

# EVALUATION OF PROBABLE SOURCES AND EXTENT OF BEEF CONTAMINATION BY SMOKE PAHS (POLYCYCLIC AROMATIC HYDROCARBONS) IN SELECTED TOWNS OF NORTHERN NIGERIA

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## ABSTRACT

Polycyclic Aromatic Hydrocarbons (PAHs) contamination of roasted beef collected from five towns in Northern Nigeria was assessed. The study revealed that the 16 priority listed PAHs contaminated roasted beef especially the USEPA human carcinogenic PAHs. The level of contamination was determined as % PAH contamination from smoke and was in the range of 0-96. The contamination of the beef by these PAHs was presumed to be by adsorption rather than absorption. Some of the PAHs like acenaphthylene, chrysene, benzo[a]pyrene, benzo[g,h,i] perylene among others significantly contaminated the beef samples at  $P < 0.05$ . Furthermore, the PAH4 were found to contribute higher concentrations from smoke than from other sources in the environment. In addition, the study showed that the source diagnostic ratio indicated that the PAHs that contaminated the beef were from pyrogenic sources rather than petrogenic sources. Generally, Benzo[a]pyrene and sum of PAH4 in the samples were within the Food Standard Agency limits of  $2 \mu\text{gkg}^{-1}$  and  $12 \mu\text{gkg}^{-1}$ .

**Keywords:** Adsorption, benzo[a]pyrene, contamination, PAHs, pyrogenic, petrogenic.

## 1.0 INTRODUCTION

A lot of animals are consumed daily in various forms. They may be eaten either as cooked, roasted or even fried meat. Pollution of an animal part in the course of the various forms of processing is possible. Food processing in the form of drying, smoking, grilling, roasting and frying have been associated with PAH generation (Zelinkova and Wenzl, 2015). One very prominent pollutant which abounds in the environment today is the polycyclic aromatic hydrocarbons (PAHs). They are produced from the incomplete combustion of materials in the environment (Lau *et al.*, 2010). Materials in the environment that could undergo combustion include garbage, petroleum products, coal, meat and tobacco. The PAHs which are also referred to as polynuclear aromatic hydrocarbons have been found to contain two or more rings which are fused together based on the number of rings in them.

The number of rings that are present in them can be used to classify them either as low molecular weight (LMW) or High Molecular Weight (HMW). It was originally viewed that PAHs can be obtained from only petroleum and its products (Muanya, 2006) but PAHs have now been associated with smoked and grilled meat (Suya) (Emerole *et al.*, 1982).

Analysis of charcoal roasted food products have indicated the presence of benzo[a]pyrene, anthracene, chrysene, benz[a]anthracene and indeno[1,2,3-cd]pyrene (Amos-Tautua *et al.* , 2013). The common polycyclic aromatic hydrocarbons based on their number of rings are naphthalene (2), acenaphthene, acenaphthylene, anthracene, phenanthrene and fluorene (3); fluoranthene, benz[a]anthracene, chrysene and pyrene (4); benzo[a]pyrene, benzo[b]fluoranthene and benzo[k]fluoranthene (5) while dibenz[a,h]anthracene, benzo[g,h,i]perylene and Indeno[1,2,3-cd]pyrene are 6 membered ring PAHs (Mzoughi and Chouba, 2012). One PAH that has been so linked with cancer is benzo[a]pyrene (Muanya, 2006). These compounds form epoxides due to metabolic activities propelled by a group of enzymes known as the CYP 1A (cytochrome phosphate 1A) and CYP 1B (cytochrome phosphate 1B) (Farombi, 2004). Essentially, the PAHs are transformed to chemicals that attach to substances in the body. The metabolites like epoxides which are reactive intermediates bind covalently to nucleic acids like deoxyribonucleic acid (DNA) leading to breakage and damage of the DNA. This damage results in mutation and tumor initiations (Harvey, 1997). Due to the fact that PAHs and the epoxides formed from them are highly toxic, mutagenic and carcinogenic to lower animals and human body systems, it is necessary to assess consumable foods for the quantity of PAHs that could enter the body.

Polycyclic Aromatic Hydrocarbons tend to emanate from natural and anthropogenic sources. PAHs from anthropogenic sources may be as a result of combustion of industrial materials, wastes among other forms of combustion (Ogungbuyi *et al.*, 2013). PAH sources from nature include bush fires and volcanic eruptions (Tobisaewski *et al.*, 2013). The ratio of some PAH will be ideally constant from the point of collection to when sample analysis takes place. Some of the PAHs used for source prediction include anthracene, phenanthrene, fluoranthene, pyrene, Benz[a]anthracene, chrysene, indeno[1,2,3-cd]pyrene and benzo[g,h,i]perylene.

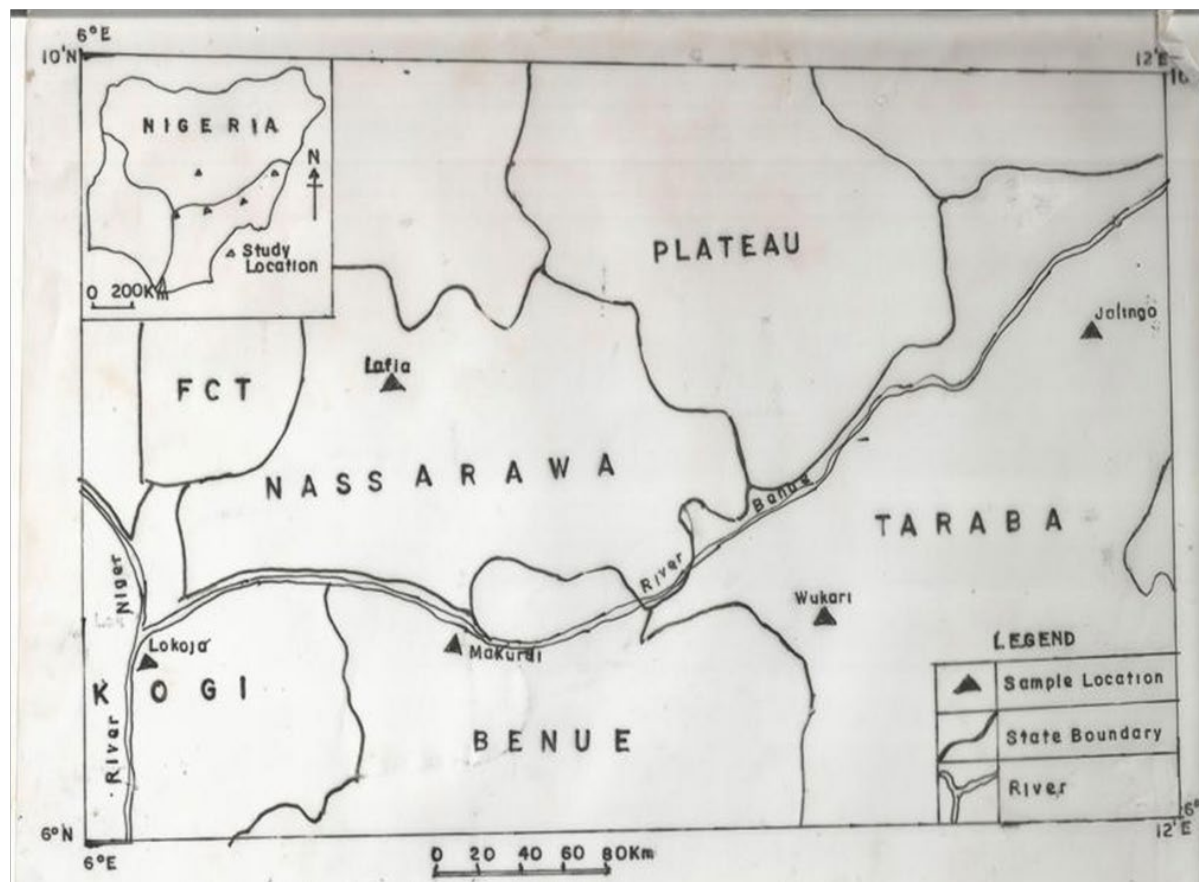
Furthermore, different methods of calculation have been reported for assessing environmental quality such as pollution indices. According to Hakanson (1980), a contamination factor describes the contamination of a given toxic substance by comparing the mean content of the substance to a reference level for substances. Furthermore, elemental and metal contamination indices have been developed based on the ratio of the difference between measured concentration ( $C_m$ ) and background concentration ( $B_m$ ) to the background concentrations which was rated on a scale from 0-100 (Meybeck *et al.*, 2004; Aikpokpodion *et al.*, 2010). There had been no records for assessing the pollution or contamination of PAH in samples contributed from smoke hence the need to monitor the extent of PAH contamination of beef surfaces from smoking.

