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Effect of smoke generation sources and smoke curing duration on the levels of polycyclic aromatic hydrocarbon (PAH) in different suites of fish

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ABSTRACT

The research studied the impact of smoke generation sources on PAH contamination in four different smoke-cured fish (mackerel, sardine, tuna and Cigar minnows). The smoke sources used included acacia, sugarcane bagasse and mangrove. PAHs in the smoke-cured fish were analysed using Varian GC/MS (3800-GC) system. The mean total PAH concentrations in the smoked fish ($n = 108$) ranged from 250.59–1376.09 μg/kg in tuna, cigar minnows, sardine and mackerel smoke-cured with sugarcane bagasse, mangrove and acacia for between 2 and 8 h. The mean BaP levels for most fish cured with smoke from acacia and mangrove for between 2 and 8 h were all above the European Commission set limit of 5.0 μg/kg. Positive correlations (at $P = 0.01$, 2-tailed) were observed between PAH levels in smoked fish and lignin contents of wood type used for the smoke generation, the fat content and the smoke-curing duration. Risk assessment conducted using benzo[a]pyrene carcinogenic and mutagenic toxicity equivalency factors (TEF and MEF respectively) showed high risk associated with consuming fish smoke-cured with hard woods (acacia and mangroves). Sugarcane bagasse was found to be relatively the best and safest smoke-generating source for smoke-curing of fish among the three wood types when using the traditional kiln.

1. Introduction

Traditional smoking or smoke curing of food, meat and fish products has been used since antiquities in many countries (Dore, 1993). The main purpose of smoke curing as applied to fish was to preserve the fish, partly by drying and partly by adding naturally produced anti-microbiological constituents such as phenols from the smoke to the fish (Kramlich et al., 1980; Serden-Basak et al., 2010). However, in recent times it is being used in addition to preservation to achieve characteristic taste and appearance of the smoked fish (Burt, 1988; Hui, 2001; McGee, 2004). In Ghana, fish smoking is the most extensively practiced fish preservation method which make use of the traditional kiln with wood burning temperature of between 300 and 700 °C usually above 80 °C of the oven's temperature (Nti et al., 2002). Statistical data published by Nti et al. (2002) shows that practically all species of fish available in Ghana can be smoked and he estimated that 70–80% of the domestic marine and freshwater catch is consumed in the smoked form.

Food safety is of prior consent in the food processing industry throughout the world. The wholesomeness of smoked fish products using the traditional kiln depends on the following: the type of wood used for the smoking process, the temperature used, the duration of smoking, the type of kiln used, the proximity of the fish from the fire, the type of fish being smoked and the fat content of the fish. It is possible to regulate all these parameters to give quality smoke-cured fish product except for the smoking temperature due to the lack of temperature regulating system on the traditional kiln. Also, the commercial fish mongers have little knowledge on the control of smoking temperature to meet quality standards. This makes it difficult to effectively control smoking temperature, hence the release of toxins like PAHs into the fish product (Philips, 1999; EC-SCF, 2002).

Wood smoke contains a wide range of antioxidant and antimicrobial chemicals such as phenols, aldehydes, acetic acids (Lawrie, 1979; Kjhallstrand and Petersson, 2001) and some toxins like PAHs (Wilson, 1981). PAHs are a class of ubiquitous organic compounds, with some known to be carcinogenic, mutagenic and endocrine disrupting (Yusty and Davina, 2005; EC, 2005; Okuda et al., 2006). PAH generation during smoking may also be affected by the type and composition of wood (EC-SCF, 2002). Thus, smoke generated from "hard" wood may differ in chemical composition from that of the "soft" woods (Maga, 1988; Varlet et al., 2007).

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The basic structural materials of wood cells; cellulose and hemicellulose are aggregate of sugar molecules (linear polysaccharides) which when burnt, effectively caramelize, producing carbonyls which provide most of the colour components and sweet scented aromas (McGee, 2004; Rowell, 2005; Garcia-Perez, 2008) which may be profound with sugarcane bagasse. The wood cells' bonding glue; lignin is a highly complex arrangement of interlocked phenolic molecules which produces a number of distinctive aromatic product when burnt, including smoky, spicy, and pungent (antimicrobial) compounds like guaiacol, syringol, PAHs and phenols (Maga, 1988; Hui, 2001; McGee, 2004; Klemm et al., 2005; Garcia-Perez, 2008). Šimko (2005) stated in his paper that different species of trees have different ratios of components; hence different types of wood smoke may impart different flavour to smoke-cured fish. For instance lignocelluloses compositions of hardwoods are; cellulose (40–50%), hemicellulose (25–35%) and lignin (20–25); Sugarcane bagasse; cellulose (43.6%), hemicellulose (33.5%) and lignin (18.1%), (Sun et al., 2004; PPRIS, 2010).

PAHs produced in wood smokes are known to originate from the thermal pyrolysis (depolymerisation) of lignin and subsequent condensation of the lignin components in lignocelluloses at temperatures above 350 °C (Kawamoto et al., 2007; Nakamura et al., 2008; Garcia-Perez, 2008). Thus, a wood with high lignin content would produce high levels of PAHs at temperatures that favour PAH production (500–900 °C) (Nakamura et al., 2008; Garcia-Perez, 2008). Nakamura et al. (2008) and Garcia-Perez (2008) found that softwood produces higher PAHs than hard wood when burnt at temperatures above 400 °C because of its high lignin content. Nakamura et al. (2008) observed that at temperature of over 400 °C after depolymerisation, it is possible to observe a change from a single aromatic ring system to a multiple ring systems in the solid phase indicating the formation of PAH precursors of char formation. He suggested that the formation of radicals on the aromatic carbons after the hemolytic release of O–CH_3 group may be the starting step for the formation of PAH-like multiple ring system (Nakamura et al., 2008).

According to the latest classification on carcinogenicity of PAHs by International Agency for Research on Cancer (IARC) monograph, it is has been established that benzo[a]pyrene is definite carcinogenic (group 1), dibenz[a,h]anthracene is probably carcinogenic (group 2A), whereas naphthalene, benzo[a]anthracene, chrysene, benzo[b]fluoranthene, benzo[k]fluoranthene, benzo[j]fluoranthene and indenol[1,2,3-c,d]pyrene are classified as possible human carcinogens (group 2B), (IARC, 2012). It is also worth noting that wood smoke has been classified by the IARC (2012) monograph as definite carcinogenic (group 1).

The Cancer Control Division of Ghana Health Service (GHS) estimated that 16,600 cases of cancer occur annually in Ghana with an occurrence rate of about 109.5 cases per 100,000 persons. The report stated that, most of the cases seen in Ghana and other West African countries identified the disease with younger people which are directly opposite to what has been reported in the developed world (GNA, 2011). This means that the continuous consumption of heavily smoke-cured fish may not exempt the consumers from all the possible health cases associated with PAHs.

