Effect of two fish smoking ovens on the nutritional composition and PAH content of smoked fish.

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Abstract

The quality of three marine fish species smoked with the Abuesi gas fish smoker (gas smoker) and the chorkor smoker (wood smoker) were analysed. The aim was to evaluate the nutritional composition and Polycyclic Aromatic Hydrocarbon (PAH) levels in the smoked fish of the two smokers. The results of the analysis showed that the protein, moisture, fat, total carbohydrate and ash content of the gas smoked samples were in the range of 54.23%-70.32%, 13.69%-24.73%, 5.28%-8.76%, 2.97%-13.21% and 2.77%-4.27% respectively, while that of the wood smoked samples were in the range of 43.38%-43.75%, 22.00%-46.5%, 2.00, 5.26%-25.78%, 2.85%-6.50% respectively. The total PAH concentration also ranged between $321.7 \mu g/kg$ - $514.41 \mu g/kg$ in the gas smoked samples and $1038.8 \mu g/kg$ - $1550 \mu g/kg$ in the wood smoked samples. The EU maximum residue limits (MRLs) for PAH4 and BAP were met in the Abuesi gas fish smoker samples but not in the chorkor smoker samples. It is concluded that the Abuesi gas fish smoker is a better choice, in terms of fish quality, for the smoking of fish than the chorkor smoker.

Keywords: Nutritional composition, Polycyclic Aromatic Hydrocarbons, Abuesi gas fish smoker, Chorkor smoker, Smoked fish

Abbreviation: HPLC: High Performance Liquid Chromatography; GRATIS: Ghana Regional Appropriate Technology Industrial Service; PAH: Polycyclic Aromatic Hydrocarbons; FAO: Food and Agricultural Organization; LPG: Liquefied Petroleum Gas

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Introduction

The highly susceptible nature of fish to early deterioration requires that it is properly handled and preserved immediately after harvesting, in order to remain fit for human consumption. Globally, various methods such as freezing, cooling, drying, salting, smoking and canning are used for fish preservation. In Ghana and the rest of West Africa, smoking is the most practiced method of fish preservation often at temperatures above 800°C, which is able to cook the fish. An estimated 70-80% of marine and freshwater catch is consumed in the smoked form [1] and practically all species of fish in the country can be smoked.

Over the years, traditional smoking ovens have been used for fish smoking but have evolved with time. These ovens include: the cylindrical metal oven, the rectangular mud oven, the square metal oven and the relatively recent chorkor smoker. Other improved fish smokers such as the Ahotor Oven, FTT-Thiaroye and the Abuesi gas fish smoker have also been introduced along the line. Traditional fish smoking methods involve treating of pre-salted fish with smoke that comes into contact with the fish from incomplete combustion of wood [1]. This way of fish smoking, which also involves thermal treatments at high temperatures, denature the proteins in fish, which is the major reason for its consumption. Idah and Nwankwo (2013) have also stated that excessively high smoking temperatures can affect the nutritional content of smoke dried fish and thus emphasized the importance of ascertaining the effect of smoke drying temperatures on the nutritional properties of the finished product. Direct contact of fish with combustion gases has also been reported to introduce high levels of Polycyclic Aromatic Hydrocarbons (PAHs) into the finished product [2].

PAHs are complex chemical compounds formed and released during incomplete combustion or pyrolysis of organic matter. They are well known for their harmful effects on human health with some known to be carcinogenic. Studies in experimental animals on individual PAHs, mainly on benzo[a]pyrene, have shown various toxicological effects such haematological effects, reproductive and developmental toxicity and immunotoxicity (Food Safety Authority of Ireland, 2015). The International Agency for Research into Cancer, in 2012 concluded that benzo[a]pyrene is a human carcinogen. Some other PAHs have also been identified as carcinogens with possible genotoxic (DNA-damaging) properties (Food Safety Authority of Ireland, 2015). Essumang et al. suggested that high consumption levels of smoked fish could be attributed to the increase in cancer cases in Ghana, especially breast cancer in the older female population [3].

