoking was terminated when the fish were properly dried (80^{o}C – 100^{o}C for 6 -10 hours). The smoked samples were analyzed for amino acid profile and polycyclic aromatic hydrocarbons.

Smoked fish from local processor: Smoked fish sample from a commercial fish processor in Ibadan was obtained in order to compare the quality with the experimental fish. The processor used open top kiln in smoking the fish.

Amino acids profile of fresh and smoked *Clarias gariepinus*

The amino acid in the samples were analysed using Waters 616/626 Transducer Pump HPLC liquid chromatograph (LC) Instrument.

Hydrolysis of the samples: 0.5g of the samples were weighed into a sterile furnace hydrolysis tube. 5nmols norleucine was added to the samples and then dried under vacuum. The tube was again placed in a vial containing 10.05HCl with a small quantity of phenol, thereby hydrolysing the protein by the HCl vapours under vacuum. This stage of hydrolysis of the sample lasted between 20 -23hrs at 108^{o}C. After the hydrolysis the samples were dissolved in ultra-pure water (HPLC) grade, containing ethylenediaminetetraacetic acid (EDTA). The EDTA chelates the metal present in the samples. The hydrolysed samples were stored in HPLC amino acid analyser bottles for further analytical operations.

Derivatisation: The hydrolysed samples were derivatised automatically on the Waters 616/626 HPLC by reacting the five amino acids, under basic situations with phenylisothiocyanate (i.e PITC) to get phenylthiocarbamyl (PTC) amino acid derivatives. The duration for this was 45 minutes per sample, as calibrated on the instrument. A set of standard solutions of the amino acids were prepared from Pierce Reference standards H (1000umol) into auto-sampler crops and they were also derivatised. These standards (0.0, 0.5, 1.0, 1.5, 2.0umpl) were used to generate a calibration file that was used to determine the amino acid contents of the samples. After the derivatisation, a methanol solution (1.5N) containing the PTC-amino acids was transferred to a narrow bore waters 616/626 HPLC system for separation.

The HPLC separation & Quantization: The separation and quantization of the PTC-amino acids were done on a reverse phase (18 silica column and the PTC chromophone were automatically and digitally detected at the wavelength of 254nm). The elution of the whole amino acids in the samples took 30minutes. The buffer system used for separation was 140nm sodium acetate pH 5.50 as buffer A and 80% acetonitrile as buffer B. The program was run using a gradient of buffer A and buffer B concentration and ending with a 55% buffer B concentration at

the end of the gradient.

Data interpretation and Calculation: The intensity of the chromatographic peaks areas were automatically and digitally identified and quantified using a Dionex chromeleon data analysis system which is attached to the Waters 616/626 HPLC system, The calibration curve or file prepared from the average values of the retention times (in minutes) and areas (in Au) of the amino acids in 5 standards runs was used. A response factor (Au/pmol) was calculated by the software that was interphase with the HPLC loaded with the standard amino acid. The response factor was used to calculate the amount of each of the amino acid (in pmols) in the sample and displayed on the system digitally. The amount of each amino acid in the sample is finally calculated by the software by dividing the intensity of the peak area of each (corrected for the differing molar absorptivities of the various amino acids) by the internal standard (i.e. Pierce) in the chromatogram and multiplying this by the total amount of internal standard added to the original sample. The software ascertained the picomole by the intensity of the height of each amino acid. Then the digital chromatographic software extrapolated back to 5nmoles of the internal standard (Norleucine), and displays for the total amount that was pippeted into the hydrolysis tube at the beginning of the analysis as shown below:

$$\text{mg/ml (in Extract)} = \text{Dilution factor} \times \text{Peak height intensity}$$

$$\text{mg/ml (in sample)} = \frac{\mu\text{g/ml in extract}}{\text{sample volume}} \times \text{Wt. of sample}$$

Analysis of Polycyclic Aromatic Hydrocarbons (PAHs)