Regulatory measures on PAHs are not present in most developing countries including Ghana. This therefore calls for research to know which wood type that would be suitable to smoke-cured fish and other product in order to minimise its impact on health. This work seeks to produce smoke from three different wood types to smoke-cure four different fish types using the traditional kiln and to determine the PAHs levels in the smoke-cured fish products. The research further seeks to estimate the risk involved in life time (70 years) ingestion of the smoke-cured products.

2. Material and methods

2.1. The wood used

Smoke from mangrove, sugarcane bagasse and Acacia were used for the smoke curing process due to their availability in the coastal Ghana. They were chosen due to their relatively high usage in smoke-curing of fish among the Ghanaian coastal communities because it is believed to produce high quality fish products. Acacia and Mangroves are classified as hardwoods whereas sugarcane bagasse is classified as grass. In preference, sugarcane bagasse gives attractive colour and sweet flavour to the smoke cured fish products.

2.2. Sample collection and preparation

Four different types of fresh fish samples namely mackerel, sardine, tuna, and Cigar minnows were collected from the Elmina landing beach and were subjected to smoking using the three wood types and the traditional kiln (Chorkor smoker), under same conditions but different time period of smoking. These types of fish were chosen because they are the most staple fish in Ghana. The smoking was done for 2 h and samples taken from it. The rest were further smoked for additional 2 h and again some samples were taken. The remaining samples were further smoked for another 4 h to improve shelf life (Eyabi-Eyabi, 1998; Abolagba et al., 2002; Arthur and Osei-Somuah, 2004). The temperatures of the fires and oven used were recorded using PHYWE digital thermometer at four different points for every 5 min interval of smoking, after initial equilibration for 15 min. This was done for 1 h within smoke-curing durations used as stated in above. The average temperatures of hardwoods fires used reached 345.9–465.8 °C while that of the bagasse reached 289.5–402.3 °C. The average temperature of the kiln used was above 90 °C (about 92–98 °C). Whole smoked fish samples of each wood type were collected on these periods, composited, descaled and homogenized according to fish type for further preparation and extraction prior to analysis using the GC/MS. The average sizes of the smoke-cured fish were as follows; Sardines: 19.56 ± 2.39 cm; Mackerel: 19.98 ± 1.48 cm; Cigar minnows: 19.27 ± 1.63 cm and Tuna: 25.77 ± 2.25 cm. A total of 108 homogenized smoked fish samples and 12 fresh fish samples (control) were analysed. Homogenized Fish samples were kept in amber bottles and refrigerated at temperatures below 4 °C prior to analysis.

2.3. Reagents

Chromatography grade dichloromethane, n-hexane (Purity (GC) ⩾99.0%, Analytical reagent, EC: 203-777-6, Product: 103876Q) and dichloromethane (HPLC grade, 99.8% purity, UN1593 EC: 200-838-9) were purchased from VWR-BDH Chemicals Limited UK. Sodium sulphate (Analytical Reagent, 99.4% purity, product: 28114.296) and glass wool were obtained from VWR-BDH PROLABO UK. Column chromatography Silica gel (mesh: 70–230, product: 36020) was purchased from Auro Avenida Export, PVT Ltd. (India). Methanol (100%, Grade: analytical reagent, Prod: 20847.320) and potassium hydroxide pellet (Purity: 86.1%, Analytical Reagent, Product: 26668.296) were purchased from VWR-BDH PROLABO UK. Petroleum ether (40–60 °C) was also obtained from BDH PROLABO UK. A PAH standard mixture containing 16 PAHs compounds (Purity: 95.9–99.9%, 47940-U) was purchased from SUPELCO-analytical, Bellefonte, PA, USA. A mixture containing four isotopically labelled PAHs namely D10-acenaphthalene, D10- phenanthrene, D12-chrysene, and D10-pyrene internal standard were also purchased from Chemservice, Westchester, PA, USA.

2.4. Dry weight determination (moisture content) and crude lipid content analysis

Standard methods as described by AOAC (1990) were employed in the determination of the moisture and crude lipid contents of the smoke-cured fish.

2.5. Extraction of PAHs

A soxhlet apparatus consisting of 500 mL round bottom flask, an extraction chamber, condenser and water circulators were mounted on a temperature controlled heating mantles for the extractions. 10 g of the smoked fish powder was homogenized in a mortar with about 10 g of Na_2SO_4 until a completely dry homogenate was obtained. The homogenate was carefully transferred into the extraction thimble made from cellulose. The cellulose thimble containing the homogenate was then placed in the extraction chamber of the soxhlet extractor. 50 mL of methanol–KOH mixture prepared by dissolving 6 g of KOH in 12 mL distilled water and making it up to the mark with methanol in 100 mL volumetric flask was added to the homogenate in the extraction chamber. Soxhlet extractions were carried out using 300 mL dichloromethane (DCM). 2.0 mL of isooctane was added to the flask as a keeper. Solvent circulation cycles were at an average of 4 cycles per hour and extraction of each sample was done for 24 h. The extract was cooled to room temperature. The aqueous layer containing the stearate was separated by addition of 100 mL methanol–water mixture (2:8 v/v) using separatory funnel. The organic layer was washed twice with 50 mL distilled water to remove all remaining stearates from the organic extracts. The extract was concentrated using Rotavapor

R-114 at a temperature of 45 °C to about 5 mL. The extracts were further concentrated to about 1 mL using a stream of an inert nitrogen gas (USEPA Method 3540C; Telli-Karakoç et al., 2002; Essumang et al., 2012).

2.6. Post-extraction clean-up

The USEPA Method 3540C's clean-up procedures used by Telli-Karakoç et al. (2002) and Essumang et al. (2012) in similar work were employed in this research.

2.7. GC/MS analysis

A Varian GC/MS (3800 GC) system with 8400 auto-sampler (mass data type: centroid) was used for the analysis. The system was also equipped with 40 m × 0.25 mm × 0.25 μm VF-5 ms fused capillary column. Helium gas was used as the carrier gas. The column head pressure was maintained at 10psi for 15 min with a constant flow rate of 1.0 mL/min. The front injector line was maintained at 250 °C. Injection volumes were 2.0 μL in the splitless mode. The column temperature was initially held at 50 °C for 1 min, and ramped to 320 °C at a rate of 20 °C/min, and then held at 320 °C for 20 min. The mass spectrometer was operated in the ionization mode and spectra were acquired using a mass range of 45 m/z to 450 m/z and automatic gain control. SIM acquisition was carried out by comparison of the base peak of each targeted PAH.

2.8. Analytical quality control

The analytical precision and recovery of the 16 PAHs were checked first with NIST standard reference material (1941b) which is marine sediment collected at the mouth of the Baltimore Harbour intended for use in evaluating analytical methods for the determination of selected PAHs, PCBs congeners and chlorinated pesticides in marine sediments and similar matrices like smoked fish powder. Further analysis of variance (ANOVA) at 95% confidence level for triplicates of smoked fish samples analysed was conducted. To evaluate the instrumental efficiency for the target compounds, recovery studies were carried out using 4 deuterated PAHs, namely acenaphthene-d10 (for naphthalene, acenaphtylene, acenaphthene and fluorene), phenanthrene-10 (for phenanthrene, and anthracene), pyrene-d_{10} (for fluoranthene, pyrene and benz[a]anthracene), and chrysene-d_{12} (for chrysene and the remaining 6).