The study was embarked upon bearing in mind that roasted beef is largely consumed in the Northern part of Nigeria especially among travelers on transit. Consequently, the study aims at determining the probable sources and contamination levels of the 16 EPA priority listed PAHs in beef samples collected from some towns in northern Nigeria.

## 2.0 MATERIALS AND METHODS

### 2.1 Study area

The study was conducted with samples collected from five locations namely Wukari and Jalingo in Taraba state, North Eastern Nigeria and Makurdi (Benue state), Lokoja (Kogi State) Lafia (Nassarawa) in the North central part of Nigeria.



## 2.2 Sample collection

A total of twenty (20) samples comprising fifteen (15) samples of the roasted samples and five (5) samples of the non roasted samples were obtained for the study. The beef prepared for roasting was obtained randomly from processors in the markets of the towns selected for the study. Part of the samples was processed by non smoke producing means while the other parts were processed normally by the processors.

## 2.3 Procedure for extraction of polycyclic aromatic hydrocarbons from the samples

Recovery experiment was carried out by spiking 3g of the pulverized beef with 1ppm of four deuterated PAH internal standards namely acenaphthene-d<sub>12</sub>, phenanthrene-d<sub>10</sub>, chrysene-d<sub>10</sub> and perylene-d<sub>12</sub>. The extraction was carried out using modified method 3550C (USEPA, 2007). Following recovery of 95.5-98.3 %, the extraction of the samples was then carried out. The pulverized samples were weighed into a 250 mL capacity beaker of borosilicate material and 50 mL of a ratio 3:1 (75 mL: 25 mL) redistilled hexane- dichloromethane mixture was added. The beaker and its content were placed in a sonicator to extract the hydrocarbons for thirty minutes. The organic layer was filtered into the 250 mL capacity borosilicate beaker. The extract was dried by passing the filtrate through a funnel containing anhydrous sodium sulphate. The dried extract was concentrated with a stream of nitrogen gas.

## 2.4 Clean up of extract

The extract in each case was cleaned up by separating it into the aliphatic and polycyclic aromatic hydrocarbons fractions as follows: Neutral alumina was packed into a column (up to 10 cm) and

cleaned properly with redistilled hexane. The extract was poured onto the alumina and allowed to run down with the aid of the redistilled hexane to elute the aliphatic profiles into a pre-cleaned 20 mL glass container. The aromatic fraction was eluted with a 3:1 mixture of hexane and dichloromethane to recover the non polar fractions while the most polar PAHs were recovered by eluting with dichloromethane into the pre-cleaned borosilicate beaker. The mixture was concentrated to 1 mL by a stream of nitrogen gas before gas chromatographic analysis using flame ionization detector (GC-FID).

## 2.5 Calibration of instrument (Gas Chromatograph)

Calibrations were obtained with standard solutions of concentrations ranging between 0.20 - 10 µg/l. Linearity was demonstrated by correlations  $r^2 \geq 0.999$ . The PAHs were identified in the samples by comparing them with the retention times of the peaks in the pure standard mix.

## 2.6 Instrumental analysis

Gas chromatographic parameters/conditions

Model: HP6890; Column: HP-1; Column length/column internal diameter/Column film: 30 m, 0.25 µm, 0.25 µm; Injection temperature: 250 °C;

Detector temperature: 320 °C; Detector: Flame ionization detector; Initial temperature: 60 °C for 5 min; First rate: 15 °C/min for 14 min and maintained for 3 mins; Second rate: 10 °C/min for 5 min and maintained for 4 mins; Mobile phase or carrier: Nitrogen; Nitrogen column pressure: 30 psi; Hydrogen pressure: 28 psi; Compressed air pressure: 32 psi

## 2.7 Quality control check

An accuracy check was carried out using blank after each 5 samples were analyzed to check if contamination from laboratory sources that would affect accuracy was present. The blank was prepared by substituting redistilled n-hexane in the place of the sample composite then performing the usual extraction procedure as described earlier.

## 2.8 Determination of Percentage (%) PAH contamination levels from smoke

The study introduced the concept of % PAH contamination level from smoke (PCL). The % PCL is calculated from the expression;

$$\% \text{ PAH contamination level from smoke (PCL)} = \frac{C_{SS} - C_{NS}}{C_{SS}} \times 100$$

$C_{SS}$  = Concentration of PAH in smoked samples

$C_{NS}$  = Concentration of PAH in non smoked samples

**Table 1: Interpretation of the % PAH contamination levels from smoke**

% contamination level from smoke	Interpretation
70-100	Very high contamination
50-69	High contamination
21-49	Fair contamination
1-20	Low contamination
0	No contamination

Source: From study

## 2.9 Statistical analysis

The data collected was analyzed for their mean values and standard deviations using MS excel software while t-test of significance was analyzed using SPSS 17 software at  $P < 0.05$ . These analyses

helped to determine the level of significant difference in the mean concentrations of the polycyclic aromatic hydrocarbons in the various samples.