PAHs are lipophilic in nature. They usually accumulate in the fatty tissues of organisms and as such are produced in the fatty tissues of fish during smoking at higher temperatures. The formation of PAHs is known to occur through pyrolysis of fat at temperatures above 200°C and it is highly stimulated at temperatures over 700°C [4]. More fatty fish thus tend to accumulate more PAHs than less fatty fish.

In order to reduce the adverse effects of PAHs in smoked fish on the health of consumers, maximum levels of PAHs have been set for smoked fish and smoked fish products. In 2005, the European Commission (EC) introduced for benzo[a]pyrene (chosen as a marker of the occurrence and carcinogenic potency of the entire class of carcinogenic and genotoxic PAHs) a maximum level of 5 μ g/kg in smoked fish and meat (Commission Regulation *Citation:* Nunoo FKE, Tornyeviadzi E, Asamoah EK, et al. Effect of two fish smoking ovens on the nutritional composition and PAH content of smoked fish. J pub health catalog. 2018;1(1):5-10.

2005/208/EC) [5]. Based on new available data in 2008, the EFSA CONTAM Panel concluded that benzo[a]pyrene was not a suitable indicator for the occurrence of PAHs in food and that 8 high molecular weight PAHs (PAH8), or a subgroup of 4 PAHs (PAH4), are the most suitable indicators of PAHs in food (EFSA, 2008). In accordance with these findings, new maximum levels were set for benzo(a)pyrene at 2.0 µg/kg and PAH4 (the sum of benzo[a]pyrene, benz[a]anthracene, benzo[b] fluoranthene and chrysene) or PAH8 (the sum of benzo[a] pyrene, benz[a]anthracene, benzo[b]fluoranthene, benzo[k] fluoranthene, benzo[ghi]perylene, chrysene, dibenz[a,h] anthracene and indeno[1,2,3-cd]pyrene) at 12.0 µg/kg, with PAH8 not providing much added value compared to PAH4 (Commission regulation (EC) 835/2011). These maximum levels are set at very low levels (as low as reasonably achievable) [6].

The high levels of PAHs in traditionally smoked fish along with other factors such as the large quantities of fuel wood consumed and high temperatures involved in the smoking process, which affects the nutritional quality of the finished products; has directed the development of improved fish smokers over the years.

The aim of this study is to compare the effect of one of these improved fish smokers, the Abuesi gas fish smoker, developed by Ghana Regional Appropriate Technology Industrial Service (GRATIS) Foundation in collaboration with the fish processors association of Abuesi, which uses Liquefied Petroleum Gas as the source of fuel and wood chirpings to provide smoke; and the common traditional fish smoker in Ghana, the chorkor smoker, which was developed by the Food Research Institute of Accra, Ghana, assisted by Food and Agricultural Organization of the United Nations (FAO) on the quality of smoked fish produced [7].

Materials and Methods

Sample collection

Raw fish samples of three different species common in Ghana, namely Barracuda (*Sphyraena sphyraena*, Sphyraenidae) (Linnaeus, 1758), Common White Grouper (*Epinephelus aeneus*, Serranidae) (Geeoff. St. Hill, 1809), and Yellowfin Tuna (*Thunnus albacares*, Scombridae) (Bonnat, 1788) were purchased from fishmongers at Abuesi, near Takoradi in the Western region and Tema fishing harbour in Ghana in September 2016. These species were chosen because they are commonly caught and consumed in smoked form in the Western Region of Ghana.

Fish smokers

Chorkor smoker: The chorkor smoker is an improvement on a traditional fish smoking oven already known and used in Ghana that proved to be readily acceptable to women who practice traditional fish smoking. It consists of a combustion chamber and a smoking unit with a set of trays. The combustion chamber is rectangular, twice as long as it is wide, divided by a wall down the middle and with two stokeholes in front. The recommended standard measurements of the combustion chamber are length-225 cm, width- 112.5 cm, height-60 cm, wall thickness, 12.5 cm, width and height of stoke hole-37.5 cm and depth of fire pit 15 cm. The combustion chamber is the base of the smoker and is generally constructed from mud, although burnt bricks and cement blocks may be used. The latter two are more expensive than mud and cement is not recommended, since it cannot withstand the high smoking temperatures. The top of the wall must be flat so that the trays fit flat and no smoke or heat can escape through gaps. The smoker is designed so that the wooden frame of the trays rests along the midline of the base walls so that they are firmly supported and do not catch fire. The smoking unit consists of a set of 5-15 smoking trays, depending on the size and quality of fish to be smoked [1]. On average, however, ten trays are used per oven for small to medium-sized fish. The chorkor smoker however has some shortfalls, it uses huge quantities of fuel wood for the smoking process, which leads to forest deforestation and is also costly. It also poses a health risk to fish processes as they have to move close to the smoker to turn the fish from time to time.