2.9. Calculating toxicity equivalency quotient (TEQ) and mutagenic equivalent using toxic equivalency factors (TEFs) and mutagenic equivalent factors

Toxic equivalency factors (TEFs) have been developed by a number of researchers and institutions for a number of individual PAHs classified as potential carcinogens. The factor for each of the PAHs expresses its potency relative to benzo[a]pyrene a definite carcinogen, which has a TEF of unity. The concentration of each of the individual PAH compounds is multiplied by its TEF, and these values are summed to yield benzo[a]pyrene equivalent concentrations, TEQ_{BaP} (AFSSA, 2003). By this means, the concentrations of a suite of PAHs can be represented by a single concentration, which reflects the overall carcinogenic potential of the PAHs within the sample for which TEFs have been assigned. This technique has been applied successfully in the past and recent times to smoked and fresh seafood monitoring studies, and other wider monitoring programmes (Law et al., 2002). The mutagenicity of individual PAHs relative to B[a]P had also been computed using the mutagenic factor (MEF) proposed by Durant et al. (1996, 1999). The sum of the concentration of each individual PAH multiplied the corresponding MEF gives the mutagenic equivalents (MEQ). That is;

$$TEQ_{BaP} = \sigma(TEF_i \times C_i) \quad (1)$$

and

$$MEQ_{BaP} = \sigma(MEF_i \times C_i), \quad (2)$$

where C_i is the measured individual PAHs concentrations for the '*ith*' compound with the assigned TEF_i or MEF_i.

The TEF (for TEQ_{BaP}) and MEF (for MEQ_{BaP}) approach has also been adopted because PAH contamination rarely consists of a single compound, but rather of mixtures of compounds that can affect the environment and human health (Engraff et al., 2011; Fisher et al., 2011). The assessment of individual PAHs irrespective of their relative potency is believed to generate inaccurate or underestimated value for carcinogenic and mutagenic risk since it focuses on single compounds. The calculated TEQ_{BaP} and MEQ_{BaP} for the seven USEPA classified carcinogens (mutagens) were used to estimate carcinogenic and mutagenic risk involved in ingestion of smoke-cured fish used herein for life time of 70 years (USEPA, 2000). The total risk due to exposure to mixtures of carcinogenic (or mutagenic) PAHs is:

$$\text{Risk(carcinogenic or mutagenic)} = SF_{BaP} \times \text{BaP equivalent dose of mixtures of PAHs} \quad (3)$$

where SF_{BaP} is the oral carcinogenic slope factor for benzo[a]pyrene (7.3 per mg/kg/day). The BaP equivalent daily dose of compound '*i*' is given as

$$BaP_{EQ}\text{Dose}_i = TEF_i \times \text{Dose}_i \quad (4)$$

Hence the daily BaP equivalent dose of mixtures of carcinogenic (mutagenic) PAH compounds was calculated for carcinogenicity and mutagenicity using the following equation.

$$\text{BaP equivalent dose of carcinogenic(mutagenic)PAHs} = \frac{\text{TEQ(or MEQ)} \times \text{IR} \times \text{EF} \times \text{ED} \times \text{CF}}{\text{BW} \times \text{AT}} \quad (5)$$

These exposure assumptions were made to be consistent with EPA guidance on default assumption on "reasonable maximum exposure" (USEPA, 1991). Where IR is the ingestion or intake rate of carcinogenic (mutagenic) PAHs in μg per day; EF is the exposure frequency to carcinogenic (mutagenic) PAHs in days per year; ED is the exposure duration in years; CF is the conversion factor (i.e. 10^{-6} kg/μg); BW is the average body weight of Ghanaian adult in Kilogram and AT is the average life time of 70 year expectancy. Mean Ingestion rate of 89 ± 31 g/day calculated base on a Mean fish of 46 ± 15 g per meal consumed by the average Ghanaian adult was used. This was obtained through an oral interview conducted. Exposure frequency of 350 day/yr, exposure duration of 30 yr, and average adult body weight of 70 kg were used for the risk assessment.

2.10. Statistical analysis

Microsoft excel's data analysis tool pak and SPSS 16.0 were employed for the Data and statistical analysis. For statistical differences in triplicate analysis evaluation, single-factor ANOVA (One-way ANOVA) was employed (factor: Result from each sample analysis in triplicate analysis; Response measured: PAH levels, μg/kg). For the rest of the statistical differences in means analysis conducted, 2-factor ANOVA (Two-way ANOVA) was employed [factors include: wood type and fish type; wood type and smoke curing duration; smoking duration and fat level (pyrolysis), and Responses measured is PAHs levels, μg/kg; other response measured is fat levels (g) with wood type and fish type as factors].

3. Results and discussion

3.1. Quality control result

There were statistically no significant differences in the PAH results for triplicates ($n = 3$) of each sample at the 95% confidence level. The percentage recovery of each triplicate internal standard analysis ranged from 76 to 106 with an average of 91%. The PAH standard mix was ran to calibrate the instrument and also along with the sample to ensure accurate result (reading). The limit of detection (LOD) and quantification (LOQ) for the individual PAHs ranged from 1.00–2.00 μg/kg to 3.00–6.00 μg/kg respectively. The regression coefficient (R^2) of the PAH standard mix calibration curves over concentration range of 1.00–10.00 μg/mL ranged from 0.9872 to 1.0000. One of the calibration chromatogram showing Peaks with good baseline is presented in Fig. 1.

The second recovery study conducted using the NIST-1941B reference material showed good recovery PAH values ranging from 61% to 103% with an average PAH recovery value of 81%. The values obtained were used to establish the reliability of the extraction system as well as the efficiency of the GC/MS instrument since there was no specific certified reference material for the sample matrix under study at the time of the analysis.

3.2. PAHs levels in fresh fish samples

The PAHs concentrations in fresh fish samples used as control were all below the detection limits (1.00–2.00 μg/kg) except for mackerel where a mean total concentration of 52.82 μg/kg being the contribution of naphthalene (8.95 μg/kg), phenanthrene (0.88 μg/kg), anthracene (0.89 μg/kg) and benzo[b]fluoranthene (42.09 μg/kg) were recorded. This is in conformity with the statement made by Stolyhwo and Sikorski (2005) that fish and marine invertebrates may naturally contain minute amounts of different PAH absorbed from the environment. Rainio et al. (1986) also said in their work that the edible parts of fish from unpolluted seas generally do not contain detectable amounts of B[a]P which also explains the below detection limits recorded for B[a]P in all the

