Full Length Research Paper

Evaluation of polycyclic aromatic hydrocarbons and some heavy metals in roasted food snacks in Amassoma, Niger Delta, Nigeria

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Accepted 1 September, 2013

Analysis of raw and roasted ready-to-eat foods namely; Atlantic mackerel (*Scomber scombrus*), suya beef and plantain (*Musa paradiasca*) sold and consumed in Amassoma town were screened for the presence of 15 polycyclic aromatic hydrocarbons (PAHs). Concentration of chromium, lead and cadmium were also determined after digestion with aqua regia and analysed using atomic absorption spectrophotometer. Column chromatography, packed with anhydrous Na₂SO ₄ and silica gel was used for PAH extraction with dichloromethane as the eluting solvent. The identification and concentration of PAHs were carried out by gas chromatography GC/FID with the aid of 15 PAH standards. Appreciable amount of benzo[a]pyrene (2.41 μ g/g) and benzo[b]fluoranthene (4.51 μ g/g) were found present in roasted mackerel fish; while a mean of 7.23 μ g/g of benzo[a]anthracene was detected in suya beef. Thirteen (13) other PAHs were present at various concentrations in the smoked fish. No PAH was detected in roasted plantain and in the raw food items. Levels of the heavy metals were found in trace amounts and were below World Health Organization limits for human consumption.

Key words: Gas chromatography, benzo[a]pyrene, chromium, lead, smoked fish, suya plantain.

INTRODUCTION

One of the ubiquitous sets of chemicals that abound in the environment is the polycyclic aromatic hydrocarbons (PAHs), also known as the poly nuclear aromatic hydrocarbons. PAHs constitute a large class of organic compounds, containing 2 or more fused aromatic rings made up of carbon and hydrogen atoms. The chemical structures of some PAHs are shown in Figure 1. Food is one source of PAH (Guillen et al., 1997). When food particularly meat, meat products and fish is smoked, roasted, barbecued, or grilled; PAHs are formed as a result of incomplete combustion or thermal decomposition of the organic materials (WHO, 2006). Pyrolysis of the fats in the meat/fish generates PAH that

become deposited on the meat/fish. PAH production by cooking over charcoal (barbecued, grilled) is a function of both the fat content of the meat/fish and the proximity of the food to the heat source (Phillips, 1999; Kazerouni et al., 2001).

Several analyses of charcoal roasted/grilled common food items have proven the presence of PAHs such as benzo[a]pyrene, anthracene, chrysene, benzo[a]anthracene, indeno[1,2,3-c,d]pyrene (Ogbadu and Ogbadu, 1989; Akpan et al., 1994; Duke and Albert, 2007; Linda et al., 2011; Akpambang et al., 2009). Most of these PAHs have been found to be carcinogenic while some are not (Bababunmi et al., 1982; Alonge, 1988;

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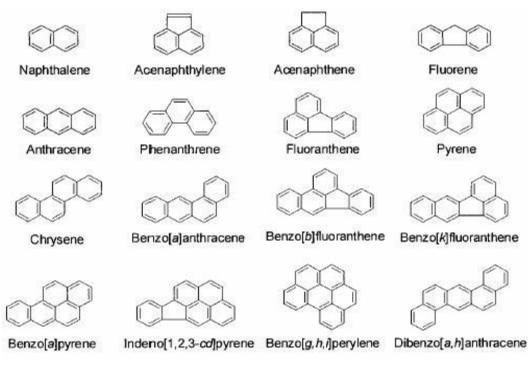


Figure 1. Chemical Structure of Some PAHs.

