Comparative analysis of the Polycyclic Aromatic Hydrocarbon (PAH) content and proximate composition of unripe *Musa paradisiaca* (plantain) fruit exposed to varying methods of roasting

F. N. Nworah¹ · C. C. Nkwocha¹ · J. N. Nwachukwu¹ · E. C. Ezeako¹

Received: 21 March 2018 / Accepted: 5 December 2018 / Published online: 3 January 2019
© Springer Nature Switzerland AG 2019

Abstract

This comparative study was carried out to ascertain the relative effect of smoking and non-smoking methods of food preparation on the concentration of PAHs in unripe plantain sample. The fruit samples were subjected to different smoking methods such as roasting with firewood, charcoal and charcoal augmented with polythene materials as well as non-smoking method such as frying and oven drying process, and compared with the fresh fruits. Gas chromatography-mass spectroscopy (GC-MS) method was used to determine the concentrations of sixteen priority PAHs content of each sample and the results were subjected to statistical analysis. The percentage compositions of crude fibre in the raw, smoked roasted (charcoal, firewood and augmented charcoal) and non-smoked (oven dried and fried) samples were; 4.4%, 3.2%, 3.1%, 3.0%, 3.5% and 4.1% respectively. Percentage composition of protein and carbohydrate of the raw food samples were found to be 3.62% and 34.5% respectively which were higher than the dried food samples. The total PAHs concentration of the fresh plantain sample (control) was $(8.0 \pm 0.1 \text{ mg/L})$. The Charcoal, firewood and augmented roasted sample had total PAHs concentration of $(19.3 \pm 0.2 \text{ mg/L})$, $(19.6 \pm 0.1 \text{ mg/L})$ and $(20.1 \pm 0.1 \text{ mg/L})$ respectively, whereas the total PAHs concentration in the fried and oven dried samples were $(9.8 \pm 0.1 \text{ mg/L})$ and $(15.3 \pm 0.2 \text{ mg/L})$ respectively. From the result, it was observed that the total PAHs concentration of the smoke roasted sample were significantly higher ($p < 0.05$) than the non-smoke roasted sample which indicates that roasting and grilling over open fire or smoke do increase the level of Polycyclic aromatic hydrocarbons (PAHs) in food. Although the concentration of PAHs in the fried sample was significantly ($p < 0.05$) lower than the oven dried sample, however, it predisposes one to increased risk of atherosclerosis and related lipidemia due to its increased fat concentration. Hence smoking method of food preparation should be substituted with other non-smoking methods such as oven drying.

Keywords Polycyclic Aromatic Hydrocarbon (PAH) · *Musa paradisiaca* · GC-MS · Charcoal · Smoking

Introduction

In Nigeria, grilled or roasted plantain is one of the most popular snacks taken by both the young and old. It could be taken at any time of the day but preferably in the afternoon. These snacks are available throughout the year. In fact, the snacks supplemented with plenty of water now form the main source of lunch for some low income earners [1]. Several method of food preparation such as roasting and smoking in open fire has

been reported to increase the level polycyclic aromatic hydrocarbon in food [2].

Polycyclic aromatic hydrocarbons (PAHs) are class of ubiquitous environmental organic pollutants, consisting of benzene rings (liner, cluster or angular arrangement), produced as a result of incomplete combustion and pyrolysis of organic materials or during the preparation of food products [3]. PAHs originate mainly from anthropogenic processes particularly from incomplete combustion of organic fuels and are distributed widely in the atmosphere. Natural process, such as volcanic eruptions and forest fires, also contribute to an ambient existence of PAHs [4]. PAHs can be present in both particulate and gaseous phases, depending on their volatility. Low molecular weight PAHs are emitted into the gaseous phase, while high molecular weight PAHs, are emitted in the particulate phase [5]. The later has been found to confer

✉ F. N. Nworah
florence.nworah@unn.edu.ng

¹ Food Safety and Toxicity Unit, Department of Biochemistry, University of Nigeria, Nsukka, Enugu State, Nigeria

carcinogenic and mutagenic effects while the former may act as synergists [6]. PAHs have been found in different food products, such as dairy products, vegetables, fruits, common delicacies such as roasted plantain, corn, yam, oils, coffee, tea, cereals and smoked meat, therefore the analysis of PAHs in food is a matter of concern [7].

Food smoking belongs to one of the oldest methods of food preservation and processing. It is known to increase the sensory quality, shelf -life, wholesomeness and market value of the food product [3]. However, in the food processing industries, food additives such as smoke flavoring products (SFP), Lubricants, solvents, propellants, glazing agents and protective coatings have been implicated to contribute to the PAHs contamination of food [8]. Over the years, different sources or route of PAHs contaminations of foods have been found which include; soils, polluted air and water [9]. Reports have shown that raw foods do not usually contain high levels of PAHs, however, industrial processes that emit PAHs into the atmosphere, may contaminate the food products [10]. Also, areas remote from urban or industrial activities could possess high levels of PAHs in unprocessed foods, as a result of background contamination.