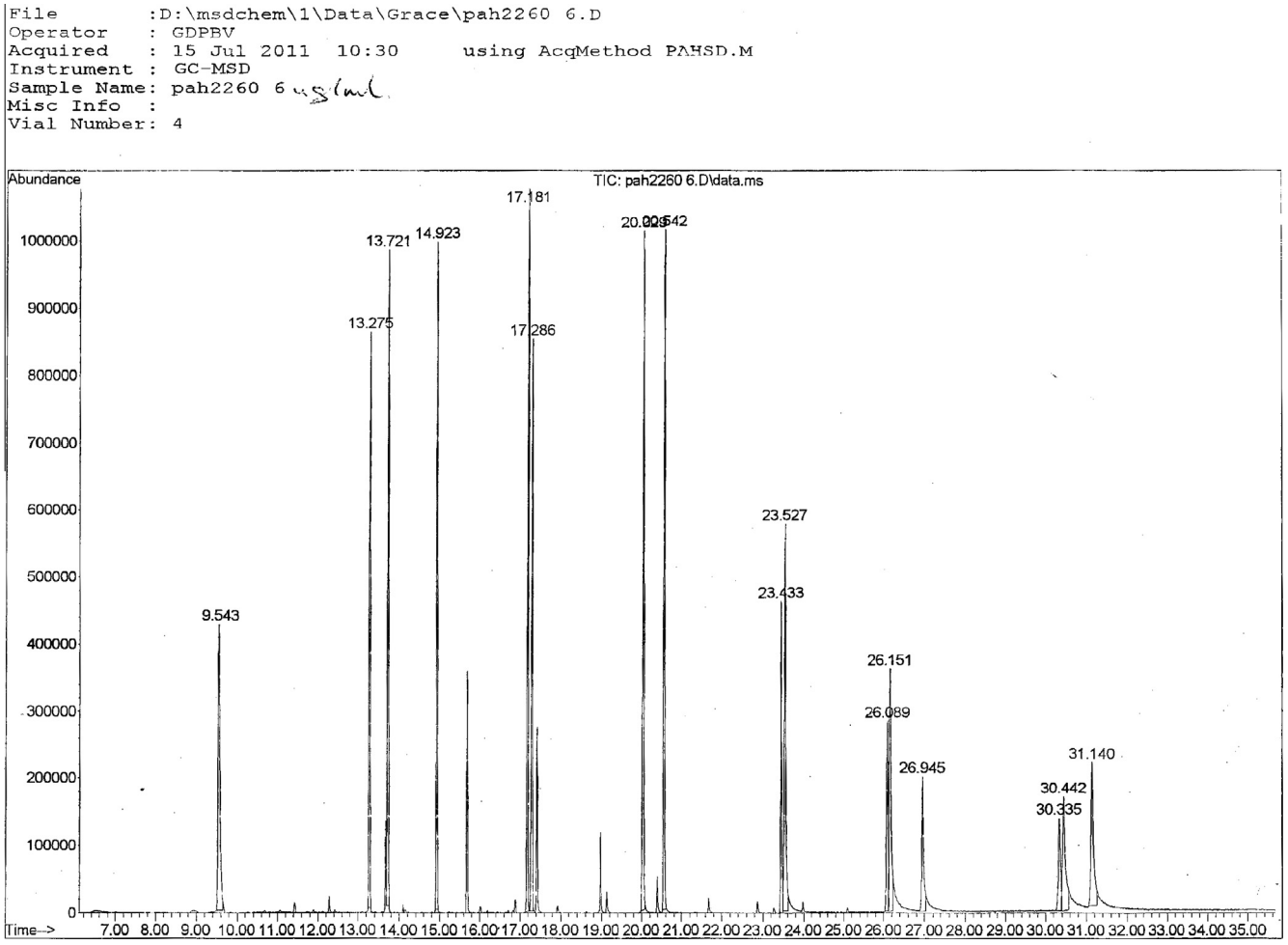

Fig. 1. Chromatogram showing the Peaks of the Calibration Standard with good baseline.

fresh fish samples in this study. It is worth noting that this mean total concentration was corrected for in the mean values for the smoke-cured mackerel samples reported.

3.3. PAHs levels in smoke-cured fish samples

Source assessment analysis in smoke-cured fish on the Ghanaian market conducted suggested combustion as the primary source of PAHs in the fish, with pyrolysis of fat also contributing significantly in samples with high lipid content (Essumang et al., 2012). The mean total PAH concentrations in the smoked fish samples recorded in this work ranged from 250.59 to 1143.51 μg/kg for 2 h of smoke curing. For 4 h of smoke curing, the mean total PAH concentrations ranged from 595.33 to 1315.66 μg/kg. Similarly, for 8 h of smoke curing, the mean total concentrations ranged from 574.97 to 1376.09 μg/kg (Tables 1–3). The individual mean PAH concentrations ranged from below detection limit (1.00–2.00 μg/kg) to 666.16 μg/kg. This maximum mean concentration of 666.16 μg/kg was recorded for benz(a)anthracene in mackerel sample smoked for 8 h with acacia. The individual PAHs found in high levels were those of lower molecular weight while those of higher molecular weight were in most cases below detection limit (1.00–2.00 μg/kg) (Tables 1–3). This could be attributed to the lower average wood temperature range of 345.9–465.8 °C used in the smoking process which is below the noted temperature range of 500–900 °C (Maga, 1988; Nakamura et al., 2008). Temperature range of 500–900 °C are known to favour the production of higher molecular weight PAHs from thermal breakdown of lignin in lignocelluloses during wood combustion (Maga, 1988; Nakamura et al., 2008) and also from pyrolysis of fats in fish (Bartle, 1991; EC-SCF, 2002). Benzo[a]pyrene used as biomarker in monitoring carcinogenic PAHs also recorded mean concentrations from below detection limit to a maximum of 111.90 μg/kg (for acacia wood) in mackerel samples smoke-cured for 8 h (Table 3). This concentration far exceeds the maximum limit of 5.0 μg/kg and 2.0 μg/kg in smoked fish product set by the European Commission (2005) and the Turkish codex regulation (2008) respectively. From the results (Tables 1–3), it was generally observed that Mackerel smoke cured with hardwoods at the various smoking times, recorded B[a]P levels higher than the set limits of 5.0 μg/kg and 2.0 μg/kg for smoked fish product. Also, the mean concentrations of B[a]P; 8.92, 17.61 and 41.62 μg/kg respectively recorded in Cigar minnows, Tuna and Sardine samples smoked for 8 h (Table 3) were all above the set limits. These high levels recorded for B[a]P may pose an elevated cancer risk in consumers and call for immediate setting of limits for search contaminants in smoked fish products by the government of Ghana to protect consumers. The results obtained on the levels of PAHs in smoked fish samples in this work are comparable to that obtained by Stolyhwo and Sikorski (2005), Ciecierska and Obiedzinski (2010), Duedahl-Olesen et al. (2010), and Silva et al. (2011) in similar works.

3.4. Wood type (smoke type) and PAHs levels

From the results (Tables 1–3), the trend of PAH contribution by the different wood types may be summarize as Acacia >

Table 1
Mean PAHs concentrations in μg/kg (dry weight) for fishes smoked with different types of fires for a 2 h ($n = 3$).