Lijinsky, 1999; Fritz and Soos, 1980; Borokovcova et al., 2005). Emerole (1980) screened for the presence of PAH in local foodstuffs available in Nigerian market. They discovered that appreciable amounts of benzo[a]anthracene and benzo[a]pyrene were found present in three varieties of smoked fish and smoked meat (suya) purchased from a popular market in Ibadan, Nigeria. In a recent study carried out by Olabemiwo et al. (2011) to assess the PAHs content of 2 smoked fish species available in Western Nigeria; it was found out that the sum of all PAHs in the smoked fish Caria gariepinnus and Tilapia guineensis ranged from 0.497 to 0.814 µg/kg and 0.519 to 0.772 µg/kg, respectively. High levels of PAHs have been reported to be associated with the dark colorations in intensively heated products. This was supported by Ova et al. (1998) who reported that the PAH levels were significantly higher in the fish skins than in the edible parts.

Certain heavy metals such as lead, cadmium, mercury, arsenic have been recognized to be potentially toxic within specific limiting values. Some of them, such as copper, nickel, chromium and iron, for example, are essential in very low concentrations for the survival of all forms of life. These are described as essential trace elements. Only when they are present in greater quantities, can these, like the heavy metals lead and cadmium which are already toxic in very low concentrations, cause metabolic anomalies (Smirjakova et al., 2005).

Cadmium is a relatively volatile element not essential to

plants, animals and humans. It is spread by air and water (sewage sludge) far over sea and land, but especially in the vicinity of heavy industrial plants. Cadmium is today regarded as the most serious contaminant of the modern age. It is absorbed by many plants and sea creatures and, because of its toxicity, presents a major problem for foodstuffs. Cadmium, like lead, is a cumulative poison, that is, the danger lies primarily in the regular consumption of foodstuffs with low contamination (Zeleznik, 1994).

Chromium in food is as a result of washing of industrial waste containing chromium into river and pond sediments as contaminants. The health effect from exposure to Cr are dermatitis, skin inflammation, chronic allergic reactant, asthma-like condition in lungs and respiratory tract, lung cancer, weak carcinogenic infections . Total Cr concentrations in food are low (Barry et al., 2000).

Roasted foods (fish, "suya" meat and plantain) are commonly sold and consumed as ready-to-eat snacks by large population in the Niger Delta region of Nigeria. Frozen fish samples mostly Atlantic mackerel (*Scomber scombrus*) are thawed, scaled and eviscerated, washed in water and steeped into palm oil mixed with dried pepper and salt. These are then placed on open charcoal fire. The "suya" meat is prepared from boneless beef

.The meat is sliced into continuous sheets, cut into pieces and staked on sticks and spiced with groundnut oil, salt, ground nut powder/flour, ginger, dried pepper and flavourings such as monosodium glutamate. The sticks are then arranged round on wire gauze placed over an open charcoal fire (Inyang et al., 2005).

Roasted plantain is prepared by firstly washing ripe/unripe plantain in water and peeled. The raw pulps are placed on wire gauze that was over an open charcoal fire to roast until they are slightly brown.

The primary purpose of this study was to report the concentration levels and distribution of PAHs and selected heavy metals in raw and roasted ready –to –eat snacks consumed by people of the Niger Delta region in Nigeria. The study is also aimed to supplement the information on food contaminants available for roasted foods and to determine possible potential health risk on consumption of these snacks.

MATERIALS AND METHODS

Collection of food items

Samples of frozen and roasted Atlantic mackerel (*Scomber scombrus*, Scombridae), suya beef, and plantain (*Musa paradiasca*) used for the study were obtained from three randomly selected sales spots in Amassoma town, Nigeria.

Extraction of food samples for PAH determination

2 g of each of the homogenized food samples was thoroughly mixed with anhydrous Na₂SO₄ salt to absorb moisture and then grade extracted with unspecified quantity of analytical dichloromethane (CH2Cl2). The dichloromethane extract was cleaned up by passing through a column packed with anhydrous Na₂SO₄ salt. The resulting extract was concentrated on a rotary evaporator to give an oily residue; which was again dissolved in 1ml CH2Cl2 and 1µL was injected into the GC for analysis. The gas chromatography used was Hewlett Packed 589 0 series II, coupled with flame ionization detector (FID) (Hewlett Packard, Wilmington, DE, USA). The identification of PAHs was based on comparison of the retention times of the peaks with those obtained from standard mixture of PAHs (standards supplied by instrument manufacturer). Quantification was based on external calibrations curves prepared from the standard solution of each of the PAHs.