There are hundreds of PAHs compounds in the environment but in practice, PAHs analysis is restricted to the determination of six (6) to sixteen (16) compounds. Of these 16 PAHs, Benzo [a] Pyrene (B(a)P) is probably the most studied and has been widely used in environmental analysis as marker for the entire PAH content [9]. The act of subjecting food materials to direct smoking product elevates the levels of PAHs in the food which could pose several health defects in humans. Hence the determination of the levels of PAHs in some of these ubiquitous food products such as plantain is very imperative, as it could aid in the fight and alleviation of cancer, and other inherent health complications.

Materials and methods

Study site

The plantain samples used in this study were obtained from Ogige market, University of Nigeria, Nsukka, Nsukka L.G.A, in Enugu State within the period of June 2017.

Methods

Collection of plantain samples and different drying method

A bunch of unripe plantain consisting of eighteen (18) heads of fresh fruits was bought from Ogige market, Nsukka. The

unripe plantain samples were divided into six (6) groups and the wet weight of each group noted.

Preparation of plantain samples

The smoking processes

The smoking processes of the plantain fruits was done at high temperature above 250 °C and involved the production of heat accompanied with smoke from smoldering wood (hard wood) and charcoal placed directly below the hanging plantain fruit laid on mesh trays. It was continuously fanned to enhance combustion. In another set of the preparation by smoking, polythene materials were used to enhance the flame and ease the smoking process.

The non-smoking processes

The plantain fruits were placed in very hot oil on a deep fryer and were fried at a temperature of 170 °C for 10 mins. After cooling, the fruits were homogenized into powder form using electric blender. After grinding the plantain fruits, powdered samples were labeled accordingly. Also, the plantain fruits were placed on an electric oven at a temperature of 220 °C for 5 min. After cooling, the samples were homogenized into powdered form using electric blender. After homogenizing the plantain fruits, powdered samples were labeled accordingly.

Experimental design

- Group A1: Fresh unripe plantain.
- Group A2: Plantain sample dried with charcoal fire.
- Group A3: Plantain sample dried with firewood.
- Group A4: Plantain sample oven dried.
- Group A5: Fried plantain sample.
- Group A6: Plantain sample dried with charcoal augmented with polythene material.

Sample preparation for the analysis of PAHs

Soxhlet extraction

Twenty grams (20 g) of the homogenized plantain sample was thoroughly mixed with 60 g of anhydrous sodium sulphate in an agate mortar to absorb moisture. The homogenate was placed into an extraction cellulose thimble covered with a whatman filter paper (125 mm diameter) and inserted into a soxhlet extraction chamber of the soxhlet extraction unit. Extraction was then carried out with 200 ml of n-hexane using EPA 3540 C method (US EPA, 1994) for 8 h. The crude extract obtained was carefully evaporated using rotary vacuum evaporator (Ribby RE 200B) at 40 °C just to dryness. The

residue was dissolved in 5 ml of n-hexane and transferred onto a 10 ml florisil column for clean-up [11].

Preparation of florisil for clean-up

This clean-up step to remove more polar substance was performed using activated florisil (magnesium silicate) and anhydrous Na_2SO_4 . The florisil was heated in an oven at 130 °C overnight and transferred to a 250 ml size beaker and place in a desiccator. Anhydrous Na_2SO_4 (1.0 g) was added to 2.0 g of activated florisil (60–100 mm mesh) on a 10 ml column which was plugged with glass wool. The packed column was filled with 5 ml n-hexane for conditioning. The stopcock on the set-up was opened to allow the n-hexane run out until it just reached the top of the sodium sulphate into a receiving vessel whilst taping gently the top of the column till the florisil settled well in the column. The extract was then transferred onto the column with a disposable Pasture pipette from an evaporating flask. The crude extract was eluted on the column with the wide opening of the stopcock. Each evaporating flask was immediately rinsed twice with 1 ml of n-hexane and added to the column by the use of the pasture pipette. The eluate was collected into an evaporating flask and rotary evaporated to dryness. The dry eluate was then dissolved in 1 ml n-hexane for Gas chromatographic analysis [11].

Instrumental analysis

Analysis of PAHs using GC-FID

Procedures One milliliter (1 ml) of the filtered residue was Dissolved in 50 ml of chloroform and transferred to a 100 ml volumetric flask and diluted to the mark, most of the chloroform was evaporated at room temperature. Then 1 ml of the reagent (20 vol% benzene and 55 vol% methanol) was added and the vessel was sealed and heated at 40 °C in a water bath for 10 min. After heating, the organic sample was extracted with hexane and water, so that the final mixture of the reagent, hexane and water, is in proportion of 1:1:1 (i.e., by adding 1 ml of hexane and 1 ml of water to the reaction mixture). The mixture was shaken vigorously by hand for 2 min, and observed to see any stable emulsion formed, which is afterward broken by centrifugation. About half of the top hexane phase was transferred to a small test tube for injection. Care was taken to remove only the organic layer, and some quantity was taken into 2 ml chromatographic vial and made up to 2 ml with toluene. Temperature condition of GC-FID was as follows; Injector temperature was set at 22 °C, detector temperature at 250 °C with integrator chart speed of 2 cm/min. The temperature gradient temperature was as follows: 70 °C maintained for 5 min at 22 °C/min up to 220 °C, then at 220 °C maintained for 2 min at 55 °C/min up to 280 °C. The recovery rate of the GC was 81% and in quality control,

standard was run and compared with the internal standard. Detection limit was 0.00001 µg/g.