### 3.0 DISCUSSION

The mean PAH concentrations ( $\mu\text{gkg}^{-1}$ ) in the roasted beef samples are presented in Table 2. The study revealed that the 16 priority listed PAHs contaminated the beef by varying degrees for all samples obtained from the different sample point. Benzo[a]pyrene concentration was the highest followed by a three ringed PAH, phenanthrene while the lowest concentrations were observed for naphthalene, acenaphthene and indeno[1,2,3-cd]pyrene. The concentrations of the PAH samples in the roasted beef did not follow any pattern in terms of molecular weight or other properties indicating that the distribution is based on their presence from the source of smoke. The total mean PAH concentration in the roasted beef is  $1.2519 \mu\text{gkg}^{-1}$  and the concentrations of the individual PAHs are in the range of  $0.0016\text{-}0.4819 \mu\text{gkg}^{-1}$ . The concentrations ( $\mu\text{gkg}^{-1}$ ) of the USEPA labelled human carcinogenic PAHs in the roasted samples are benz[a]anthracene (0.0295), benzo[a]pyrene (0.4819), benzo[b]fluoranthene (0.0030), benzo[k]fluoranthene (0.0039), chrysene (0.0254), dibenz[a,h]anthracene (0.0266) and benzo[g,h,i]perylene (0.0263). A study conducted in Amassomma (Amos Tautua *et al.*, 2013) revealed that concentrations were not detected in the roasted beef for naphthalene, acenaphthylene, fluorene, phenanthrene, anthracene, fluoranthene, pyrene, benzo[b]fluoranthene, benzo[a]pyrene, indeno[1,2,3-cd]pyrene, and dibenz[a,h]anthracene. The concentrations of benzo[a]anthracene ( $7.23 \mu\text{g}^{-1}$ ) and chrysene ( $2.95 \mu\text{g}^{-1}$ ) were found to be lower than detected for the samples in the present study. Furthermore, from another study which was carried out on smoked meat and other foods in Cote d'ivoire (Manda *et al.*, 2012), it was revealed that the concentrations of USEPA human carcinogenic PAHs in smoked meat had higher concentrations of benz[a]anthracene, benzo[a]pyrene, benzo[b]fluoranthene, chrysene, dibenz[a,h]Anthracene and benzo[g,h,i]perylene than was found for these PAHs in the present study. Only benzo[b]fluoranthene which was not detected in the smoked meat had relatively higher concentrations in the present study. Benzo[a]pyrene levels in charcoal grilled and smoked meat were found to be high in heat treated foods from China (Yong-Hong *et al.*, 2012) which may corroborate the higher levels of the B[a]P in the roasted beef studied relative to the other PAHs that were detected. In addition, a study conducted by Ogbuagu and Adedepo (2012) on roasted beef showed that the total concentrations of PAHs in the samples was  $0.0372 \text{ mg/kg}$  which is higher than the values reported in the present study.

PAHs were found to contaminate surfaces of smoked meat than their interior parts (Ciecieska and Obiezinki, 2007). This tends to suggest that PAH contamination of meat from smoke sources occur by adsorption rather than by absorption. Table 3 presents PAH concentration in non roasted beef whereby the total concentrations of the PAHs ( $0.4363 \mu\text{gkg}^{-1}$ ) is lower than determined for the roasted beef. The non- roasted beef were prepared by non -smoking sources hence the PAH contamination was from sources prior to smoking. The average background values have been reported to be usually in the range of  $0.01\text{-}1.00 \mu\text{gkg}^{-1}$  in uncooked foods (FAO/WHO, 2004). In addition to this there is a report that meat, milk, poultry and eggs do not record high levels of PAHs due to their rapid metabolisms in the species of origin (Food Safety Authority of Ireland, 2009). This could have been responsible for the low level of total PAH concentrations in the samples studied.

Since contamination could be due to adsorption rather than absorption, the extent of contamination of each PAH on the surface of the beef is required. Table 4 presents the extent of contamination from



smoke for each of the PAH detected in the study. The study introduced the concept of % PAH contamination levels from smoke in smoked samples. This was necessary since quantifying the level of PAH contamination of samples from the smoking would guide the analyst to infer on which sample source should be of concern especially regarding benzo[a]pyrene, chrysene, benzo[b]fluoranthene and benz[a]anthracene and other USEPA human carcinogenic PAH. The study therefore relied on these to develop the % PAH contamination levels from smoke based on the ratio of the difference between the concentration of a particular PAH in the roasted sample and the concentration of the PAH in the non-roasted sample to the concentration of the PAH in the roasted sample multiplied by 100. The reason for variations in contaminations of the PAH from the smoke to samples as shown by the PAH contamination levels is not clear but it goes to show that intensity of combustion could be a factor that enhances the deposition of some of the PAH. Generally, data reported in literature on quantitative basis have been shown to be highly variable (USEPA, 1993).

Contamination levels of 50 % and above reflects more contamination while less than 50 % is less contamination and 0 % indicates that the PAH did not contaminate the sample from smoke but from other environmental sources. Table 1 describes the extent of contaminations in % contamination ranges. To this extent, acenaphthylene, phenanthrene, benzo[a]anthracene, benzo[a]pyrene, dibenz[a,h]anthracene and benzo[g,h,i]perylene very highly contaminated the beef samples which corroborates with the test of significance level analysis at  $p < 0.05$  (Table 7). The implication is that the listed PAHs had difference in mean concentrations between the roasted and non-roasted beef that were statistically significant. The differences in mean concentrations of the other PAHs were not significant statistically. Generally, the other PAHs may have contaminated the samples from the smoke as shown by their contamination levels but their levels of contaminations were not significant. The B[a]P and PAH4 concentrations in the study are presented in Table 5. The values in this study were  $0.4819 \mu\text{gkg}^{-1}$  and  $0.5538 \mu\text{gkg}^{-1}$  respectively. However, the PAH4 content for smoked meat in a study conducted in Latvia was found to be  $1.22 \mu\text{gkg}^{-1}$  (Miculis *et al.*, 2011). This was higher than the values of PAH4 in the present study. The concentrations of the PAH4 contributed from the smoke were higher than that from other sources in the environment. They include benz[a]anthracene (0.0225, 0.0070); chrysene (0.0134, 0.0120) benzo[b]fluoranthene (0.0016, 0.0014) and benzo[a]pyrene (0.3603, 0.1216). This reveals that smoking could contribute these PAHs even more than other sources in the environment. The concentrations of some other PAHs though contributed from smoke were lower than those from other sources in the environment while acenaphthene and fluorene in the beef did not come from the smoke. This could imply that the concentrations of the PAHs from other sources to the beef degraded to lower concentrations in the smoked beef due to heat associated with the process. The % PAH contamination levels also indicated that the PAH4 had more contaminations from the smoke. The concentrations were generally below the Food Standard Agency permissible limits of  $2 \mu\text{gkg}^{-1}$  for benzo[a]pyrene and  $12 \mu\text{gkg}^{-1}$  for  $\Sigma\text{PAH}_4$ .