The PAHs of the farmed smoked *C. gariepinus* and smoked fish from LFP were determined using Gas Chromatographic techniques using the method of Garcia-Falcon *et al.*, (1996). For the determination of the PAHs content, 5.0g of each type of smoked fish were weighed into amber glass bottles and extracted sequentially by ultrasonication using 25 ml of n-hexane for 1 hour. After ultrasonication, the supernatant of the extracts were decanted into a vial and 15 ml of fresh solvent was added for another 1hour of ultrasonication. The process was repeated with another 10 ml of fresh solvent for 1h. The combined extracts (50ml) were centrifuged at 2500 rpm for 10 min and the supernatant was decanted. The supernatant was cleaned-up using the Whatman nylon filter membrane. Further clean-up was done using the solid phase extraction (SPE) cartridges. The sorbent of the SPE cartridges were first conditioned with n-hexane, after which the filtered extracts were loaded on to the cartridges, the analytes were eluted with dichloromethane. The volume of the dichloromethane was blown down to dryness and extract was reconstituted in 200µl of acetonitrile. After purification of the sample with n-hexane solution on Silica SPE column, it was concentrated and analyzed on Agilent Model 6890 gas chromatograph equipped with the mass selective detector Model 5973. PAH determination is mostly limited to the analysis of 17 compounds by GC. The quantification of PAH in the sample is done by the use of calibration standards. All calculations were done by the computer application (HP Chemstation) software.

The response factor (RF) and the PAH concentration (mg/l) is calculated from the chromatogram of the standard mixture as follows:

$$RF = \frac{\text{Concentration (mg/l)}}{\text{Area counts}}$$

C = RF x Area x V/Q
Where;
V = volume in which sample has been dissolved before injection
Q = amount of sample extract in ml.

Data Analysis

Data obtained were subjected to Student's T test to compare means and significance at $\alpha_{0.05}$ using IBM SPSS Statistics version 20.

Results

Amino Acid Profile of Fresh and Smoked *C. gariepinus*

The composition of amino acids in fresh and smoked *C. gariepinus* is shown in Tables 1 and 2. Total amino acids in fresh farmed *C. gariepinus* ranged from 204.91mg/g to 220.22mg/g. In smoked fish, total amino acids ranged from 141.18mg/g to 163.18mg/g. The most abundant amino acids in fresh and smoked fish were leucine, lysine, asparagine, valine, phenylalanine, isoleucine, methionine and tryptophan. In this study the percentage concentration of leucine in smoked *C. gariepinus* decreased with storage time while percentage concentration of lysine increased with storage time. Other amino acids remained fairly stable throughout the storage period.

Table 1: Amino acid composition of fresh *Clarias gariepinus* under different culture systems

Amino Acids (mg/g)	Earthen Pond	Concrete Tank	Local Fish Processor
Tryptophan	8.41	10.22	7.56
Phenylalanine	13.39	9.55	13.21
Isoleucine	6.71	8.17	12.21
Tyrosine	9.01	5.11	6.05
Methionine	6.11	7.15	8.09
Proline	4.39	4.77	5.16
Valine	15.96	21.35	17.09
Threonine	8.43	11.22	9.56
Histidine	2.95	5.08	5.19
Alanine	7.81	5.96	8.11
Glutamine	5.57	4.81	6.73
Glutamic acid	3.08	4.11	3.29
Glycine	12.6	18.2	13.1
Serine	8.73	7.74	8.56
Arginine	12.29	20.12	15.16
Aspartic acid	5.88	4.32	6.12
Asparagine	20.86	19.66	21.61
Lysine	16.31	20.19	17.73
Leucine	31.61	22.16	31.56
Cysteine	4.81	6.15	4.13
Total	204.91	216.04	220.22

Table 2: Amino acid composition of farmed smoked *C. gariepinus* stored under ambient condition