Determination of proximate and antinutrient content

The standard method of AOAC [11] hot air oven method 925.10 of analysis was used. Four porcelain dishes were thoroughly cleaned and dried in hot air oven at 100 °C for 30 min. They were cooled in desiccators for five minutes to remove the surface moisture. The porcelain dishes were weighed and 2 g of the samples was weighed into them and placed in the oven at 150 °C for six hours for moisture elimination. The porcelain dishes were removed from the oven and cooled in the desiccators after drying to prevent moisture re-absorption.

After cooling the porcelain dishes were weighed again and the moisture percentage calculated.

$$\% \text{ Moisture} = \frac{X-Y}{Z} \times 100$$

Where:

- X sum of weight of the dishes and sample before drying;
- Y weight of the dish and sample after drying;
- Z initial weight of the sample in gram

Determination of ash content

The AOAC method [11] was used to determine the ash content of the two samples. Four ashing dishes were weighed and dried in a hot air oven at 100 °C for 25 min and cooled in desiccators. Two grams of samples were weighed into the porcelain dishes and charred on desiccators to remove carbon. The samples were put in muffle furnace at 600 °C until ashing was completed. Dishes and content was cooled in the desiccators after ashing.

Determination of protein content

The micro Kjeldahl method described by AOAC [11] was used in the determination of the crude protein, which involved two stages:

Digestion Ten grams of each sample were digested with a mixture of concentrated sulphuric acid; a pinch of copper sulphate and sodium sulphate crystals to act as catalyst. The mixtures were heated till the black liquid cleared. Heating continued till the sample becomes completely digested. The digested samples were transferred into 100 ml volumetric flask, which was weighed in preparation for distillation.

Distillation Kjeldahl distillation apparatus was used. Boric acid 10 ml was combined with two drops of methyl blue and

methyl red indicators in a conical flask of 100 ml. The conical flask was positioned into the receiving side of the distillation unit using a clamp. Ten millilitres of the digested samples was first introduced into the distillation unit followed by addition of 10 ml of concentrated NaOH gradually. The distillation process lasted for five to ten minutes, during which ammonia (NH_4) was trapped in excess boric acid. The presence of ammonia changed the purple color of boric acid to green which is a common characteristic of alkaline gas. Ammonia trapped in boric acid indicator was titrated using 0.1N HCl. Crude protein content was calculated by the formula.

$$\% \text{ Nitrogen} = \frac{T \times N \times DF \times MWN}{\text{weight of Sample}}$$

Where:

T Titre volume
N Normality of Acid
DF Dilution Factor (100 ml/5 ml) = 20
MWN Molecular weight of nitrogen = 14.01

Therefore to convert nitrogen content into protein content = $N \times 6.25\%$

Protein = % Nitrogen $\times$ 6.25

Where 6.25 = Conversion factor of nitrogen to protein

Determination of fat content

The soxhlet extraction method described by AOAC [11] was used. Two grams of the samples were weighed into dry soxhlet thimbles and put in soxhlet condenser attached to a weighed extraction flask containing 100 ml of petroleum ether. The thimbles were removed from the chamber separating the petroleum ether completely, cooled in desiccator and weighed. The percentage fat for each sample was obtained.

$$\% \text{ Fat} = \frac{Y - X}{Z} \times 100$$

Where:

X weight of flask and extracted fat (g)
Y weight of the sample and flask (g)
Z weight of the sample (g)

Determination of crude fibre content

The AOAC method [11] was used. Ten grams of the sample were hydrolysed in a beaker and 100 ml of 1.25% diluted sulphuric acid (H_2SO_4) added in the flask to digest for 30 min. The content was filtered using a Buckner flask, which is white calico cloth, fitted with a vacuum pump attached. To the digested sample, (1%) HCl was added to neutralize the

NaOH present and were cleaned with methylated spirit to remove any trace of oil. The residue was put into a weighed crucible, dried in an oven for 30 min, cooled in desiccators and reweighed. The sample was put into a muffled furnace for two hours and calculated thus:

Determination of carbohydrate content

The carbohydrate content of the samples was obtained by difference in hundred and sum of other proximates
Carbohydrate = $100 - (\% \text{ moisture} + \% \text{ ash} + \% \text{ crude fat} + \% \text{ crude protein} + \% \text{ crude fibre})$.

Determination of oxalate concentration

The oxalate content of the samples was determined using titration method. Each sample of A and B (2 g) was placed in a 250 ml volumetric flask suspended in distilled water (190 ml). HCl (10 ml, 6 M) solution was added to each of the samples and the suspension digested at 100 °C for 1 h. The samples were then cooled and made up to (250 ml) mark of the flask. The samples were filtered and duplicate portion (125 ml) of the filtrate were measured into beaker and four drops of methyl red indicator was added, followed by the addition of concentrated NH_4OH solution (drop wise) until the solution changed from pink to yellow colour. Each portion was then heated to 90 °C, cooled and filtered to remove the precipitate containing ferrous ion. Each of the filtrate was again heated to 90 °C and 10 ml of 5% CaCl_2 solution was added to each of the samples with stirring consistently. After cooling, the samples were left overnight. The solutions were then centrifuged at 2500 rpm for 5 min. The supernatant were decanted and the precipitates completely dissolved in 10 ml of 20% H_2SO_4 . The total filtrates resulting from digestion of 2 g of each of the samples were made up to 200 ml. Aliquots of 125 ml of the filtrate was heated until near boiling and then titrated against 0.05 M standardized KMnO_4 solutions to a pink colour which persisted for 30 s. The oxalate contents of each sample were calculated [12].