These challenges with the chorkor smoker have led to the development of the new Abuesi Gas Fish Smoker.

Abuesi Gas Fish Smoker: The Abuesi Gas Fish Smoker is a Liquefied Petroleum Gas (LPG) operated fish smoker, developed by the GRATIS Foundation, Ghana in collaboration with the Abuesi Fish Processing Association, Abuesi, Ghana. It is a double-chamber oven with the dimensions of 1.2 m \times 2.4 m \times 1.8 m. The frame/body is made of a stainless steel to prevent rusting. At the bottom of each chamber is a perforated metal coil through which the gas burns to heat the entire oven. The coil is connected externally to a hose that also connects to a gas cylinder. The oven is also equipped with a thermometer for temperature determination. Within each chamber is a suction fan that sucks moisture from the oven since the heated oven moderately cooks the fish during the process of smoking. Each chamber is designed with a number of grooves where oven wire meshes can be slotted for fish to be arranged on them for smoking. The oven also has a chamber that can be filled with mostly agricultural byproducts like coconut husks and sugarcane bagasse to generate smoke during the smoking process. A fully loaded oven takes about 0.7 tons of fish and the smoking process can be accomplished in two hours or more, depending on whether the end product would be soft or hard smoked. The smoking process can also be staggered such that after the fish are smoked in one oven chamber, the gas can be turned off allowing the heat in the chamber to further dry the fish whiles fresh fish can be loaded into the second chamber for smoking. This would allow one fish processor to be doing different work such as descaling, smoking and packaging within the two hours. The gas smoker has many advantages; processors do not come into direct contact with contact with fire and smoked as with fuel wood operated ovens, it prevents deforestation, It is more hygienic, the threat to the health of processors by smoke/ sooth is prevented, processors can do other tasks while fish is in the oven. It takes a shorter time to process the fish than the traditional smoking methods.

Fish Smoking Processes

The Abuesi gas fish smoker: The fresh samples of the different species were weighed, descaled, gutted, washed thoroughly

with clean water and cut in preparation for smoke curing. Fishes were dipped in freshly prepared salt solution followed by draining. The fish samples were arranged onto removable wire mesh trays and smoked using the Abuesi Gas Fish Smoker. The burning of Liquefied Petroleum Gas (LPG) generated heat. Smoking was done for approximately 3 hours. During smoking, fish samples were turned upside down in mid period, to make the sample smooth and steady in texture and appearance. The samples were cooled for 20 -30 minutes at ambient temperature after smoking. The cooled smoked fish samples were packaged. Smoke-dried fish products were then kept at room temperature for further analysis.

The chorkor smoker: Fresh fish samples of the same species as the ones smoked with the Abuesi Gas fish smoker, were purchased at the Tema fishing harbour, processed using the chorkor smoker and immediately sent to the laboratory for analysis.

Proximate composition: For each of the samples smoked by the two smoking methods (Abuesi Gas Fish Smoker and the chorkor smoker), the moisture content, crude protein, crude lipid, total carbohydrates and ash content were determined using the method of analysis as described by the Association of official Analytical Chemists (AOAC, 1990). The analyses were carried out at the Department of Food Science and Nutrition of the University of Ghana, Legon. Samples were taken from the dorsal, anterior section and mid region of the fish. Each analysis was carried out in replicate.

After the laboratory analyses, the data were subjected to t Test: Paired two samples for means to determine mean differences between crude protein, fat content, moisture content, total carbohydrates and ash content of the fish samples.