	Farmed smoked fish sample			Smoked fish from LFP		
Amino Acids (mg/g)	**12 weeks**	**24 weeks**	**36 weeks**	**12 weeks**	**24 weeks**	**36 weeks**
Tryptophan	7.56	8.26	9.65	8.33	8.26	7.22
Phenylalanine	8.66	7.56	6.98	6.84	5.68	9.21
Isoleucine	4.98	9.06	6.54	6.38	7.62	7.77
Tyrosine	7.25	4.69	4.49	3.69	5.66	5.62
Methionine	4.22	5.26	7.25	3.69	6.66	4.69
Proline	3.05	3.56	3.65	3.05	4.28	4.52
Valine	9.88	12.36	9.68	19.35	10.31	15.44
Threonine	6.35	7.26	6.94	9.65	7.55	6.26
Histidine	1.56	3.02	2.94	2.14	3.01	2.99
Alanine	4.93	6.35	4.99	4.05	5.06	5.62
Glutamine	4.12	4.69	4.03	3.99	3.89	3.66
Glutamic acid	1.98	1.87	1.96	3.01	1.65	2.03
Glycine	9.98	9.99	11.14	10.20	9.87	10.66
Serine	6.55	6.57	4.65	5.68	5.22	5.68
Arginine	9.98	9.77	10.23	10.43	9.96	8.97
Aspartic acid	3.69	4.26	2.98	3.05	3.21	3.75
Asparagine	16.64	16.55	12.37	13.24	10.89	12.36
Lysine	9.98	10.14	10.24	10.33	9.77	9.86
Leucine	29.64	26.64	18.96	13.89	17.75	23.54
Cysteine	3.77	5.32	6.03	4.56	4.88	4.98
Total	154.77	163.18	145.70	145.55	141.18	154.83

The result of paired sample test revealed that values of amino acids obtained in smoked fish under storage were significantly different ($p>0.05$) from the values obtained from fresh fish (Table 3).

Table 3: Result of Paired samples Test of amino acid values in fresh and smoked *Clarias gariepinus* under different culture systems

Fish Samples	**df**	**t**	**Sig.(2-tailed)**
Farmed smoked fish			
Fresh - ST12	19	5.406	0.000
Fresh – ST24	19	5.067	0.000
Fresh – ST36	19	4.874	0.000
Smoked fish from LFP			
Fresh – ST12	19	4.325	0.000
Fresh – ST24	19	4.846	0.000
Fresh – ST36	19	5.455	0.000

ST12, ST24, ST36 = Storage time at 12, 24, and 36 weeks respectively

Polycyclic aromatic hydrocarbons (PAH) in Farmed Smoked *Clarias gariepinus*

The results of the polycyclic aromatic hydrocarbons in smoked fish samples are presented in Figures 1 and 2. The total concentration of PAHs detected in fresh *C. gariepinus* raised in earthen pond, concrete tank and local fish processor was 0.0961µg/kg. In this study, the PAHs in farmed smoked catfish were 0.039 ± 0.004µg/kg, 0.034 ± 0.005 µg/kg and 0.053 ± 0.005 µg/kg from EPs, CTs and LFP respectively. Benzo (a) pyrene (B(a)P) which is widely accepted as indicator for the level of Polycyclic Aromatic Hydrocarbons (PAH) in food is not in the detectable range in the smoked samples. The results of PAH4 [Anthracene (0.003 µg/kg – 0.005 µg/kg), Chrysene (0.001 µg/kg – 0.004 µg/kg), Pyrene 0.05 µg/kg – 0.07 µg/kg) and Fluovanthene (0.03 µg/kg – 0.07 µg/kg)] obtained in this study were below the recommended maximum permissible limit. Therefore, the values of PAHs obtained in this study were still within FAO/WHO limit and the fish are safe for human consumption.

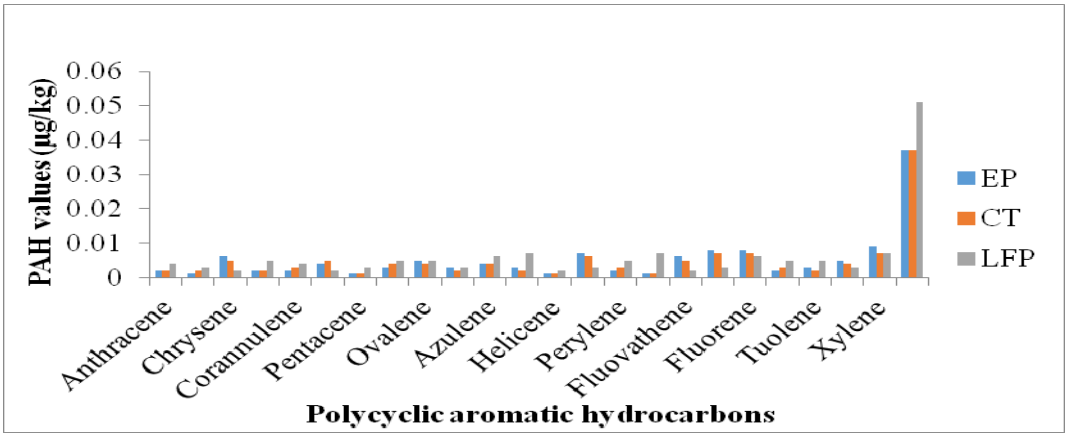

Figure 1: Polycyclic aromatic hydrocarbons in smoked farmed *Clarias gariepinus* from EPs, CTs and LFP at 0week of storage