Determination of phytate concentration

The phytate concentration of each of the samples was determined through phytic acid determination using the procedure described by [13]. This entails the weighing of 2 g of each sample into 250 ml conical flask. Hundred millilitre of 2% conc. HCl was used to soak the samples in the conical flask for 3 h and then filtered through a double layer filter paper 50 ml of each of the sample filtrate were placed in a 250 ml beaker and 107 ml of distilled water added to give/ improve proper acidity. Ten millilitre of 0.3% ammonium thiocyanate solution was added to each sample solution as indicator and titrated with standard iron chloride solution which contained

0.00195 g iron/ml and the end point was signified by brownish-yellow colorations that persisted for 5 min. The percentage phytic acid was calculated [14].

Determination of hemagglutinin concentration

Two grams of each of the sample was added to 20 ml of 0.9% NaCl and suspension shaken vigorously for 1 min. The supernatants were left to stand for 1 h. The samples were then centrifuged at 2000 rpm for 10 min and the suspension filtered. The supernatants in each were collected and used as crude hemagglutinin extract [14].

Statistical analysis

The data obtained were analyzed using computer software known as Statistical Product and Solution Service (SPSS) version 18. One way analysis of variance (ANOVA) was used for comparison across the different groups and differences were considered significant at $p < 0.05$. While, Post Hoc test was used to separate and compare the mean.

Result

Initial wet weight of the unripe plantain samples studied

Table 1 shows the wet weight of the unripe plantain samples obtained from the University market, University of Nigeria, Nsukka, Nsukka Local Government Area, in Enugu State studied. The percentage water loss of the different drying employed varied significantly among the treatment groups. The Oven dried plantain sample had the highest percentage water loss of 17.54% followed by the charcoal dried sample. While the fried augmented charcoal dried and firewood dried sample had a relatively less percentage water loss of 15.0%, 13.60 and 13.14% respectively. From the statistical analysis, it was observed that the mean value of the various drying methods employed showed significant difference ($p < 0.05$) compared to the control (fresh sample), which indicates that

there was significant differences in the percentage water loss of all the various treatment groups.

Proximate compositions of the plantain samples

Table 2 shows the proximate compositions of the plantain samples subjected to different treatments. All the raw food samples had the highest percentage of crude fibre (4.4%) than the other samples. The percentage compositions of crude fibre in the smoked roasted food samples; charcoal, firewood and augmented charcoal dried samples were; 3.2%, 3.1% and 3.0% respectively while the non-smoked dried sample; oven dried and fried samples were 3.5% and 4.1% respectively. Percentage composition of protein and carbohydrate of the raw food samples were found to be 3.62% and 34.5% respectively which were higher than the dried food samples.

Anti-nutrient compositions of the plantain samples

Table 3 shows that the levels of antinutrient content of the dried food samples were significantly ($p < 0.05$) lower than the control. The Oven dried samples had the least phytate and haemagglutinin level of (11.9 ± 2) and (12.33 ± 0.1) respectively. While the augmented smoked sample had the highest haemagglutinin levels of (26.01 ± 0.1) . The fresh samples had the highest phytate and oxalate levels.

Discussion

Table 4 shows that in the statistical analysis, a significant amount of PAHs in the raw unripe plantain sample was observed. This may be from the contaminated soil of which the plantain fruits were cultivated upon. The work of Nikolaou et al. [15] suggested that Soils and sediments are often the ultimate repository for most of the hydrophobic organic contaminants such as PAHs bonded with particles and that most anthropogenic PAHs are restricted to the top layer of the soils. Kim et al. [16] reported that; particularly 90% of the total PAHs are located in soil and are usually present in the environment as complex mixtures. Amodio et al. [1] also supported the

Table 1 Weight of plantain samples

Components	Wet weight (g)	Dry wet (g)	% water loss	% water $\pm$ SD
Fresh unripe plantain(A1)	143.2 $\pm$ 0.2	–	–	–
Charcoal roasted unripe plantain (A2)	144.3 $\pm$ 0.3	122.4 $\pm$ 0.3	15 $\pm$ 0.3	15 $\pm$ 0.3
Firewood roasted unripe plantain (A3)	143.8 $\pm$ 0.2	124.9 $\pm$ 0.2	13.14 $\pm$ 0.2	13.14 $\pm$ 0.2
Oven roasted unripe plantain (A4)	142.4 $\pm$ 0.4	121.3 $\pm$ 0.4	17.54 $\pm$ 0.2	17.54 $\pm$ 0.2
Fried unripe plantain (A5)	145.3 $\pm$ 0.2	119.8 $\pm$ 0.2	14.82 $\pm$ 0.4	14.82 $\pm$ 0.4
Charcoal + polythene roasted unripe plantain(A6)	143.4 $\pm$ 0.3	123.9 $\pm$ 0.3	13.6 $\pm$ 0.3	13.6 $\pm$ 0.3