The study further revealed that from the diagnostic ratios presented in Table 6, the PAHs entered the samples via combustion. The other diagnostic ratio values also indicate that the sources of the PAHs were linked to combustion. Infact, Indeno[1,2,3-cd]pyrene: indeno[1,2,3-cd]pyrene + benzo[g,h,i]Perylene ratios reveal that the combustion of the roasted beef samples were actually attributed to coal, grass or wood sources. The interesting revelation from the source diagnostic ratio is that petrogenic sources did not contribute PAHs to the roasted beef. A study on smoked fish contaminations by PAHs in Ghana revealed that they were from petrogenic sources (Palm *et al.*, 2011). This contrasts the report of the present study.

## CONCLUSION

The study revealed that 16 priority listed PAHs contaminated roasted beef collected from some towns in Northern Nigeria. The contamination was presumed to be due to adsorption rather than absorption. The level of contamination was determined as % PAH contamination from smoke. Some of the USEPA human carcinogenic PAHs significantly contaminated the beef samples at  $P < 0.05$ . The PAH4 contributed higher concentrations from smoke than from other sources in the environment. However, Benzo[a]pyrene and sum of PAH4 in the samples were within the Food Standard Agency limits of  $2 \mu\text{kg}^{-1}$  and  $12 \mu\text{kg}^{-1}$  (Food Standard Agency, 2012)

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**Table 2:** Mean PAH concentrations ( $\mu\text{g}/\text{kg}$ ) in roasted beef samples (n=15)

PAH	Range	Mean**	SD
Naphthalene	0.0002-0.0027	0.0016	0.0012
Acenaphthylene	0.0043-0.0247	0.0119	0.0085
Acenaphthene	0.0066-0.0145	0.0097	0.0030
Fluorene	0.0012-0.1107	0.0250	0.0479
Phenanthrene	0.1068-0.5346	0.3752	0.2040
Anthracene	0.0051-7.0288	0.1437	0.2746
Fluoranthene	0.0055-0.2650	0.0611	0.1141
Pyrene	0.0317-0.1292	0.0865	0.0378
Benz[a]anthracene	0.0110-0.0332	0.0295	0.0017
Chrysene	0.0001-0.0590	0.0254	0.0259
Benzo[b]fluoranthene	0.0002-0.0078	0.0030	0.0286
Benzo[k]Fluoranthene	0.0002-0.0057	0.0039	0.0030
Benzo[a]pyrene	0.0040-	0.4819	0.3509



	0.8518		
Indeno[1,2,3-cd]pyrene	0.0003- 0.0034	0.0017	0.0011
Dibenz[a,h]anthracene	0.0026- 0.0354	0.0266	0.0262
Benzo[g,h,i]perylene	0.0009- 0.0354	0.0263	0.0273
Total PAH		1.2619	

**Table 3:** PAH concentrations ( $\mu\text{g}/\text{kg}$ ) in non-roasted beef

PAH		Mean	SD
Naphthalene	0.0003- 0.0012	0.0006	0.0005
Acenaphthylene	0.0004- 0.0066	0.0034	0.0026
Acenaphthene	0.0027- 0.0378	0.0102	0.0015
Fluorene	0.0027- 0.1378	0.0565	0.0731
Phenanthrene	0.0020- 0.0492	0.0235	0.0192
Anthracene	0.0378- 0.1893	0.0735	0.0727
Fluoranthene	0.0148- 0.1178	0.0545	0.0366
Pyrene	0.0381- 0.0907	0.0648	0.0204
Benz[a]anthracene	0.0016- 0.0161	0.0070	0.0071
Chrysene	0.0001- 0.0318	0.0120	0.0164
Benzo[b]fluoranthene	0.0007- 0.0026	0.0014	0.0008
Benzo[k]Fluoranthene	0.0001- 0.0038	0.0016	0.0019
Benzo[a]pyrene	0.0037- 0.3002	0.1216	0.1608
Indeno[1,2,3-cd]pyrene	0.0001- 0.0115	0.0007	0.0007
Dibenz[a,h]anthracene	0.0011- 0.0059	0.0035	0.0022
Benzo[g,h,i]perylene	0.0002- 0.0022	0.0010	0.0009
Total PAH		0.4363	

**Table 4:** PAH concentrations ( $\mu\text{g}/\text{kg}$ ) from smoke in roasted beef (RB) samples

PAH	PAH Conc. in RB	PAH Conc. in NRB	PAH Conc. from smoke	% contamination level of PAH from smoke
Naphthalene	0.0016	0.0011	0.0005	31
Acenaphthylene	0.0119	0.0033	0.0086	72
Acenaphthene	0.0097	0.0102	0.0000	00
Fluorene	0.0250	0.0565	0.0000	00
Phenanthrene	0.3141	0.0236	0.2906	93
Anthracene	0.1437	0.0735	0.0702	49
Fluoranthene	0.0611	0.0545	0.0066	11
Pyrene	0.0865	0.0648	0.0217	25
Benz[a]anthracene	0.0295	0.0070	0.0225	89

Chrysene	0.0254	0.0120	0.0134	53
Benzo[b]fluoranthene	0.0030	0.0014	0.0016	53
Benzo[k]Fluoranthene	0.0039	0.0016	0.0023	59
Benzo[a]pyrene	0.4819	0.1216	0.3603	75
Indeno[1,2,3-cd]pyrene	0.0017	0.0007	0.0001	59
Dibenz[a,h]anthracene	0.0266	0.0035	0.0231	87
Benzo[g,h,i]perylene	0.0263	0.0010	0.0253	96

**Table 5:** B[a]P and  $\Sigma$ PAH4 concentrations ( $\mu\text{g}/\text{kg}$ ) in Roasted beef

Sample	Jalingo	Wukari	Lokoja	Lafia	Makurdi	Mean
Benzo[a]pyrene	0.0040	0.2461	0.7332	0.8518	0.5742	0.4819
$\Sigma$ PAH	0.0274	0.3677	0.8285	0.8676	0.6780	0.5538

**Table 6:** Source predictor values relative to Roasted and non-Roasted samples

Diagnostic ratio	Petrogenic	Fuel combustion	Coal, grass, wood burning	RB in the study	NRB in the study
$\frac{\text{Anthracene}}{(\text{Anthracene} + \text{Phenanthrene})}$	<0.1	>0.1	-	0.31	0.76
$\frac{\text{Fluoranthene}}{(\text{Fluoranthene} + \text{Pyrene})}$	<0.4	0.4-0.5	>0.5	0.41	0.46
$\frac{\text{Benz[a]Anthracene}}{(\text{Benz[a]Anthracene} + \text{Chrysene})}$	<0.2	>0.35	0.2-0.35	0.54	0.37
$\frac{\text{Indeno[1,2,3-cd]Pyrene}}{(\text{Indeno[1,2,3-cd]pyrene} + \text{Benzo[g,h,i]Perylene})}$	<0.2	0.2-0.5	>0.5	16.4	0.41