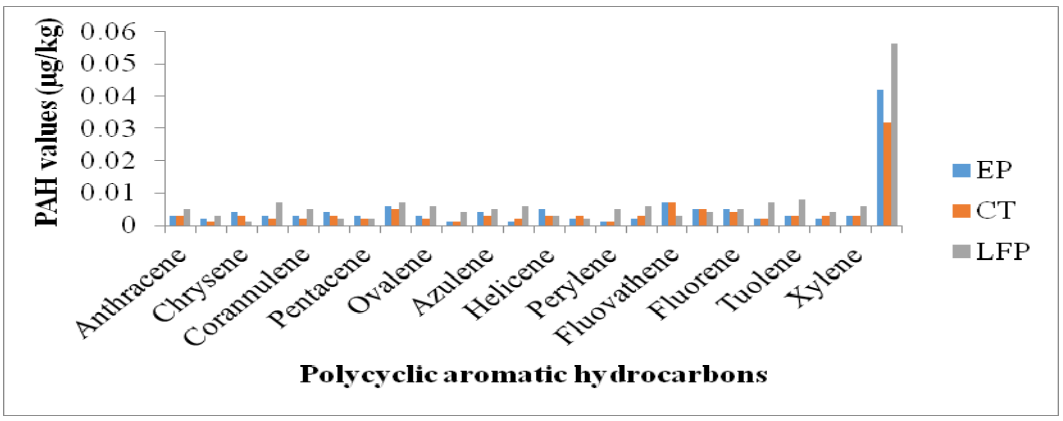

Figure 2: Polycyclic aromatic hydrocarbons in smoked *Clarias gariepinus* from EPs, CTs and LFP at 16week of storage

DISCUSSION

mino Acid Profile of Fresh and Smoked *C. gariepinus*

All essential amino acids that are very important for human body are all present in the fish. These essential amino acids are lysine, leucine, valine, isoleucine, threonine, phenyl alanine, methionine, histidine and tryptophan. Ibhadon *et al.,* (2015) reported total amino acid value of 107.96mg/g in farm-raised catfish in Kaduna, Nigeria. Osibona (2011) also reported value of 165.84mg/g of amino acid in *C. gariepinus*. Amino acids are also important in healing processes and the composition of amino acids in fish is required by man for good health. Therefore, people can acquire essential amino acids in abundance and proper balance by eating fish (Osibona *et al.,* 2009). The essential amino acids cannot be manufactured in the human body, but can be obtained from food. Deficiency in the essential amino acids may hinder the healing recovery process (Osibona *et al.,* 2009). Leucine promotes the healing of bones, skin and muscle tissues. Isoleucine is necessary for haemoglobin formation, stabilizing and regulating blood sugar and energy. Glycine which is one of the major components of human skin, collagen, together with other essential amino acids such as alanine form a polypeptide that will promote re-growth and tissue healing (Mohanty *et al.,* 2014). Other reports of similar nature provided valuable information on selected fish and fish oils for nutritional purposes (Osibona, 2011). All the essential amino acids needed for normal body functioning are present in *Clarias gariepinus*.