Fire type	Acacia				Sugarcane bagasse				Mangroves			
PAH	Sardine	Cigar minnows	Tuna	Mackerel	Sardine	Cigar minnows	Tuna	Mackerel	Sardine	Cigar minnows	Tuna	Mackerel
Naphthalene	397.10	52.00	ND	17.90	15.99	205.21	13.02	93.15	ND	38.74	134.03	117.90
Acenaphthyelene	35.75	47.00	19.50	29.33	27.54	11.04	4.40	ND	79.06	47.66	ND	ND
Acenapthene	106.80	ND	ND	ND	12.00	15.53	3.98	68.49	ND	62.27	ND	ND
Fluorene	81.40	94.64	46.16	85.56	30.09	28.55	8.44	43.49	144.57	21.49	48.20	ND
Phenanthrene	86.50	245.07	113.84	271.37	97.53	44.42	40.58	98.48	308.11	192.09	31.30	71.55
Anthracene	90.30	129.74	60.26	142.57	97.47	45.92	44.64	95.33	154.03	143.37	21.30	71.55
Fluoranthene	10.50	46.15	37.08	96.33	57.96	16.00	25.46	258.68	53.72	3.92	ND	ND
Pyrene	13.90	48.40	36.08	96.33	85.38	16.04	36.47	ND	55.80	3.68	4.85	225.15
Chrysene	ND	ND	ND	167.48	ND	ND	ND	54.89	ND	1.34	ND	ND
Benz(a)anthracene	ND	76.25	ND	53.48	ND	ND	ND	59.76	ND	2.75	ND	42.10
Benzo(b)fluoranthene	4.05	ND	ND	50.96	29.97	70.76	ND	ND	ND	ND	ND	168.38
Benzo(k)fluoranthene	125.30	ND	84.60	126.29	ND	19.07	73.60	97.44	ND	ND	154.50	88.50
Benzo(a)pyrene	5.10	1.35	ND	5.90	ND	ND	ND	ND	2.52	7.40	ND	13.50
Indeno(1,2,3-cd)pyrene	21.65	ND	7.12	ND	ND	ND	ND	4.05	ND	ND	70.60	ND
Dibenz(a,h)anthracene	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Benzo(g,h,i)perylene	41.70	ND	ND	ND	ND	ND	ND	6.17	ND	ND	42.10	ND
Total	1020.05	740.59	404.64	1143.51	453.93	472.54	250.59	879.91	795.29	524.71	506.88	798.63

ND means concentration was below the detection limit used.

Mangroves > sugarcane bagasse. Sugarcane bagasse contributed total PAHs levels as low as one third that of Acacia and one half that of mangrove in most of the smoke-cured fish products for the smoking duration (2–8 h). The low PAH levels in fish smoked with sugarcane bagasse may be attributed to the relatively low lignin to celluloses content of the bagasse as compared to the Acacia and mangroves used in this work (Sun et al., 2004; PPRIS, 2010). Also, this may be attributed to the high oxygen content of the lignocelluloses in the bagasse which made smoking possible even at relatively low burning temperature of 289.5–402.3 °C as well as in oxygen restricted system (low burning temperature system). The high PAH levels in fish smoked with Acacia compared to other smoke generation sources used may be attributed to the high lignin content of the wood (Acacia) (PPRIS, 2010) which makes it to burn hot (averagely 345.9–465.8 °C) hence producing high PAH levels (Houck and Tiegs, 1998) than sugarcane bagasse and mangrove.

Though mangroves is classified as a hardwood, it is believed to exhibit malfunctioning water transport tissue, which implies reduced lignin content (Wilson and Fischer, 2011; Tyree and Zimmermann, 2002) since it only survives in water logged area. This reduced cell lignin may have contributed to the reduced PAH levels in fish smoked with Mangrove as compare to that of Acacia. Strong positive correlations (>0.6) were observed for PAH levels in smoked fish (Table 1–3) and the known lignin content of wood used for the smoking process (Sun et al., 2004; PPRIS, 2010) at the 95% confidence level. Analysis of variance conducted at the 95% confidence level (CL) on data showed significant difference ($P < 0.05$) between wood types (smoke type) with respect to PAH levels in each fish type smoked. This implied, different smoke

Table 2
Mean PAHs concentrations in μg/kg (dry weight) for fishes smoked with different types of fires for 4 h ($n = 3$).

Fire type	Acacia				Sugarcane bagasse				Mangroves			
PAH	Sardine	Cigar minnows	Tuna	Mackerel	Sardine	Cigar minnows	Tuna	Mackerel	Sardine	Cigar minnows	Tuna	Mackerel
Naphthalene	57.52	ND	ND	17.90	65.31	87.79	46.04	207.25	235.32	144.53	291.90	198.80
Acenaphthyelene	120.00	78.85	79.18	86.13	48.77	27.45	28.61	ND	104.27	18.40	ND	ND
Acenapthene	34.98	ND	ND	ND	14.90	17.77	7.57	ND	33.10	13.85	17.20	79.50
Fluorene	259.90	171.84	174.84	161.30	27.50	33.47	37.40	49.13	64.70	49.47	87.90	43.90
Phenanthrene	457.00	313.80	344.91	322.60	168.96	88.53	172.43	121.67	56.43	149.38	26.90	58.20
Anthracene	257.20	164.05	180.28	170.74	169.46	88.51	172.16	174.50	63.65	156.80	51.80	60.80
Fluoranthene	47.95	70.53	ND	144.05	126.05	44.43	17.08	ND	11.10	26.67	75.50	100.00
Pyrene	48.02	76.01	22.45	153.82	120.59	62.53	91.71	184.22	30.02	62.63	51.50	64.00
Chrysene	3.66	ND	ND	84.73	1.88	ND	40.72	113.02	9.27	ND	ND	ND
Benz(a)anthracene	3.87	ND	ND	ND	3.56	17.64	29.12	68.89	40.37	ND	99.60	106.80
Benzo(b)fluoranthene	0.53	5.09	2.25	168.38	1.13	15.71	ND	1.26	74.65	ND	ND	84.19
Benzo(k)fluoranthene	0.68	ND	ND	ND	1.26	97.09	11.72	ND	82.85	38.40	ND	135.90
Benzo(a)pyrene	3.04	ND	ND	6.01	2.53	ND	ND	1.59	2.27	1.24	ND	22.28
Indeno(1,2,3-cd)pyrene	ND	ND	ND	ND	ND	14.41	ND	ND	61.50	ND	ND	79.70
Dibenz(a,h)anthracene	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	44.53	ND
Benzo(g,h,i)perylene	ND	ND	ND	ND	0.08	ND	ND	ND	ND	ND	ND	ND
Total	1294.35	880.17	803.91	1315.66	751.96	595.33	654.56	921.51	869.48	661.37	746.83	1034.07

ND means concentration was below detection limit used.

generation sources (wood type) imparted significantly different amount of PAHs to each fish smoked.