Source: Tobiszewski, 2014; present study

Table 7: T-test analysis result of 16 priority listed PAHs between roasted and non roasted beef

S/NO	Name of PAHs	t-value, $p < 0.05$
1.	Naphthalene	0.072 <sup>a</sup>
2.	Acenaphthylene	0.034 <sup>b</sup>
3.	Acenaphthene	0.949 <sup>a</sup>
4.	Fluorene	0.257 <sup>a</sup>
5.	Phenanthrene	0.002 <sup>b</sup>
6.	Anthracene	0.359 <sup>a</sup>
7.	Fluoranthene	0.840 <sup>a</sup>
8.	Pyrene	0.136 <sup>a</sup>
9.	Benzo[a]anthracene	0.003 <sup>b</sup>

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10.	Chrysene	0.203 <sup>a</sup>
11.	Benzo[b]fluoranthene	0.222 <sup>a</sup>
12.	Benzo[k]fluoranthene	0.065 <sup>a</sup>
13.	Benzo[a]pyrene	0.031 <sup>b</sup>
14.	Indeno[1,2,3-cd]Pyrene	0.045 <sup>b</sup>
15.	Dibenz[a,h]anthracene	0.054 <sup>b</sup>
16.	Benzo[g,h,i]perylene	0.002 <sup>b</sup>

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a- Indicates no significant difference in means; b- indicates significant difference in means

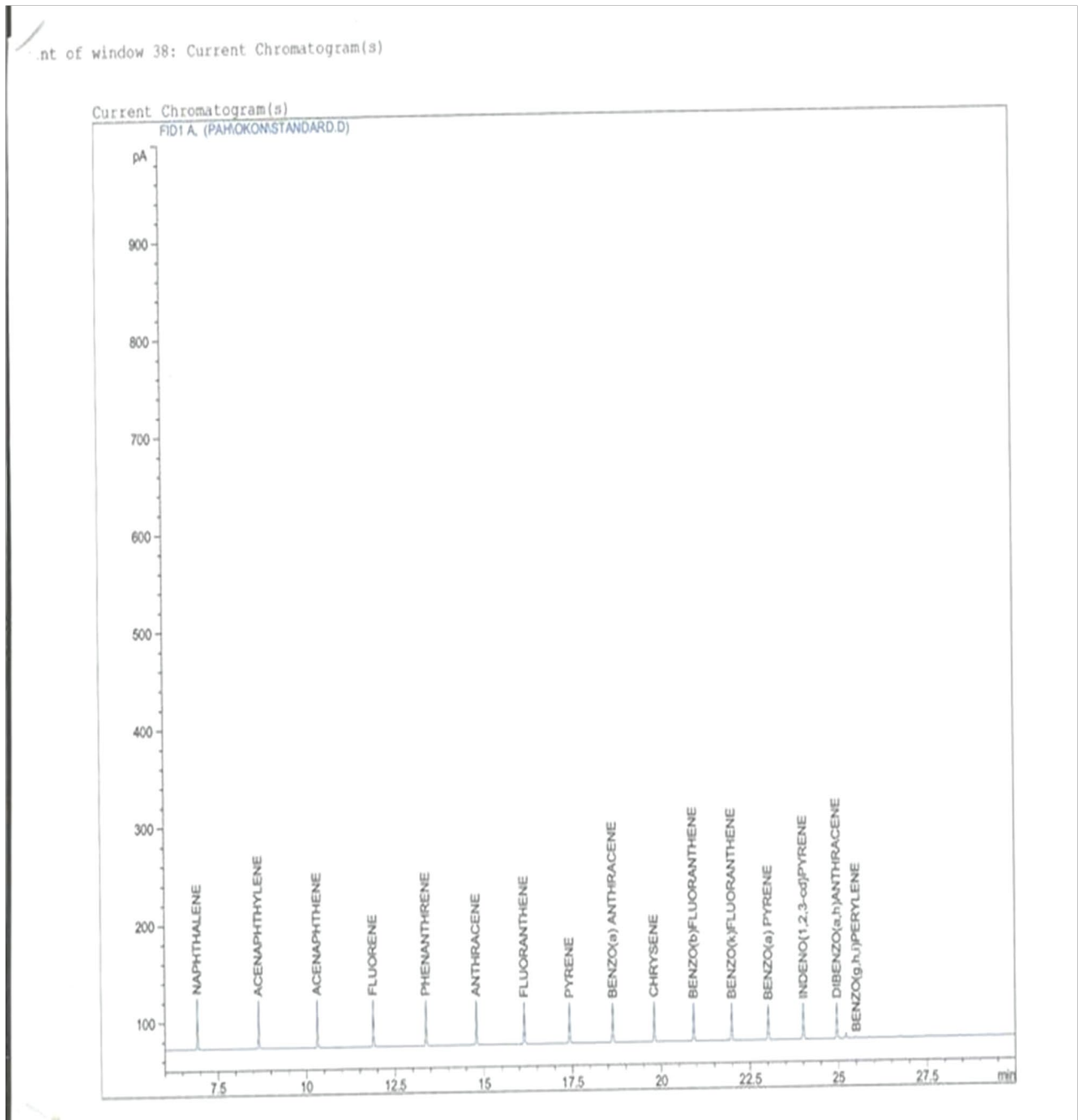
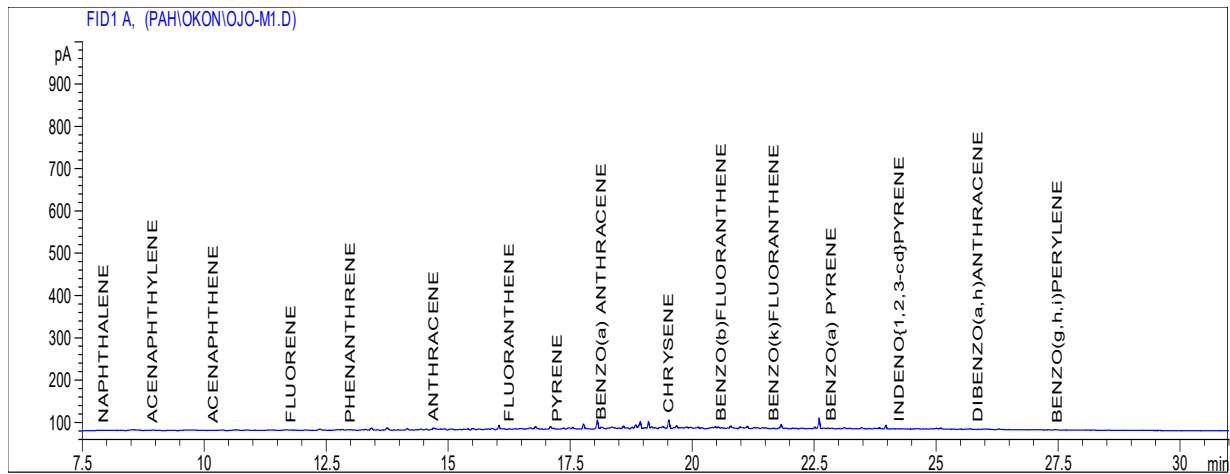
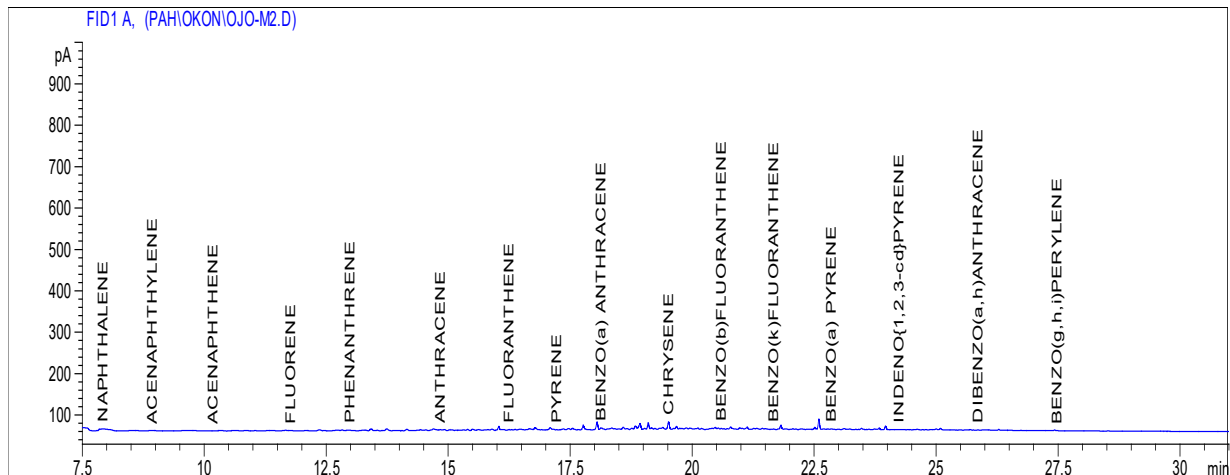