Results are expressed in mean $\pm$ SD, $n = 3$

Table 2 Proximate compositions of plantain samples

Plantain samples	Proximate compositions (%)					
	Crude Fibre	Protein	CHO	Moisture	Fat	Ash
Fresh unripe plantain(A1)	4.4 ± 0.3	3.62 ± 0.3	34.5 ± 0.3	52.54 ± 0.3	4.7 ± 0.3	4.24 ± 0.1
Charcoal roasted (A2)	3.2 ± 0.2	3.57 ± 0.2	33.8 ± 0.2	50.71 ± 0.2	4.5 ± 0.2	4.22 ± 0.2
Firewood roasted (A3)	3.1 ± 0.1	3.54 ± 0.1	33.3 ± 0.1	51.64 ± 0.1	4.2 ± 0.1	4.22 ± 0.1
Oven roasted (A4)	3.5 ± 0.1	3.27 ± 0.1	33.5 ± 0.1	51.52 ± 0.1	4.0 ± 0.1	4.21 ± 0.1
Fried (A5)	4.1 ± 0.2	3.30 ± 0.2	34.1 ± 0.2	46.62 ± 0.2	7.9 ± 0.2	4.25 ± 0.2
Charcoal + polythene roasted (A6)	3.0 ± 0.1	3.51 ± 0.1	33.1 ± 0.1	51.76 ± 0.1	4.4 ± 0.1	4.23 ± 0.1

Results are expressed in mean ± SD. n = 3

proposition that the lipophilic nature of PAHs makes it easy to attach to soils with high organic content, Lucas and Markaka [13] added that plants absorb nutrients including chemical compounds such as PAHs and its derivatives from the soil and via translocation deposits it in peripheral organs like fruits, stem and vegetative organs, which ultimately serve as fuel in the smoking processes.

The result showed that the total PAHs concentration of the smoked samples were significantly ($p < 0.05$) higher than those subjected to non-smoking treatment (frying and oven drying). This could be as a result of the higher PAHs levels produced in the process. Lee and Vu [4] reported that smoking method is accompanied with the incomplete combustion of organic material used as fuels such as firewood, charcoal and polythene materials resulting in the release of PAHs as byproducts in the smoke.

The total PAHs concentration of the plantain smoked with smoldering firewood were higher than those smoked with charcoal, (although no significant difference exist between the groups) suggesting that the smoke from the firewood contributed more PAHs to the food samples than the charcoal, which corresponded with the work of [5]. It could also be inferred that prior to when it was used as fuel, the charcoal must have lost most of its PAHs concentration during its formation from wood burning, whereas the logs of wood had its PAHs contents intact before it was used, resulting in increased

PAH levels in firewood drying. Also the increased PAH levels in the firewood smoking treatment could be due to higher heat energy involved, when logs of wood are used as fuel, compared to when charcoal is used. This is supported by the work of Okonkwo and Ajuonuma [6] who reported that the combustion temperature during the generation of smoke seems particularly critical and PAHs is formed as a result of varying temperature degrees whenever wood, coal or oil is burnt.

The non-smoked samples showed significantly lower ($p < 0.05$) levels of PAHs relative to the roasted treatment compared to the positive control. However, it was observed that the levels of PAHs in the smoked samples showed a significant increase ($p < 0.05$) compared to the oven dried samples. This could be as a result of the increased heat intensity and sufficient O₂ concentrations in the open air which enhances combustion and as a result, increased the level of emission of gaseous and particulate mixture of PAHs, suspended in the smoke [6].

Also, the oven dried samples had a significant increase ($p < 0.05$) in PAHs level compared to both the positive control and the fried sample. This may be due to the increased heat intensity involved in the oven drying process. This is in tandem with the work of Hiba et al. [17] which reported that PAHs emission increases with increasing temperature from 200 °C to 700 °C but negatively correlate with the moisture content in the foodstuff. Lim et al. [16] also reported that the

Table 3 Anti-nutrient compositions of plantain samples

Components	Anti-nutrient compositions (%)		
	Phytate	Oxalate	Haemagglutinin
Fresh unripe plantain (A1)	43.7 ± 0.1 ^d	34.3 ± 0.1 ^d	18.4 ± 0.1 ^c
Charcoal roasted (A2)	16.8 ± 0.2 ^c	20.21 ± 0.2 ^b	25.90 ± 0.1 ^d
Firewood roasted (A3)	15.38 ± 0.2 ^b	17.39 ± 0.2 ^a	18.39 ± 0.2 ^c
Oven roasted (A4)	14.9 ± 0.3 ^b	18.11 ± 0.3 ^a	12.33 ± 0.1 ^a
Fried (A5)	11.9 ± 0.2 ^a	28.0 ± 0.1 ^c	14.9 ± 0.1 ^b
Charcoal+ polythene roasted (A6)	16.5 ± 0.1 ^c	20.17 ± 0.1 ^b	26.01 ± 0.1 ^d

Results are expressed in mean ± SD. Values having different letter as superscript across the rows are considered significant at $p < 0.05$. n = 3