3.5. Fish type and PAH Levels

The different fish species had different PAH (levels) contribution from the smoking process. This could be attributed to the differences in fat and moisture contents, the mussel arrangement and the nature of skin cover (Maga, 1988). Smoked mackerel on the average recorded the highest mean levels of PAHs for all the wood types and durations of smoking followed by sardine whereas smoked Cigar minnows and tuna recorded the least levels of PAHs (Tables 1–3). The moisture content showed a reverse pattern as that of the lipid content in the smoked fish (Appendix 1). Thus Tuna recorded the highest mean moisture content, followed by Cigar minnows, then by sardine with mackerel recording the least moisture content. The high mean levels of PAHs, especially benzo[a]pyrene in smoked mackerel and sardine may partly be attributed to the high lipid contents in the fish which might have been pyrolysed back into the fish during the smoking period, even at 200 °C (EC-SCF, 2002). The pyrolysis of lipid turns to produce more PAHs in addition to that emanating from the wood combustion. The relatively low level of PAHs obtained in smoked tuna may also be attributed to its relatively high moisture content but low lipid content as compared to the other smoked fish used in the study. It was also observed that the longer the smoking period, the higher the reduction in the lipid content of the fish through pyrolysis (Appendix 2). This may have contributed a significant amount of PAHs in fish with higher lipid contents like mackerel and sardine during a lengthy period of smoke curing (Appendix 2; Table 3). These results are comparable with the results from Varlet et al. (2007) and Duedahl-Olesen et al. (2010) in a similar research.

Interrelation analysis conducted on the result (Tables 1–3 and Appendix 2) using SPSS 16 at 99% ($p = 0.01$) CL (2-tailed), showed strong correlations (+0.65–1.0) between the levels of PAHs and the lipid content of the smoked fish. This result is comparable to the result obtained by Serden-Basak et al. (2010) in similar work. Two-way ANOVA analysis conducted at 95% confidence level on the crude lipid data showed no statistical significant differences ($P > 0.05$) between wood types in fat pyrolysis with respect to each fish type smoked. Which implied that the amount of PAHs contributed through pyrolysis of fat in smoked fish is independent of the firewood type used. This may be attributed to the fact that lipid pyrolysis usually start at mean temperature of 200 °C and is favoured in the production of PAHs at temperatures ⩾500 °C (Bartle, 1991; EC-SCF, 2002) but the fire temperatures used in this work ranged from 289.5 to 465.8 °C. The low levels of PAHs in smoked tuna may also be attributed to the highly compacted fillets of tuna species as compared to the other fish species analysed. This subsequently reduced the penetration of PAHs into the fish mussels (Maga, 1988). Further analysis of variance (two-way ANOVA) conducted on the data at 95% CL showed significant difference ($P < 0.05$) in PAH levels between fish type with respect to all the wood type used and the smoke curing duration. Thus PAH levels in smoked fish are also species dependent.

3.6. Smoking duration and PAH Levels

The time used in smoke curing is known to have significant influence on both the quality and the levels of PAHs in smoked fish (Varlet et al., 2006; 2007). According to the local fish mongers, longer smoking time is known to improve on the shelf life of the fish by reducing significantly the moisture and lipid content which would otherwise cause rancidification and spoilage of the smoke-cured fish (McGee, 2004). However, this turns to increase significantly the PAH levels of the smoke cured fish. From the result (Appendix 1 and 2), there was indeed a significant decrease in both the moisture and lipid content of the smoked fish with respect to long period of smoking for all the wood types used. Fish smoked for 2 h had the highest levels of moisture and lipid, whilst the least was recorded for samples smoked for 8 h. On the contrary, less PAH accumulation occurred for less smoking period (Appendix 1, 2; Tables 1–3) for all the wood types used. A similar trend was observed by Varlet et al. (2007) and Duedahl-Olesen et al. (2010) in their work. In some few cases, the PAH levels in fish smoked for 8 h were insignificantly lower than that smoked for 4 h. This may be attributed to the fact that PAHs adsorbed on fish surfaces were either easily detached or converted to volatile ones and released into the surroundings other than the fish when being heated for long time. Statistical correlation conducted on the data (SPSS) showed a strong positive correlations (>0.90) between PAH levels and the smoking duration at the 99% CL ($P = 0.01$ for 2-tailed). Thus increasing the smoking duration may increase the PAH levels in the final smoke cured product.

Table 3
Mean PAHs concentrations in μg/kg (dry weight) for fishes smoked with different types of fires for 8 h ($n = 3$).

Fish type	Acacia				Sugarcane bagasse				Mangroves			
PAHs	Sardine	Cigar minnows	Tuna	Mackerel	Sardine	Cigar minnows	Tuna	Mackerel	Sardine	Cigar minnows	Tuna	Mackerel
Naphthalene	73.93	ND	62.37	55.52	55.53	17.81	157.91	162.10	ND	56.89	385.00	231.13
Acenaphthyelene	52.58	84.50	54.43	ND	49.93	ND	ND	62.70	28.38	58.78	ND	144.67
Acenapthene	29.79	10.56	2.57	5.72	14.12	5.22	121.49	36.90	ND	15.42	144.80	46.67
Fluorene	66.64	160.13	109.29	12.72	22.42	18.22	37.54	84.10	118.05	10.60	49.20	125.80
Phenanthrene	293.04	331.93	218.36	22.60	272.81	21.87	25.95	142.40	147.61	293.39	69.20	104.67
Anthracene	262.28	175.69	115.58	23.04	274.26	26.89	33.98	86.70	78.13	295.03	90.60	105.00
Fluoranthene	84.77	52.94	ND	314.13	50.56	ND	219.25	ND	293.63	48.12	ND	22.20
Pyrene	97.08	52.25	20.25	122.30	48.22	396.80	4.46	74.80	241.49	48.22	ND	67.40
Chrysene	30.22	ND	ND	ND	5.37	ND	ND	ND	ND	3.49	ND	18.53
Benz(a)anthracene	30.01	ND	46.82	666.16	5.23	57.57	ND	89.60	ND	3.68	ND	43.20
Benzo(b)fluoranthene	ND	67.40	ND	42.00	1.21	ND	ND	117.00	ND	0.52	ND	168.38
Benzo(k)fluoranthene	ND	53.85	ND	ND	ND	18.46	27.67	165.00	0.50	0.66	ND	57.80
Benzo(a)pyrene	41.62	ND	17.61	111.90	1.54	8.92	ND	2.20	ND	ND	ND	9.87
Indeno(1,2,3-cd)pyrene	50.72	ND	ND	ND	ND	3.23	ND	ND	ND	ND	ND	ND
Dibenz(a,h)anthracene	2.78	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Benzo(g,h,i)perylene	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Total	1115.46	989.25	647.28	1376.09	801.19	574.97	628.24	1023.50	907.78	834.80	738.80	1145.31

ND means concentration was below detection limit used.

Table 4
Average PAHs mean concentration in μg/kg (dry weight) in smoked fish samples for all time smoke curing with respect to fire types ($n = 9$).