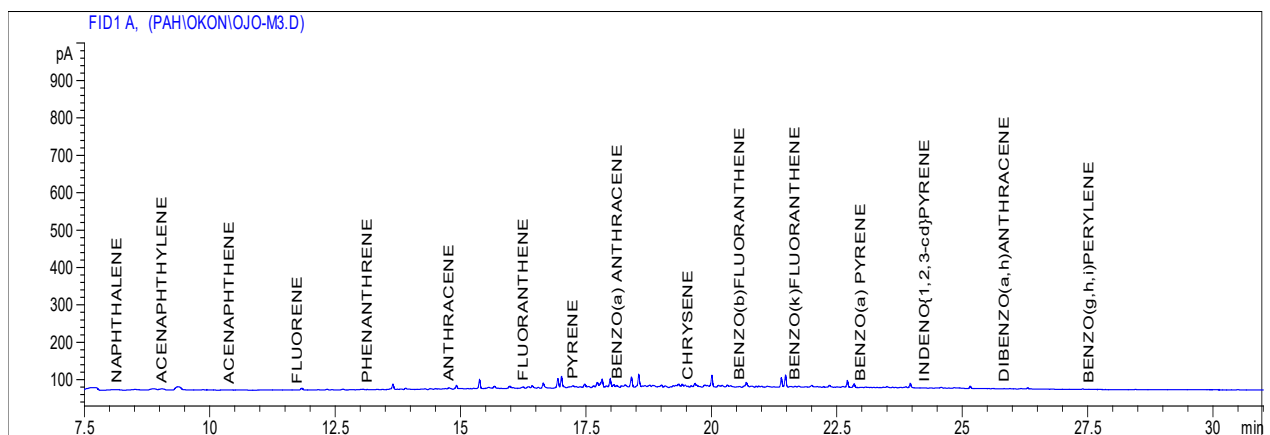
Fig 1: Chromatogram for Pure standard mix



Chromatogram for roasted beef from Sample location 1A (Wukari)

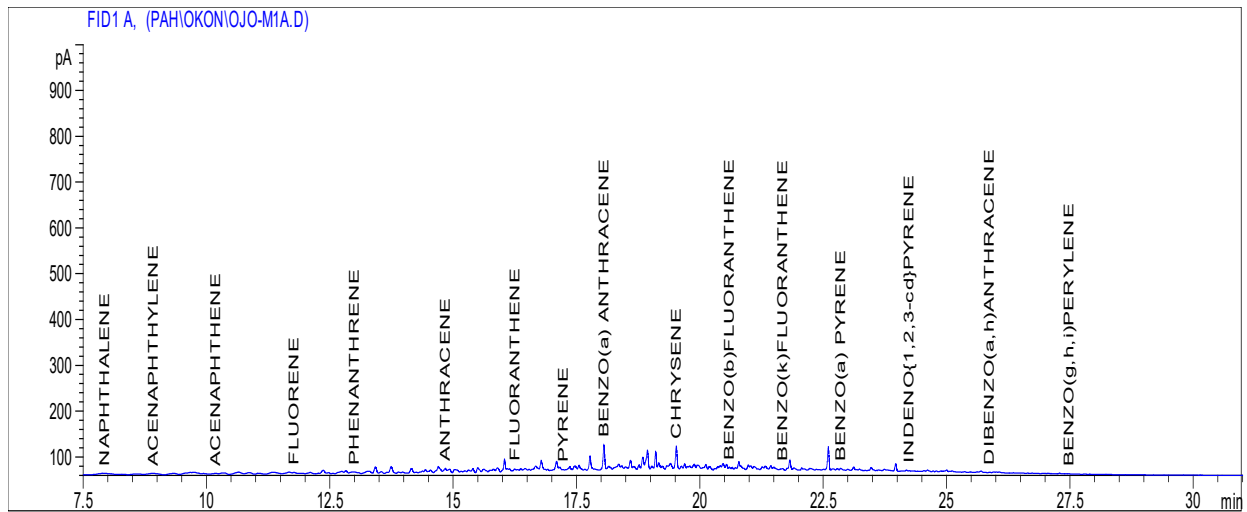


Chromatogram for roasted beef from Sample location 1B (Wukari)