PAH Analysis

The PAH analyses were carried out at the Ghana Standards Authority Laboratory. For each of the fish samples smoked using the two ovens, 16 PAHs were targeted (Napthalene (NAP), Acenaphthalene (ACA), Acenaphthene (ACE), Fluorene (FLU), Phenanthrene (PHE), Anthracene (ANT), Fluoranthene (FLT), Pyrene (PYR), Chrysene (CHR), Benzo(b)fluoranthene (BBF), Benzo(k)Fluoranthene (BKF), Benzo(a)pyrene (BAP), Indeno(1,2,3,c,d)pyrene (IND), Dibenzo(a,h)anthracene (DAA), Benzo(g,h,i)perylene (BGP) and Benzo(a)anthracene (BAA). These 16 were targeted because they are the commonly occurring PAHs. The Agilent Bond Elut QuEChERS extraction procedure was used. This method was chosen because it is easy, less expensive and is validated in terms of accuracy, specificity, linearity and quantification limits [8].

Fish samples per species weighed between 120 g to 200 g. The

samples were deboned, blended, bagged and labeled. 5 g of each of the samples was weighed with a Mettler Toledo PG1003-S electronic balance into a Poxygrid 15 ml conical tube. 10 ml of Acetonitrite was added for extraction to each of the samples and homogenized for one minute using Ultra turrax homogenizer IKA- T25. 6 g of Magnesium sulphate and 1.5 g sodium chloride salts were added to the samples and shaken for one minute using the Vortex mixer for liquid-liquid partitioning. The samples were centrifuged for five minutes using a Hermle Z300 centrifuge at a speed of 4RPM.

6 ml of the organic layer was put into a Poxygrid 15 ml conical tube after separation and c18 (High Performance Liquid Chromatography or HPLC), magnesium sulphate and primary secondary amines (PSA) added for clean-up and shaken for 30 seconds. The sample was centrifuged again for 5 minutes at a speed of 4RPM. 4 ml of the sample was put into a pear-shaped flask. The sample was then put in Buchi rotary evaporator for evaporation below 40°C. The sample was redissolved in 1 ml acetonitrite and put in an ultra-sonic water bath to knock the sample stuck to the walls of the flask into the reagent. The sample was put into a 2 ml GC val for analysis of PAHs. A Gas Chromatograph-Mass Spectrometry (GC/MS) was used for the analysis.

The total PAHs, BaP, PAH4, as well as, the sum of 8 SCF -15, which have been deemed genotoxic compounds by the European Scientific Committee on Foods (Food Safety Authority of Ireland, 2015) were calculated.

Results

Proximate composition

The results of the proximate analysis carried out on the smoked fish are presented in Table 1. For the sake of this research, the fish smoked using the Abuesi Gas smoker will be referred to as gas smoked fish (A); while the fish smoked using the chorkor smoker will be referred to as wood smoked fish (C).

The average values of the crude protein, fat content and ash were generally higher in the gas smoked samples. The results however showed a decrease in moisture content of the gas smoked fish compared to those smoked with wood [9,10].

PAH analysis

Table 2 reports concentrations (μ g/kg) of each single PAH in both gas and wood smoked samples. All 16-targeted PAHs were detected in the wood smoked fish except acenaphthalene. The total PAHs, BaP, PAH4 and 8 SCF -15 were higher in the wood smoked samples. The PAH with the highest concentration in the gas smoked samples was naphthalene. The PAHs not detected (nd) were assumed to be absent in the fish samples.

Table 1: The Proximate composition	of the fish smoked using the two smokers.
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Species	%Crude Protein		%Crude fat		%Total carbohydrates		%Moisture content		%Ash content	
	Α	С	Α	С	Α	С	Α	С	Α	С
Tuna	54.23 ± 0.71ª	43.75 ± 0.01 ^b	5.28 ± 0.74^{a}	2.00 ± 0.00^{a}	13.21 ± 0.28ª	5.26 ± 2.82^{a}	24.73 ± 0.09^{a}	46.5 ± 2.12 ^b	2.7 ± 0.04^{a}	2.50 ± 0.71 ^a
White Grouper	61.14 ± 0.53ª	43.73 ± 0.04 ^b	7.05 ± 0.21ª	2.00 ± 0.00^{b}	6.49 ± 0.59ª	25.78 ± 2.09ª	22.48 ± 0.21ª	22.00 ± 2.83ª	2.85 ± 0.06^{a}	6.50 ± 0.71ª
Barracuda	70.32 ± 0.12ª	43.38 ± 0.53 ^b	8.76 ± 0.15ª	2.00 ± 1.41 ^b	2.97 ± 0.49 ^a	13.13 ± 3.01 ^b	13.69 ± 0.43ª	38.50 ± 2.12 ^b	4.27 ± 0.04^{a}	3.00 ± 0.00 ^a
Different superscript letters (a, b) indicate a significant difference (p<0.05) within the smokers for the different species										