Fire type	Acacia				Sugarcane bagasse				Mangrove			
PAH	Sardine	Cigar minnows	Tuna	Mackerel	Sardine	Cigar minnows	Tuna	Mackerel	Sardine	Cigar minnows	tuna	Mackerel
Naphthalene	176.18	17.33	20.79	30.44	45.61	103.60	72.32	154.17	78.44	80.05	270.31	182.61
Acenaphthyelene	69.44	70.12	51.04	38.49	42.08	12.83	11.00	20.90	70.57	41.61	0.81[a]	48.22
Acenapthene	57.19	3.52	0.86	1.91	13.68	12.84	44.35	35.13	11.03	30.51	54.00	42.06
Fluorene	135.98	142.20	110.10	86.53	26.67	26.74	27.79	58.91	109.11	27.19	61.77	56.57
Phenanthrene	278.85	296.93	225.70	205.52	179.77	51.61	79.65	120.85	170.72	211.62	42.47	78.14
Anthracene	203.26	156.49	118.71	112.12	180.39	53.77	83.59	118.84	98.60	198.40	54.57	79.12
Fluoranthene	47.74	56.54	12.36	184.84	78.19	20.14	87.26	86.23	119.48	26.24	25.17	40.73
Pyrene	53.00	58.89	26.26	124.15	84.73	158.46	44.21	86.34	109.10	38.18	18.78	118.85
Chrysene	11.29	0.68[a]	0.68[a]	84.07	2.41	0.68[a]	13.57	55.97	3.09	1.61	0.68[a]	6.18
Benz(a)anthracene	11.29	25.42	15.61	239.88	2.93	25.07	9.71	72.75	13.46	2.14	33.20	64.03
Benzo(b)fluoranthene	1.53	24.16	0.75	87.11	10.77	28.82	0.83[a]	39.42	24.88	0.17	0.83[a]	140.32
Benzo(k)fluoranthene	41.99	17.95	28.20	42.10	0.42	44.87	37.66	87.48	27.78	13.02	51.50	94.07
Benzo(a)pyrene	16.59	0.45	5.87	41.27	1.36	2.97	0.62[a]	1.26	1.60	2.88	0.62[a]	15.22
Indeno(1,2,3-cd)pyrene	24.12	1.17[a]	2.37	1.17[a]	1.17[a]	5.88	1.17[a]	1.35	20.50	1.17[a]	23.53	26.57
Dibenz(a,h)anthracene	0.93	0.69[a]	0.69[a]	0.69[a]	0.69[a]	0.69[a]	0.69[a]	0.69[a]	0.69[a]	0.69[a]	14.84	0.69[a]
Benzo(g,h,i)perylene	13.90	0.67[a]	0.67[a]	0.67[a]	0.70[b]	0.67[a]	0.67[a]	2.06	0.67[a]	0.67[a]	14.03	0.67[a]
Total	1143.29	873.21	620.65	1280.95	670.62	547.61	515.11	942.33	859.72	676.16	667.11	994.03

[a] Means values were calculated base on one-half the limit of detection and multipliers. These values are incorporated for used in risk and TEQ calculations.
[b] Means was detected but average of mean value fell below detection limit. Hence was recalculated using detection limit and multiplier.

3.7. Risk assessment for PAHs in smoked fish

The carcinogenic toxicity (TEQ_{BaP}) and mutagenic toxicity (MEQ_{BaP}) relative to B[a]P were calculated for the carcinogenic and mutagenic risk associated with ingestion of the smoke-cured fish (Tables 4 and 5). While TEQ_{BaP} is directly associated with carcinogenicity, MEQ_{BaP} (mutagenic activity) may not be directly associated with cancer (Zeiger, 1998, 2001) and may have implications for other non-cancerous adverse health effects like pulmonary diseases, birth defects, impotency, low IQ etc.(DeMarini et al., 2004; Seagrave et al., 2002). From the result (Table 6), the total toxicity equivalencies for the seven USEPA priority carcinogens ranged from 2.871 for tuna smoked with sugarcane bagasse to 75.281 for mackerel smoked with acacia (2–8 h smoking duration). The high TEQ-BaP calculated for mackerel smoked with acacia compared to other smoke generation sources used, signified that ingestion of this smoke-cured product may pose a high carcinogenic risk. The corresponding BaPEQ daily dose and carcinogenic risk for an adult involved in life time of 70 years ingestion of the smoked products were also calculated to be 1.50 and 39.34 μg/kg per day for a risk of 1.1×10^{-5} and 2.9×10^{-4} respectively (Table 6). These risk values mean that for a tuna smoked with sugarcane bagasse ingestion, 1 out of 100,000 adults are likely to suffer from cancer in their life time and for ingestion of mackerel smoked with acacia, 29 out of 100,000 people are likely to suffer from cancer in their life time. This means that the consumption of tuna smoked with sugarcane bagasse may pose very low risk, because it is just equal to the USEPA (1993, 2009) carcinogenic unit risk of 1×10^{-5} (carcinogenesis threshold). Generally, relatively higher ΣBaP-TEQ and cancer risk values above the acceptable USEPA (1993, 2009) unit risk were recorded for all the four fish samples smoked with Acacia while that smoked with sugarcane bagasse recorded the least though values were just above the USEPA (1993, 2009) unit risk of 10^{-5}.

Also, the mutagenic equivalent for these PAHs calculated ranged from 4.649 for sardine smoked with sugarcane bagasse to 89.341 for mackerel smoked with acacia (2–8 h smoking duration) (Table 6). The corresponding BaP_{EQ} daily doses were also calculated to be 2.429 and 46.681 μg/kg per day for sardine and mackerel respectively. Hence, the mutagenic risk involved in ingestion of these smoked sardine and mackerel for a life time of 70 years were calculated to range from 1.8×10^{-5} to 3.4×10^{-4} respectively. This imply that for adult's life time ingestion of sardine smoked with sugarcane bagasse, and mackerel smoked with acacia; 2 out of 100,000 and 34 out of 100,000 people are like to suffer from non-cancer and other cancer related disease in their life time respectively. Generally, relatively higher ΣBaP-MEQ and mutagenic risk values above the acceptable USEPA (1993, 2009) unit risk were recorded for all the four fish samples smoked with Acacia while that smoked with sugarcane bagasse recorded the least though values were just above the USEPA (1993, 2009) unit risk of 10^{-5}.

These high risks associated with ingestion of fish smoked with acacia may render the wood type unfavourable for smoke curing of fish while sugarcane bagasse may be considered quite suitable for smoke curing amongst the three wood types used herein. Smoked mackerel recorded the highest ΣBaP-TEQ and ΣBaP-MEQ values with risk levels far exceeding the USEPA moderate unit risk level of 1×10^{-5} (USEPA, 1993, 2009) among the smoked fishes for all the wood types used. This was on the average followed closely by values recorded for smoked sardine, with Cigar minnows and tuna recording comparatively lower risk values. From these results it may be said that mackerel smoke-cured with hard woods have high cancer and mutagenic risk and may hence be considered unsafe for consumption. Tuna, sardine and Cigar minnows smoked with the hardwoods may also pose some moderate level of carcinogenic and mutagenic risk

Table 5
Proposed benzo(a)pyrene equivalent factors for carcinogenic (TEF) and mutagenic toxicity (MEF).