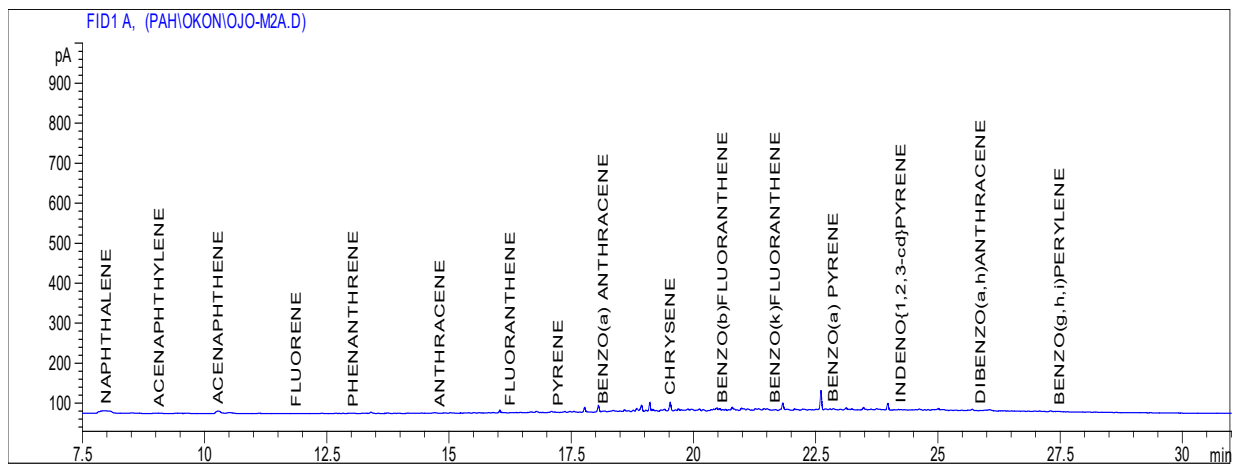


Chromatogram for Roasted beef from Sample location 1C (Wukari)

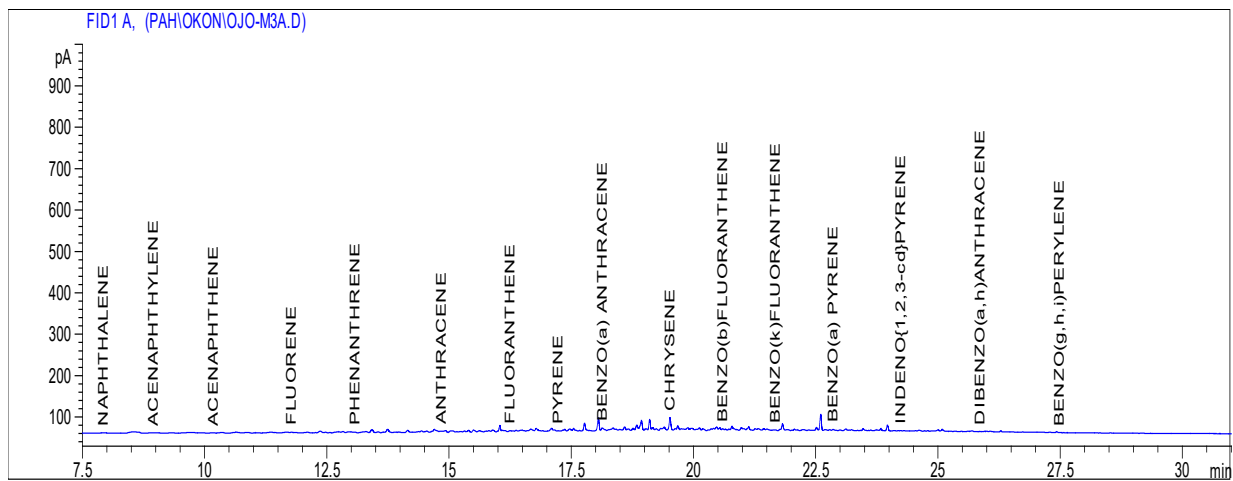




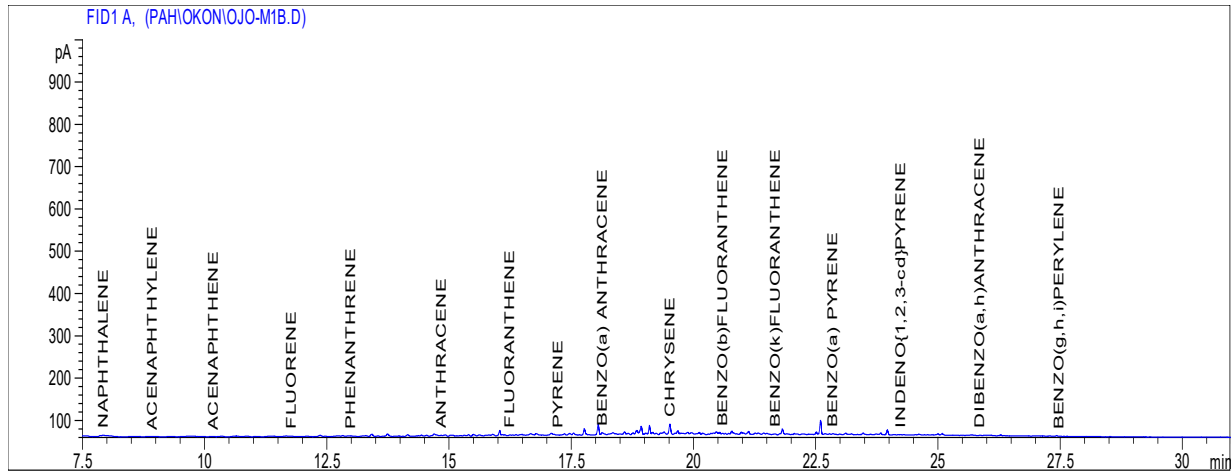
Chromatogram for Roasted beef from Sample location 2A (Lokoja)



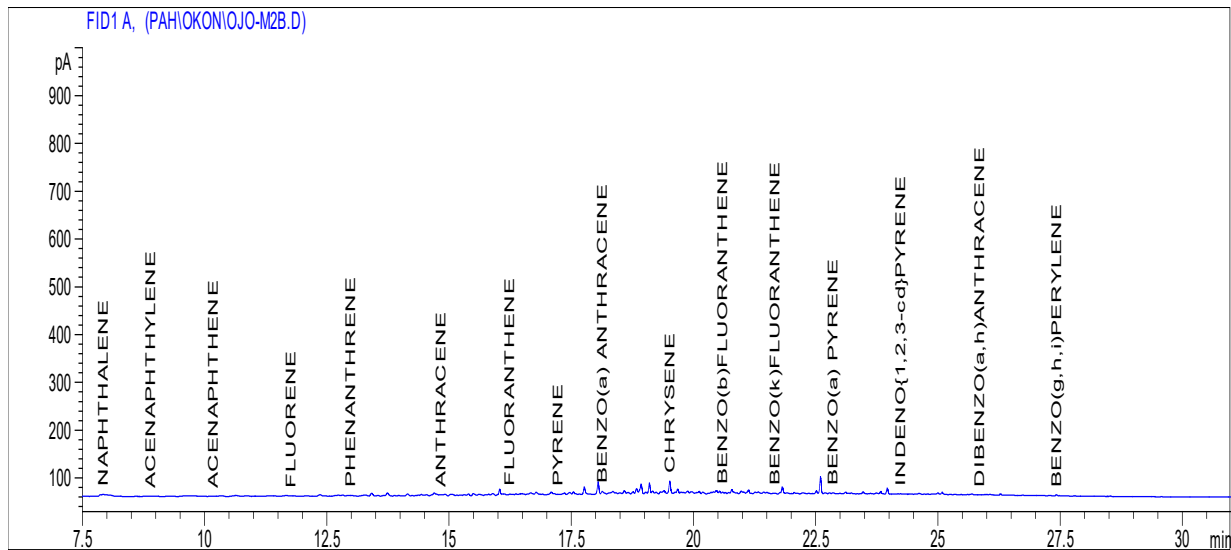
Chromatogram for Roasted beef from Sample location 2B (Lokoja)