Table 4 PAHs component of plantain samples

PAHS Components	Fresh unripe plantain	Charcoal unripe plantain	Firewood roasted unripe plantain	Oven roasted unripe plantain	Fried unripe plantain	Charcoal + polythene roasted unripe plantain	Recovery rate of individual PAH (%)
Acenaphthene	0.7 ± 0.1 ^a	1.2 ± 0.2 ^b	1.4 ± 0.1 ^b	0.8 ± 0.2 ^a	0.6 ± 0.1 ^a	10.0 ± 0.2 ^c	85
Acenaphthylene	ND	ND	ND	ND	ND	ND	85
benzo(ghi)perylene,	ND	ND	ND	ND	ND	ND	85
Benz(a)anthracene	ND	ND	ND	ND	ND	ND	85
Anthracene	ND	ND	ND	ND	ND	1.3 ± 0.2 ^a	65
Benzo (a) pyrene	0.1 ± 0.1 ^a	0.2 ± 0.2 ^b	0.2 ± 0.1 ^b	0.1 ± 0.2 ^a	0.1 ± 0.1 ^a	ND	73
Benzo(b) fluoranthene	ND	ND	ND	ND	ND	0.9 ± 0.2 ^a	85
Benzo(k) fluoranthene	ND	ND	ND	ND	ND	ND	85
Chrysene	ND	ND	ND	ND	ND	ND	85
Dibenzy (a,h) anthracene	1.2 ± 0.1 ^a	7.0 ± 0.2 ^d	7.1 ± 0.1 ^d	5.9 ± 0.2 ^c	2.9 ± 0.1 ^b	ND	85
Flouranthene	0.5 ± 0.1 ^a	0.5 ± 0.2 ^a	0.5 ± 0.1 ^a	0.5 ± 0.2 ^a	0.4 ± 0.1 ^a	5.0 ± 0.2 ^b	62
Indeno(1,2,3-cd)pyrene	ND	ND	ND	ND	ND	ND	85
Naphthalene	0.9 ± 0.1 ^a	4.1 ± 0.2 ^d	4.1 ± 0.1 ^d	3.4 ± 0.2 ^c	1.3 ± 0.1 ^b	0.8 ± 0.2 ^a	75
Phenanthrene	ND	ND	ND	ND	ND	ND	85
Pyrene	3.7 ± 0.1 ^b	3.7 ± 0.2 ^b	3.7 ± 0.1 ^b	3.7 ± 0.2 ^b	3.7 ± 0.1 ^b	1.0 ± 0.2 ^a	70
Fluorene	0.9 ± 0.1 ^a	2.6 ± 0.2 ^c	2.6 ± 0.1 ^c	0.9 ± 0.2 ^a	0.8 ± 0.1 ^a	1.1 ± 0.2 ^b	85
Total PAHS	8.0 ± 0.1	19.3 ± 0.2	19.6 ± 0.1	15.3 ± 0.2	9.8 ± 0.1	20.1 ± 0.2	85

ND Not Detected

Results are expressed in mean ± SD. Values having different letter as superscript across the row are considered significant at $p < 0.05$. $n = 3$

levels of PAHs increased with decreasing moisture content of the food material.

Also, from the result it was observed that the concentration of PAHs in the fried samples was significantly lower ($p < 0.05$) than the other treatment. This could be due to the less heat energy involved in frying as well as the less PAHs content of the oil compared to the smoke. Nevertheless, the fat content of the fried sample increased significantly ($p < 0.05$). However there was a significant increase (at $p < 0.05$) in the PAHs content of the fried sample when compared to the positive control. The additional PAHs may be from the vegetable oil used, as supported by the work of [8] which showed that vegetable oil do contain PAHs.

Furthermore, nine types of PAHs were observed in all the groups, of which only four of it belong to the group declared as carcinogenic by the EPA (Environmental Protection Agency) [18–20]. Nevertheless the levels of these carcinogenic PAHs in all the food samples were less than the maximum permissible level set by the EPA.

Also, the augmented smoke sample did not correspond with the other sample in the types of PAHs present in it. It contained some PAHs that were not detected in other groups while other PAHs that were present in other groups were not detected in it.

The statistical Analysis of the individuals PAHs in various treatment showed that there were no significant differences

($p < 0.05$) in the level of flouranthene and Pyrene. This suggests that these PAHs may be from the plantain fruit, as supported by the work of Olabemiwo [5], who reported that unprocessed food materials do contains limited amount of PAHs. Flouranthene and Pyrene are four ring high molecular weight (HMW) PAHs (i.e. PAHs having four or more rings). Rajendran et al., [9] added that HMW PAHs are usually found exclusively bounded to a particulate matter, and not free in the atmosphere.

The level of acenaphthene and Benzo(a) pyrene were not significantly different ($p < 0.05$) among the roasted treatment, however, there was a significant increase ($p < 0.05$) in their concentration in the roasted sample relative to the non-roasted samples. This significant increase ($p < 0.05$) could be from atmospheric contamination of the food samples during the drying process which is usually carried out in open air condition. Huertero et al. [21] reported that the total PAHs level in ambient air is about 0.02–12 ng/m³ in urban areas. Also Nikolaou et al. [15] supported that PAHs are also a component of concern in particulate matter suspended in the air. The lipophilic nature of PAHs makes them easy to be attached to soils with high organic content as reported by Amodio et al. [1].