Digestion of food samples for heavy metal determination

3 g of each food sample was accurately weighed after pulverization and homogenization. The homogenized samples were weighed into the digestion tubes. 10 cm³ conc. H₂SO₄ and 5 cm³ conc. HNO₃ was added. The sample was digested and its volume was reduced to 2 cm³. The digestion was continued until the solution was colourless. This ensured the removal of all HNO₃. The sample was allowed to cool and 15 cm³ of water was added with gentle swirling. 1 M NaOH was added dropwise until a pink tinge, brown or colourless solution was produced. The solution was filtered using a Whatman filter paper No.42 followed by dilution to the mark in a 25 cm³ volumetric flask. The digested food samples were analysed for Pb, Cd and total Cr concentration using SOLAAR M Atomic Absorption Spectrophotometer. All determinations were carried out in triplicate and reported as mean mineral content in µg/g.

RESULTS AND DISCUSSION

A summary of the concentrations of various PAHs present in both raw and roasted food samples is shown in Table 1. All the 15 targeted PAHs were detected in reasonably quantity in the roasted fish and only 3 were found in suya beef; but none was detected in the raw food items and roasted plantain. This is in agreement with other inferences made by other researchers that raw foods do not normally contain high levels of PAHs but they are formed during processing, roasting, baking, smoking or frying (Kayali et al., 1999; RAS, 2004; Olabemiwo et al., 2011). However appreciable amounts of PAHs were observed in both raw plantain (0.19 ppm) and roasted plantain (40.33 ppm) in a recent study carried out by Adetunde et al. (2012).

The concentration of PAH in the roasted fish ranges between 0.50 and 14.68 μ g/g; while in suya, it ranges from 1.43 TO 6.48 µg/g. The distribution of average PAH contents in roasted fish and suya are shown in Figure 2. As can be seen from Table1, the average total PAH level of suya (14.83 μ g/g) was lower than that of roasted fish (63.43 µg/g). This could be ascribed to the high fat content of the fish compared to that of beef. Akpan et al. (1994) reported that strong correlation exists between fish lipids and PAH compounds; since PAH compounds are stored in fatty fish tissue. PAHs with maximum concentrations detected in roasted fish was phenanthrene (9.98 µg/g) while benzo(a)anthrancene (7.23 µg/g) was detected as maximum concentration in suya. PAHs with minimum concentrations detected in suya was chrysene (1.43 µg/g) while indeno(1,2,3cd)pyrene with concentration of 0.50 µg/g was detected as minimum concentration in the roasted fish.

In this study, the sum of the average amounts of the low molecular weight PAHs (those containing 2 to 4 aromatic rings) such as naphthalene, acenaphthene, and pyrene, were found higher ($42.48 \ \mu g/g$) than the high molecular weight PAHs ($20.95 \ \mu g/g$), those having 4 to 6 aromatic rings such as benzo(a)anthracene, benzo(a)pyrene [BaP], indeno(1,2,3,cd)pyrene in roasted fish samples. Similar results were obtained by Borokovocova et al. (2005), who determine the levels of PAHs in samples of the food basket of the Czech Republic.