Polycyclic aromatic hydrocarbons (PAH) in Farmed Smoked *Clarias gariepinus*

The actual levels of PAHs in smoked foods depend on several variables in the smoking process including type of smoke generator, combustion temperature and degree of smoking (Silva *et al.*, 2011). The composition of the smoke and the conditions of process affect the sensory quality, shelf life and wholesomeness of the product. Potential health hazards associated with smoked foods may be caused by carcinogenic components of wood smoke; mainly PAHs, derivatives of PAHs, such as nitro-PAH or oxygenated PAH and to a lesser extent heterocyclic amines (Silva *et al.*, 2011). The smoke for smoking of food develops due to the partial burning of wood, predominantly hardwood, softwood and bagasse. Anyakora and Coker (2007) used the presence of high value Benzo(a)pyrene (2.32μg/kg) detected in *Clarias gariepinus* as indicator PAH-compound for fresh fishes caught in highly polluted rivers of Niger Delta Region of Nigeria. Olabemiwo *et al.,* (2011) reported total concentration of PAHs of 0.508, 0.497, 0.900, 0.814 and 0.570μg/kg in smoked *Clarias gariepinus* sampled in Ibadan, Ilorin, Ketu, Oyo and Ogbomoso, respectively. They also reported total PAHs concentration of 0.519, 0.738, 0.950, 0.772 and 0.613μg/kg for smoked *Tilapia guineensis* obtained from Ibadan, Ilorin, Ketu, Oyo and Ogbomoso, respectively. Ajai *et al.,* (2012) reported value of PAHs between 0.75 μg/kg – 2.25 μg/kg, 0.40 μg/kg – 2.00 μg/kg and 0.25 μg/kg – 1.75 μg/kg in smoked catfish obtained from fishing zones in Niger State, Nigeria using different extraction methods. Mičulis *et al*, (2011) reported higher values of PAHs 15.30μg/kg in Hake, 380μg/kg in Trout and 20.20μg/kg in Herring. PAHs with maximum concentrations detected in fresh fish are 2-methyl-naphtalene (0.071μg/kg), Acenaphthalene (0.011μg/kg), Fluorene (0.011 μg/kg) and Phenanthrene (0.0022 μg/kg). The FAO and WHO have set a maximum permissible concentration of B(a)P in food of 10 μg/kg (Joint FAO/WHO

Expert Committee on Food Additives, 1987) while a level of 5µg/kg has been set by the European Commission (2005). For smoked-dried fish, this level applies only to the final ready to eat product. Maximum levels of Benzo (a) pyrene allowed was 5.0µg/kg until 31st August, 2014 and has been reduced to 2.0µg/kg as from 1st September, 2014. New maximum levels for the sum of four substances (PAH4) benzo(*a*) pyrene, benz(*a*)anthracene, benzo(*b*) fluoranthene and chrysene was 30.0 µg/kg as from 1st September, 2012 until 31st August, 2014 and reduced to 12.0 µg/kg as from 1st September, 2014. (EC, 2011).

Conclusion

Amino acids are important in healing and metabolic processes in humans. The essential amino acids needed by man are present in fish. Therefore, people can get enough essential amino acids by eating fish. The PAHs values obtained in this study were lower than the limit set by the Joint FAO/ WHO which made the fish samples safe for consumption and may not pose the problem of carcinogenicity usually associated with smoked fish.

REFERENCES

Ajai A. Ikechukwu, Suleiman M. Tanko, Dauda B. Emmanuel, Sadiku O. Suleiman and Aberuagba, F. 2012: Effect of Extraction Methods on the Polycyclic Aromatic Hydrocarbons Content Smoked Catfish Species in Niger State of Nigeria. *Jordan Journal of Biological Sciences* 5(1),71 - 80

Anyakora, C and Coker H 2007. Assessment of Polynuclear aromatic hydrocarbon content in four species of fish in Niger Delta by Gas chromatography/ mass spectrometry *African Journal of Biotechnology* 6, 737-743

Da Silva A. L. V. 2002. Hazard Analysis Critical Control Point (HACCP), Microbial safety and shelf life of smoked Blue Catfish (*Ictalurus furcatus).* A M.Sc. Thesis submitted to the graduate Faculty of the Louisiana State University and Agricultural and Mechanical College.

Chimezie A and Herbert C. 2006. Determination of polynuclear aromatic hydrocarbons (PAHs) in selected water bodies in the Niger Delta. *African J of Biotechnology,* 5 (21): 2024-2031.

Easton M.D.L, Luszniak D and Von der Geest E. 2002: Preliminary examination of contaminant loadings in farmed salmon, wild salmon and commercial salmon feed. *Chemosphere,* 46: 1053-1074.