There was no significant difference ($p < 0.05$) in the level of Dibenzo(ah) anthracene (DBahA) and Naphthalene (Naph) between the charcoal and firewood roasted sample, however,

there was a significant increase in the level of these PAHs in the smoked samples aforementioned relative to the other groups when compared with the positive control. This suggests that the smoking process and the ambient air composition may have contributed to the additional PAHs concentration in the sample, as reported by [22]. Naphthalene is a two ring low molecular weight (LMW) PAHs, found in the atmosphere while DB(ah)A is a five ring HMW PAHs found bounded to particulate matters almost exclusively [8].

Also the oven dried samples showed a significant increase ($p < 0.05$) in DB(ah)A and Naphthalene compared to the positive control. This could be due to the decrease in moisture content of the sample as a result of the drying process at high temperature, which may have increased the PAHs concentration of the oven dried sample compared to the fresh sample. This is in accordance with the work of Comandini and Brezinsky [23] who reported that PAHs emission increase with increasing temperature from 200 to 700 °C but negatively correlate with the moisture content in the foodstuff.

The Augmented smoke sample released two other types of PAHs namely; Anthracene and Benzo(b) flouranthene. These were not detected in the other samples, probably because they were not in detectable concentration. However the use of polythene materials must have increased the concentration to detectable levels.

Augmentation of smoke is reported to be an ill- practice that must be shunned strictly. It increases to a very high level, the total concentration of PAHs in food product. The levels of Acenaphthene (Ace) in the augmented smoked treatment were tenfold greater than the other treatment. Also the level of Flouranthene (Fln) in the augmented smoke was tenfold greater than the other treatment groups, compared to the control. Moreover, no significant difference ($p < 0.05$) in Flouranthene levels existed among other treatment groups. Also, there was no significant difference ($p < 0.05$) in concentration of fluorine in the augmented samples compared to the control. It can be inferred that such increase in the PAHs level of the sample may be as a result of the high PAHs content of the polythene material used. Brezinsky et al. [24] showed that; Flouranthene, Anthracene and Acenaphthene among others are example of PAHs used in the manufacturing of synthetic products such as; plastics and cellophane materials Brazkova and Krastanov [25] also added that Naphthalene (Naph), Acenaphthene, Anthracene, fluorathene, fluorine, phenanthrene and pyrene are used in the manufacturing of plastics materials, among other uses.

Flouranthene and Acenaphthene are not included among the eight PAHs declared as carcinogenic by the EPA [26, 27]. However on accumulation, they may act synergistically to cause disease [6]. The other PAHs probably were not in detectable level. According to Igwo-Ezikpe [28] the non-detection of low molecular weight (LMW) PAHs in these samples may be due to instrument sensitivity not within the

range of the PAHs detection or the relative low stability of LMW PAHs. Olabemiwo [5] also stated that the thermal decomposition of this group of PAHs is a possibility because the samples were exposed to direct heat during grilling and smoking.

Conclusion

Based on the result of this study it could be inferred that smoking methods of food preparation significantly increases the level of carcinogenic compounds such as polycyclic aromatic hydrocarbon (PAHs). Although smoking of food add some desirable quality to the food product, it predisposes one to higher risk of contracting cancer and other health complications. Therefore smoking method of food preparation should be replaced with non-smoking processes such as frying and oven drying, but preferably oven drying. Since fried food product do have elevated levels of fat as shown by the result of the proximate analysis and as such could predispose one to other complication such as atherosclerosis, fatty liver and cholesterol related diseases. Therefore homes and commercial food preparation settings should take note and possibly substitute the smoking method of food preparation for electric oven drying method.

Acknowledgements The authors would like to thank the staff and the participants of the various institutions visited for their contributions.

Compliance with ethical standards

Conflict of interest The authors declare no conflict of interest.

Publisher's Note Springer Nature remains neutral with regard to jurisdictional claims in published maps and institutional affiliations.

References

1. Amodio M, Andriani E, De Gennaro G, Di Gilio A, Ielpo P, Placentino C, et al. How a steel plant affects air quality of a nearby urban area: a study on metals and PAH concentrations. *Aerosol Air Qual Res.* 2013;13:497–508.
2. Abdel-Shafy HI, Mansour MSM. A review on polycyclic aromatic hydrocarbons: source, environmental impact, effect on human health and remediation. *Egypt J Pet.* 2016;25:107–23.
3. Omodara NB, Obijole AO, Adekunbi EA, Oloyede HO, Oyeade A. Investigation of polycyclic aromatic hydrocarbon concentration in roasted plantain, roasted meat and smoked fish in Ondo. *Nigeria. Int J Innov Environ Stud Res.* 2015;3(1):27–31.
4. Lee BK, Vu VT. Sources, distribution and toxicity of polyaromatic hydrocarbons (PAHs) in particulate matter. In: Villanyi V, editor. *Airpollution*, vol. 9; 2010. p. 119–28.
5. Olabemiwo OM. Levels of polycyclic aromatic hydrocarbons in grilled/roasted maize and plantain sold in Ogbomoso Nigeria. *Int J Basic Appl Sci.* 2013;13(3):87–93.