Reports from previous publications have revealed that PAHs with higher molecular weight (HMW) are more carcinogenic than the lower molecular weight (LMW) PAHs (García Falcón et al., 1999; Alonge, 1988). In our study, the HMW carcinogenic PAHs constitute about 33% of the total PAHs in the roasted fish, of which 7% is accounted for by BaP. Surprisingly, benzo (a)pyrene was not detected at all in the suya beef in our study and this is contrary to results obtained by Bababunmi et al. (1982) who reported 8.5 µg/ kg of BaP in suya meat. Duke and Albert (2007) also found BaP contents ranging from 6.5 to 21.5 μ g/ kg in suya meat from 4 different selling points. More recently, Akpambang et al. (2009) reported BaP at levels ranging from 2.4 to 31.2 µg/kg wet weights smoked fish and meat samples. Benzo(a)pyrene is the most studied PAHs, and it is often used as a marker for PAHs

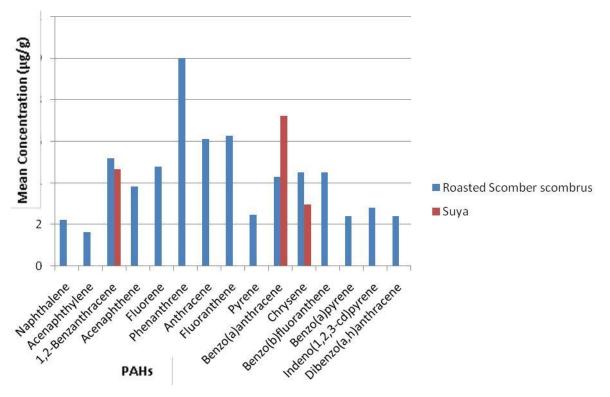


Figure 2. PAH Distribution in Roasted Scomber scombrus and Suya beef.

DAHo	Roasted Scomber scombrus (µg/g)			Suya beef (µg/g)			Plantain	
PAHs	Range	Mean	Raw	Range	Mean	Raw beef	Raw	Roasted
Naphthalene	2.01-2.45	2.23	ND	ND	ND	ND	ND	ND
Acenaphthylene	1.24-1.99	1.62	ND	ND	ND	ND	ND	ND
1,2-Benzanthracene	3.22-7.16	5.19	ND	2.31-6.98	4.65	ND	ND	ND
Acenaphthene	3.44-4.23	3.84	ND	ND	ND	ND	ND	ND
Fluorene	1.81-7.75	4.78	ND	ND	ND	ND	ND	ND
Phenanthrene	5.28-14.68	9.98	ND	ND	ND	ND	ND	ND
Anthracene	2.15-10.08	6.12	ND	ND	ND	ND	ND	ND
Fluoranthene	3.79-8.73	6.26	ND	ND	ND	ND	ND	ND
Pyrene	2.24-2.67	2.46	ND	ND	ND	ND	ND	ND
Benzo(a)anthracene	1.35-7.24	4.30	ND	6.73-7.73	7.23	ND	ND	ND
Chrysene	2.98-6.03	4.51	ND	1.43-4.48	2.95	ND	ND	ND
Benzo(b)fluoranthene	2.69-6.33	4.51	ND	ND	ND	ND	ND	ND
Benzo(a)pyrene	1.65-3.17	2.41	ND	ND	ND	ND	ND	ND
Indeno(1,2,3-cd)pyrene	0.50-5.09	2.80	ND	ND	ND	ND	ND	ND
Dibenzo(a,h)anthracene	1.25-3.58	2.42	ND	ND	ND	ND	ND	ND
Total	35.60-91.26	63.43	ND	10.47-9.19	14.83	ND	ND	ND

Table 1. Summary of PAH Concentrations in Food Samples.

ND=Not detected.

in foods. According to EU Scientific Committee on Food (2002), BaP can be used as an indicator of occurrence, concentration and effect of the carcinogenic HMW PAHs

in foods. The levels of BaP found in roasted *Scomber* scombrus with concentration ranging from 1.65 to 3.17 μ g/g were far higher than the recommended maximum

Element	Scomber scombrus		B	eef	Plantain	
	Frozen(raw)	Roasted	Raw Beef	Suya beef	Raw	Roasted
Pb	0.0013	0.0012	0.0029	0.0027	0.0074	0.0048
Cd	<0.0010	<0.0010	<0.0010	<0.0010	<0.0010	<0.0010
Total Cr	0.0759	0.0991	0.0859	0.0936	0.0651	0.1033

Table 2. Mean Concentration $(\mu g/g)$ of Heavy metals in Food Samples.

permissible concentration of 5.0 μ g/ kg or 0.005 μ g/g fixed for BaP in smoked meat, fish and smoked meat and fishery products (JECFA, 2005).