PAH	TEF USEPA (1993)	MEF Durant et al. (1996, 1999)
Chrysene	0.001	0.017
Benz(a)anthracene	0.100	0.082
Benzo(b)fluoranthene	0.100	0.250
Benzo(k)fluoranthene	0.010	0.110
Benzo(a)pyrene	1.000	1.000
Indeno(1,2,3-cd)pyrene	0.100	0.310
Dibenz(a,h)anthracene	1.000	0.290

Table 6
Risk assessment based on carcinogenic and mutagenic equivalency (μg/kg), calculated using TEF and MEF for samples smoked with 3 different types of woods for all time (n = 9).

Carcinogenic equivalency	Acacia				Sugarcane bagasse				Mangrove			
	Sardine	Cigar minnows	Tuna	Mackerel	Sardine	Cigar minnows	Tuna	Mackerel	Sardine	Cigar minnows	Tuna	Mackerel
chrysene	0.011	0.001	0.001	0.084	0.002	0.001	0.014	0.056	0.003	0.002	0.001	0.006
benz(a)anthracene	1.129	2.542	1.561	23.988	0.293	2.507	0.971	7.275	1.346	0.214	3.320	6.403
benzo(b)fluoranthene	0.153	2.416	0.075	8.711	1.077	2.882	0.083	3.942	2.488	0.017	0.083	14.032
benzo(k)fluoranthene	0.420	0.180	0.282	0.421	0.004	0.449	0.377	0.875	0.278	0.130	0.515	0.941
benzo(a)pyrene	16.587	0.450	5.870	41.270	1.358	2.973	0.620	1.262	1.597	2.879	0.620	15.216
indeno(1,2,3-cd)pyrene	2.412	0.117	0.237	0.117	0.023	0.588	0.117	0.135	2.050	0.117	2.353	2.657
dibenz(a,h)anthracene	0.927	0.690	0.690	0.690	0.690	0.690	0.690	0.690	0.690	0.690	14.840	0.690
ΣBaP-TEQ	21.639	6.395	8.716	75.281	3.447	10.090	2.871	14.234	8.452	4.050	21.732	39.944
BaPEQ daily dose $(\mu g/kg)day^{-1}$	11.306	3.342	4.554	39.335	1.801	5.272	1.500	7.437	4.416	2.116	11.355	20.871
Carcinogenic risk	8.3E−05	2.4E−05	3.3E−05	2.9E−04	1.3E−05	3.9E−05	1.1E−05	5.4E−05	3.2E−05	1.5E−05	8.3E−05	1.5E−04
Mutagenic equivalency												
chrysene	0.192	0.012	0.012	1.429	0.041	0.012	0.231	0.951	0.053	0.027	0.012	0.105
benz(a)anthracene	0.926	2.084	1.280	19.670	0.240	2.056	0.796	5.965	1.103	0.176	2.722	5.251
benzo(b)fluoranthene	0.382	6.041	0.188	21.778	2.693	5.897	0.208	9.855	6.221	0.043	0.208	35.079
benzo(k)fluoranthene	4.619	1.975	3.102	4.630	0.046	4.936	4.143	9.623	3.056	1.432	5.665	10.347
benzo(a)pyrene	16.587	0.450	5.870	41.270	1.358	8.209	0.620	1.262	1.597	2.879	0.620	15.216
indeno(1,2,3-cd)pyrene	7.478	0.363	0.736	0.363	0.071	0.334	0.363	0.418	6.355	0.363	7.295	8.236
dibenz(a,h)anthracene	0.269	0.200	0.200	0.200	0.200	1.393	0.200	0.200	0.200	0.200	4.304	0.200
ΣBaP-MEQ	30.453	11.124	11.387	89.341	4.649	22.836	6.560	28.274	18.585	5.121	20.825	74.433
BaPEQ daily dose $(\mu g/kg)day^{-1}$	15.912	5.812	5.950	46.681	2.429	10.035	3.423	14.773	9.711	2.676	10.881	38.892
Mutagenic Risk	1.2E−04	4.2E−05	4.3E−05	3.4E−04	1.8E−05	7.3E−05	2.5E−05	1.1E−04	7.1E−05	2.0E−05	7.9E−05	2.8E−04

ΣBaP-TEQ and ΣBaP-MEQ are the total benzo[a]pyrene toxicity equivalents for carcinogenicity and mutagenicity respectively.
BaPEQ = benzo[a]pyrene toxicity equivalent.

because the values recorded were just higher or equal to the USEPA unit risk, with the least risk coming from samples smoke-cured with sugarcane bagasse as shown in Table 6.

3.8. Conclusion

The results showed that fish smoked with acacia (hardwood) had elevated levels of PAH contamination with an associated elevated cancer and mutagenic risk posed when ingested by consumers. This implies the use of acacia for both short and long duration in smoke curing of fish using the traditional kiln (Chorkor smoker) may be an unsafe practice. Alternative use of mangrove (hardwood) for smoke curing of fish may be appropriate but must be done at short smoking time duration of at most 4 h, which may be enough to give the smoke-cured fish the required good shelf life. Generally, it may be said that fish smoke-cured with any of the hardwoods for a longer duration ($\geqslant$4 h) using the traditional kiln may be unsafe for consumption. Also, it could be said that smoke curing of fish with sugarcane bagasse using the traditional kiln may be the safest and the best fish smoking practice at short time duration. A comparatively low ΣTEQ-BaP and ΣMEQ-BaP values were obtained for samples smoke-cured with sugarcane bagasse. This on the average showed little carcinogenic and mutagenic risk compared to mangrove and acacia. Mackerel and sardines happened to accumulate more of the PAHs due to their high lipid content. This showed high TEQs-BaP and MEQ-BaP values suggesting elevated carcinogenic and mutagenic risk as calculated. It could therefore be said that, there may be elevated risk of cancer and non-cancer diseased associated with life time (70 years) consumption of mackerel and sardine smoke-cured with hardwoods especially at longer smoking durations because of their high lipid contents. Hence, it may be safe to discourage the consumption of these fish when smoke-cured using this unsafe practice. Generally, it could be said that it may be safe to consume tuna smoke-cured with hardwoods or sugarcane bagasse for a short smoking time duration using the traditional kiln. Sugarcane bagasse, the best smoke generator in this work is very abundant in Ghana as a waste product from traditional alcohol production. Its use for all time smoke generation for smoke curing of fish should be encouraged by the government of Ghana and the agencies involved. This may help minimize the risk associated with consuming fish smoked with hardwoods using traditional kilns and reduce the cost involved in collection and disposal of the large volumes of sugarcane bagasse generated as waste daily by the local alcohol industries in Ghana.

Conflict of Interest

The authors declare that there are no conflicts of interest.

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Appendix A. Supplementary material

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.fct.2013.04.014.

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