6. Okoronkwo NE, Ajuonuma DA. Evaluation of polycyclic aromatic hydrocarbons levels in roasted food samples. *Acad J Sci*. 2014;3(1): 1–8.
7. Okali, E. E. (2015). Levels of Polycyclic Aromatic Hydrocarbons in Some Brands Of Pastas In Nigerian Market: An Assessment Of Possible Potential Health Risk. M.Sc Thesis: University of Nigeria, Nsukka. Pp 76.
8. Rengarajan T, Rajendran P, Nandakumar N, Lokeshkumar B, Rajendran P, Nishigaki I. Exposure to polycyclic aromatic hydrocarbons with special focus on cancer. *Asian Pac J Trop Biomed*. 2015;5(3):182–9.
9. Rajendran P, Kambaram G, Sakthisekaran D. Cytoprotective effect of benzo(a)pyrene-induced lung carcinogenesis in swiss albino mice. *Basic Clin Toxicol*. 2008;103:137–42.
10. Hiba AM, Nimir AS. Identification of polycyclic aromatic hydrocarbons emissions from wood of aromatic plants (Dokhan Incense) used in Sudan. *J Chem Cheml Sci*. 2015;5(10):575–84.
11. AOAC. Official methods of analysis. 15th ed. Washington, DC: Association of Official Analytical Chemists; 1990.
12. Munro AB. Oxalate in Nigerian vegetables. *Biol Appl Chem*. 2000;12(1):14–8.
13. Lucas GM, Markaka P. Phytic acid and other phosphorus compound of bean (*Phaseolus vulgaris*) *J. Agric Educ Chem*. 1975;23: 13–5.
14. Russel HS. India-New England before the may flower. New England: University Press of New England; 1980. p. 1–237
15. Nikolaou A, Kostopoulou M, Petsas A, Vagi M, Lofrano G, Meric S. Levels and toxicity of polycyclic aromatic hydrocarbons in marine sediments. *Trends Anal Chem*. 2009;28(6):653–64.
16. Kim KH, Jahan SA, Kabir E, Brown RJ. A review of airborne polycyclic aromatic hydrocarbons (PAHs) and their human health effects. *Environ Int*. 2013;60:71–80.
17. Hiba AM, Nimir AS, Tawfeeg EM. Identification of polycyclic aromatic hydrocarbons emissions from Oud Incense – Al – Baha City Southwest Saudi Arabia. *Int J Chem Sci Res*. 2015;5(6):412–22.
18. IARC. Overall evaluation of carcinogenicity: an updating of IARC monographs, IARC monographs on the evaluation of carcinogenic risks to humans. International Agency for Research on Cancer. 1987;1:42.
19. IARC. IARC monographs on the evaluation of carcinogenic risks to humans. In: CYP1A1 and aryl hydrocarbon receptor (AhR) in the placenta. *Curr Pharmacol Biotechnol*. 2010;12:715–30.
20. IARC. (2012). IARC Monographs 92, 100F Available at: <http://monographs.iarc.fr/ENG/Monographs/vol100F/mono100F-14.pdf> [Accessed on 20th July, 2017].
21. Huertero FA, Vega EA, Perez GM, Cuevas DV, Tinoco JT. Molecular markers associated with the biological response to aromatic hydrocarbons from urban air in humans. In: Moldoveanu AM, editor. Air pollution-new developments. London: InTech; 2011. p. 87–112.
22. Ziech D, Franco R, Pappa A, Panayiotidis MI. Reactive oxygen species (ROS)-induced genetic and epigenetic alterations in human carcinogenesis. *Mutat Res*. 2011;711(1):167–73.
23. Comandini A, Brezinsky K. Theoretical study of the formation of naphthalene from the radical/ π -bond addition between single-ring aromatic hydrocarbons. *J Phys Chem Anal*. 2011;115:5547–59.
24. Brezinsky K, Malewick T, Comandini A. Chemistry of polycyclic aromatic hydrocarbon formation from phenyl pyrolysis and reaction of phenyl and acetylene. *J Phys Chem*. 2012;116(10):2409–34.
25. Brazkova M, Krastanov A. Polycyclic aromatic hydrocarbons: sources, effects and biodegradation. *J Biol Chem*. 2013;10(2):52–6.
26. ATSDR (1995) Polycyclic Aromatic Hydrocarbons (PAHs), ATSDR Toxicological Profiles, US Department of Health and Human Services, Public Health Service, Agency for Toxic Substances and Disease Registry, CRC Press LLC,. Available at <http://www.atsdr.cdc.gov/toxprofiles/tp69.html> [Accessed on 17th July, 2017].
27. ATSDR, Agency for Toxic Substances and Disease Registry. (2006). Toxicological profile for polycyclic aromatic hydrocarbons. Atlanta, GA, USA. Available at: <http://www.food.gov.uk/sites/default/files/research-report-total-diet-study.pdf> [Accessed on 23th July, 2017].
28. Igwo-Ezikpe MN, Gbenle OG, Ilori MO, Okpuzor J, Osuotoki AA. Mixture of high molecular weight polycyclic aromatic hydrocarbons: biodegradation by tropical Bacteria and via co-metabolism with Phenanthrene. *Res J Environ Sci*. 2010;4:317–26.