European Commission (EC) 2011: Draft COMMISSION REGULATION (EU) No …/…of amending Regulation (EC) No. 1881/2006 as regards maximum levels for polycyclic aromatic hydrocarbons in foodstuffs. Brussels, C (2009) SANCO/10616/2009 rev. 7 (11.4.2011)

European Commission 2005 Directive No. 208/2005 of 4 February 2005, amending Regulation (EC) No. 466/2001 as regards polycyclic aromatic hydrocarbons. *Official Journal of European Union*, Brussels, Belgium

Eyo, A. A. 2001. Fish Processing Technology in the Tropics. University of Ilorin press, Nigeria. 403pp

Garcia-Falcon M. S. G, Amigo G. S, LMA Y, Villaizan M. J, and Lozano S. L 1996. Enrichment of benzo(a)pyrene in smoked food products and determination by high-performance liquid chromatography fluorescence. *Journal Chromatograph* 753; 207-215.

Ibhadon S, M. S. Abdusalami, M.C. Emere and V. Yilwa 2015: Comparative study of Proximate, Fatty and Amino Acids Compo-

sition of Wild and Farmed Raised African Catfish *C. gariepinus* in Kaduna, Nigeria. *Pakistan Journal of Nutrition* 14 (1): 56 -61

Ikechukwu A. A, Suleiman M.Tanko, Dauda B.Emmanuel, Sadiku O.Suleiman and Aberuagba, F. 2012: Effect of Extraction Methods on the Polycyclic Aromatic Hydrocarbons Content Smoked Catfish Species in Niger State of Nigeria. *Jordan Journal of Biological Sciences* Volume 5, Number 1, Pages 71 - 80

Joint FAO/WHO Expert Committee on Food Additives (JECFA) 1987. Evaluation of certain food additives and contamination. *WHO Technical Report Series,* 31, 759 p

Mohanty B, Mahanty A., Ganguly S, Sankar T. V, Chakraborty K, and Rangasamy A, *et al*, 2014: Amino Acid Compositions of 27 Food Fishes and Their Importance in Clinical Nutrition Journal of Amino Acids Volume 2014 (2014), Article ID 269797, 7 pages http:// dx.doi.org/10.1155/2014/269797 assessed October, 2015

Mičulis J, Valdovska A, Šterna V and J. Zutis 2011: Polycyclic Aromatic Hydrocarbons in Smoked Fish and Meat *Agronomy Research* 9 (Special Issue II), 439–442

Olabemiwo, O.M; Alade, O.A; Tella, A.C and Adediran, G.O 2011. Assessment of Polycyclic Aromatic Hydrocarbons content in smoked *Clarias gariepinus* and *Tilapia guineensis* fish species available in Western Nigeria *International Journal of Basic and Applied Sciences* IJBAS-IJENS vol.11 No 02 pg. 135-150

Osibona A. O. 2011. Comparative study of proximate composition, amino acids and fatty acids of some economically important fish species in Lagos, Nigeria. *African Journal of Food Science* 5: 581-588.

Osibona A.O, Kusemiju K. and Akande G. R 2009. Fatty acid composition and Amino acid profile of two Freshwater species, African catfish (*Clarias gariepinus*) and Tilapia (*Tilapia zilli*).*African Journal of Food Agriculture Aesthetics and Development* (AJDFAND online) Vol. 9 No. 1 pp 609 - 621 Assessed 13/8/2012

Silva, B. O; Adetunde O. T.; Oluseyi T. O; Olayinka K.O. and B.I Alo B. I. 2011. Effects of the methods of smoking on the levels of Polycyclic aromatic hydrocarbons (PAHs) in some locally consumed fishes in Nigeria *African Journal of Food Science* Vol 5(7) pp384-391 July 2011 Retrieved Nov. 2012 from http://www.academicjournals.org/ajfs

Usydus, Z., Szlinder-Richert J. and Adamczyk. M. 2009. Protein quality and amino acid profiles of fish products available in Poland. *Food Chemistry*, 112: 139-145.

Wretling S, Eriksson A, Eskhult G A and Larson B. 2010. Polycyclic aromatic hydrocarbons in Swedish smoked meats and fish. *J of Food Composition and Analysis,* 23(1)**:** 264-272.

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