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	Tuna (µg/kg)		Barracu	da (µg/kg)	White grouper(µg/kg)		EU MRL (µg/kg
PAH compound (Abbreviated)	Α	C	Α	С	Α	С	
NAP	482.6	2.2	440.1	69.2	251	52.3	
ACA	1.56	43.7	2.1	88.5	1.7	45.5	
ACE	-	-	-	-	-	-	
FLU	0.28	34.6	0.92	93.8	-	67.8	
PHE	-	252.5	-	79.2	22.4	59.6	
ANT	-	251.5	0.4	383.1	27.1	273.8	
FLT	16.8	125.6	17.5	208.2	11.6	152.9	
PYR	0.27	125.1	9.2	208	7.7	152.1	
CHR	3.1	69.7	2.3	420.4	0.2	142.3	
BBF	0.5	26.1	-	66.5	-	49.4	
BKF	1.3	25.3	-	70.3	-	49.9	
BAP	1.4	26.7	-	71.1	-	50.3	
IND	-	9.1	-	18.6	-	14.2	
DAA	-	9.4	-	18.1	-	14.1	
BGP	3.4	9.3	-	18.5	-	14.2	
BAA	3.2	73	2.4	307.7	0.1	123.8	
TOTAL PAH	514.41	1038.8	474.9	1550.4	321.7	1262.2	
Total BAP	1.4	26.7	-	71.1	-	50.3	2
Total PAH4	8.2	195.5	4.7	865.7	0.3	365.8	12
FLT/PYR	62.2	1	1.8	1	1.5	1	
Sum of 8 SCF-15	12.9	248.6	4.7	991.2	0.3	408.3	

Table 2: Summary of PAHs in fish smoked with the gas (A) and wood (C) smokers

LOD: 0.01 µg/kg; "-": Not Detected

Discussion

Proximate Composition

Moisture content is one of the factors that can be used as an indicator of the rate at which deterioration occurs in fish samples resulting in early decomposition [11]. The moisture content of the gas smoked fish samples were less than 25% while the moisture content of the wood smoked fish samples were above 25% except for the white grouper. It has been documented that well dried fish with moisture reduced to 25% (wet weight) can be preserved for a longer period of time as microbial activities are retarded. It can be seen from the results that safe moisture content was achieved in all gas smoked fish products. The crude protein was significantly higher in the gas smoked fish samples than in the wood smoked fish samples. The increase in crude protein could be attributed to its concentration in the fish, as a result of the relatively higher loss of moisture in the gas smoked samples from the smoking process. This inference falls in line with Aliya et al. who stated that there is an inverse relationship between protein and moisture content i.e. protein content increases as moisture content decreases [4]. Holma and Maalekuu (2013) stated the a high crude protein content in fish offered high dietary status due to the essential amino acids they provide [12].

Idah and Nwankwo (2013) reported lower fat content of wood smoked fish and attributed it to high smoking temperatures in the oven. This could be the probable reason for the lower fat content in fish processed with the chorkor smoker as compared to the gas smoker in this study [13]. There was no significant difference between the total carbohydrates content of the gas and wood smoked fish samples. The amount of carbohydrate in fish is generally too small to be given any significance in a diet (FAO, 2001). Ash content is a measure of the total amount of minerals present in the fish. There was no significant difference between the ash content of the gas smoked fish and wood smoked fish samples except in the Barracuda where the ash content was significantly higher in the gas smoked fish than the wood smoked fish.