Sources of polycyclic aromatic hydrocarbons (PAHs) detected in samples

Ratios of fluoranthene (Fla) to pyrene (Pyr) and phenanthrene (Ph) to anthracene (An) are often used to verify the sources of the PAHs detected in the fish samples. Ratio of fluoranthrene to pyrene greater than one (Fla/Pyr>1) is attributed to pyrolytic source while Fla/Pyr<1 is attributed to petroleum hydrocarbon source (Collins et al., 1998). Similarly, ratio of phenanthrane to anthracene less than ten (Ph/An<10) indicates combustion source and Ph/An>10 is attributed to petrogenic source (Benlachenet al., 1997). In our study, the ratios of Ph/An and Fla/Pyr obtained from the roasted fish are in the range of 1.24 to 6.83 and 1.42 to 5.92 respectively. This suggests that the PAHs detected from the fish samples originated from the roasting process.

Levels of Pb, Cd and Total Cr in food samples

The result of the mean concentrations of Pb, Cd and total Cr in the food samples analyzed are presented in Table 2.

Concentrations of Pb varied from 0.0013 to 0.0074 μ g/g in the raw food items and are slightly higher than the levels in the roasted foods. On the contrary, the raw food samples contain lower levels of Cr than in the roasted snacks. Among the three heavy metals determined, the total Cr concentration ranging from 0.0651 to 0.1033 μ g/g, was generally the highest while the Cd level which was found in trace amounts, was the lowest in all the food samples analyzed. Raw plantain has the highest value 0.0074 μ g/g for Pb, while the roasted plantain has the highest value of 0.1033 μ g/g for Cr. The levels of the three metals recorded for *Scomber scombrus* in this study are below the values reported by Asegbeloyin et al, 2010; Atunaya et al, 2011; Oluyemi and Olabanji, 2011.

Levels of the three heavy metals investigated in the food items are generally below the maximum permissible levels set by World Health Organization (Brain and Allen, 1993) for Pb(0.3 ppm); Cd(0.2 ppm) and Cr(0.5 ppm) and hence pose no consumption risk.

Health Implication of polycyclic aromatic hydrocarbons (PAHs)

Studies have shown that eating a charcoal-broiled food may expose one to the same quantity of PAHs as one would receive from smoking 600 cigarettes (Ziegler, 2000). Epidemiological studies carried out by Bababunmi et al (1982), Fritz and Soos (1980), Emerole (1980) and Kazerouni et al. (2001) indicated a statistical correlation between the increased occurrence of cancer of the intestinal tract and frequent intake of roasted food. Our results are corroborated by Alonge (1988) who reported that PAHs are common and may constitute health hazards in Nigeria. The roasted fish as presently consumed by the people of Bayelsa State may therefore create high health risk.

Conclusion

This study has shown that levels of PAHs in roasted fish and suya as sold in Amassoma exceeded the permissible limits. It seems evident from our results that the roasted fish as presently consumed by the people of Bayelsa State may create high health risk. In order to reduce this chemical hazard, it has been suggested that the charred skin of roasted fish, meat or poultry should not be eaten since reports have indicated that PAH levels are significantly higher in the fish skins than in the edible parts. However, levels of the heavy metals investigated in the food items are generally below the maximum permissible limits set by World Health Organization hence one may conclude that the concentration of the metals in the food samples may not be harmful to health.

ACKNOWLEDGMENT

The authors are grateful to the management of Anal Concept Limited, Port-Harcourt for their assistance during the analyses of samples with GC and AAS.

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