PAHs Assessment

Higher concentration of PAHs was seen in the wood smoked fish samples. This is in line with report by Akpambang et al. that direct smoking of fish, using fuel wood at high heating temperatures may be responsible for high PAH levels in processed foods. More PAHs were also detected in the wood smoked samples than the gas smoked samples; this demonstrates that burning log fire may introduce many PAHs to the finished product when used for fish smoking. The level of contamination was largely reduced when fish was smoked using gas as the source of fuel. It seems more likely that the use of gas instead of burning wood, as a source of fuel could reduce the level of PAHs contamination in smoked fish products. The different levels of PAHs in fish smoked by the two methods is in line with report by Visciano et al. that the type of combustible used affects the PAH levels in smoked fish products [14-17]. Even though the total concentration of PAHs was higher in the wood smoked fish samples than the gas smoked ones, the total PAHs concentration of the gas smoked fish samples was quite high, and exceeding 300 µg/kg in all the gas smoked fish samples. This relatively high PAHs concentration in the gas smoked fish samples was mainly as a result of high naphthalene concentrations.

The ratio of fluoranthene to pyrene was greater than one (FLT/ PYR>1) in all the fish samples. This dictates that the PAHs detected could be attributed to pyrolytic sources as reported by Amos-Tautau et al. Also, all samples analyzed had similar PAH profile for each smoking method, and could indicate that

sources of contamination of the samples were the same for each smoking method [18]. There was no evident correlation between fat content of the smoked fish samples and PAHs concentration. Essumang *et al.* reported that formation of PAHs is known to occur through the pyrolysis of fat at temperatures above 200°C. The lower PAHs concentrations in the gas-smoked samples could partly be attributed to the less time spent in the smoking process.

All the 8 targeted SCF-15 PAHs considered to be potentially genotoxic and carcinogenic to humans (Food Safety Authority of Ireland, 2015), were found in the wood smoked fish samples while only two were found in the gas smoked fish, except for the gas smoked tuna which had six. The individual PAHs concentrations were also higher in the wood smoked fish samples. This result indicates that using wood as the source of fuel introduced higher concentrations of potentially genotoxic and carcinogenic PAHs to the finished product.

Compliance with EU MRL

From the results, gas smoked fish samples were within the specified limits for PAH4 and BAP, whereas the wood smoked samples had PAH4 and BAP concentrations far exceeding the EU MRL limits. This can be attributed to the different smoking methods used. The BAP levels in the wood smoked fish are in line with researchers such as Essumang et al. and Akpambang et al. who reported BAP levels ranging from 8.5 µg/kg and 73.78 μ g/kg and 2.4 μ g/kg and 31.3 μ g/kg respectively in traditionally smoked fish samples. The presence of high levels of BAP in the wood smoked fish samples is an indication of heavy contamination of commonly consumed smoked fish in Ghanaian diets. This may imply an increased risk of carcinogenic and mutagenic hazards in the Ghanaian population [19]. The noncompliance of the traditionally smoked fish in this study also means that such processed fish products cannot be exported to the EU market.

Practical Applications

This research will add to already existing knowledge concerning nutritional composition of smoked fish products in Ghana and the rest of West Africa. It also provides knowledge on the levels of PAHs introduced into traditionally smoked fish products in the country and the need to intensify such for safer smoking method. It also suggests an alternative to the current commonly used fish smoker for better quality smoked fish products.

Conclusion

The nutritional profile of fish processed with the gas smoker was found to be better than those smoked with the wood smoker. The most significant observation in the proximate composition was the high crude protein in the gas smoked fish as compared to the wood smoked fish. Also, the moisture content of the gas smoked samples was less than 25% while the moisture content of the wood smoked samples was above 25% except for the white grouper. Total PAHs concentration was lower in the gas smoked fish than the wood smoked fish. The MRL for PAH4 and benzo(a)pyrene required by the EU for smoked fish muscle was met in the gas smoked fish, but the wood smoked fish was found to be heavily contaminated with PAH4 and BAP, far exceeding the MRL acceptable by the EU for smoked fish muscle. It is concluded that the Abuesi gas fish smoker produced better quality smoked fish product than the chorkor smoker.

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