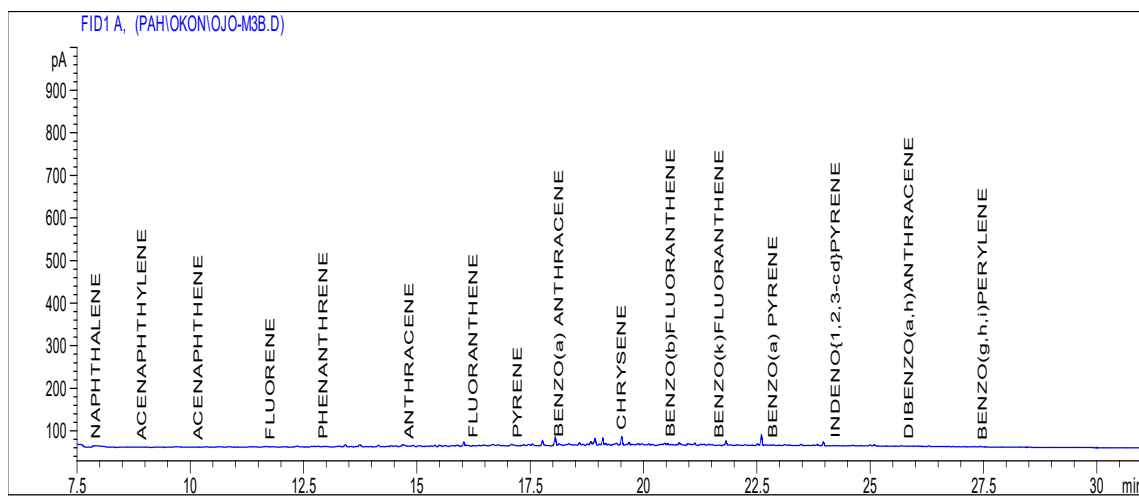
Chromatogram for Roasted beef from Sample location 2C (Lokoja)



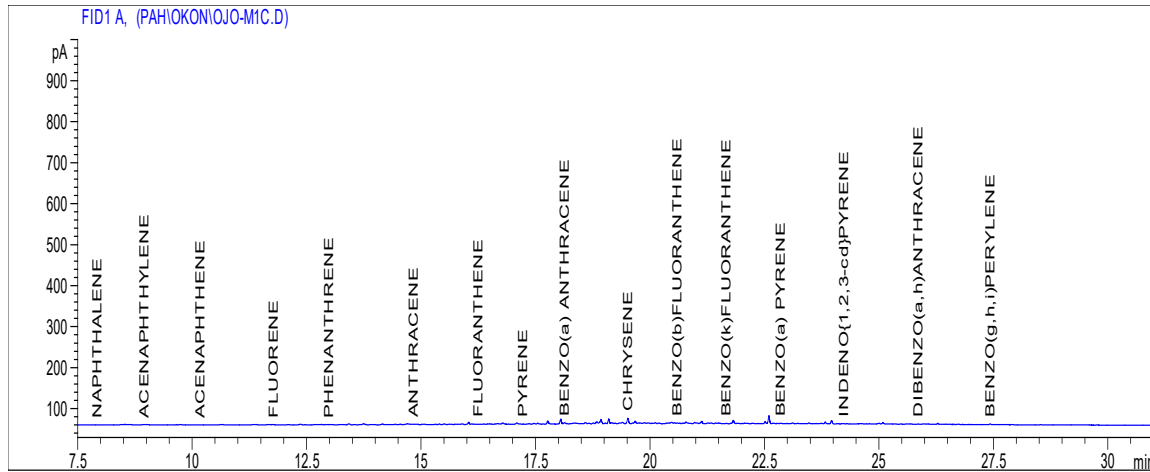
Chromatogram for Roasted beef from Sample location 3A (Jalingo)



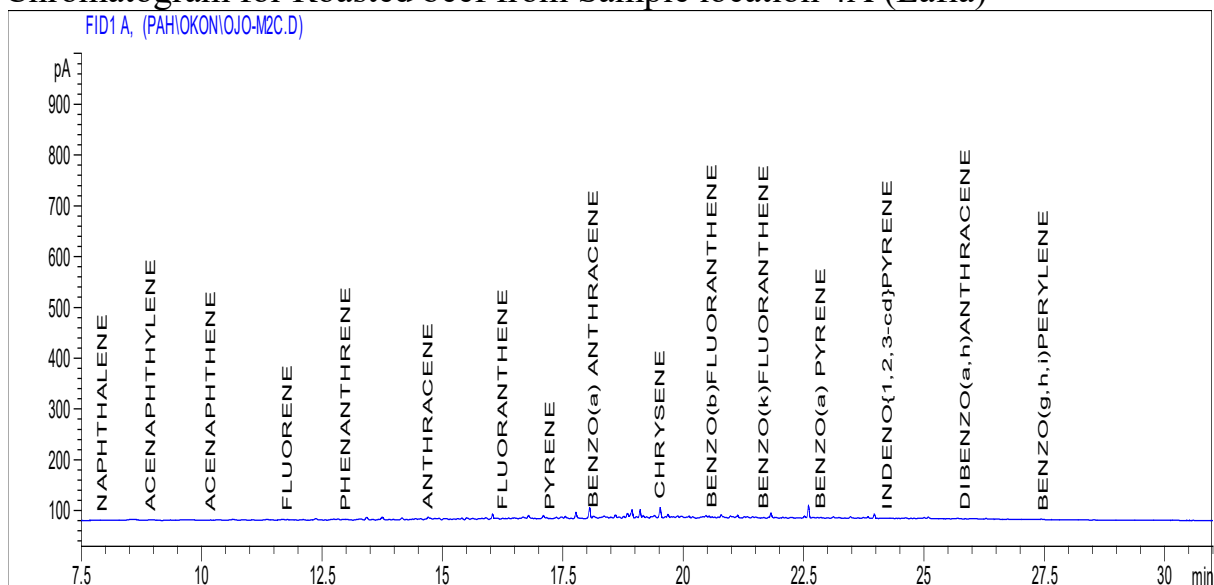
Chromatogram for Roasted beef from Sample location 3B (Jalingo)



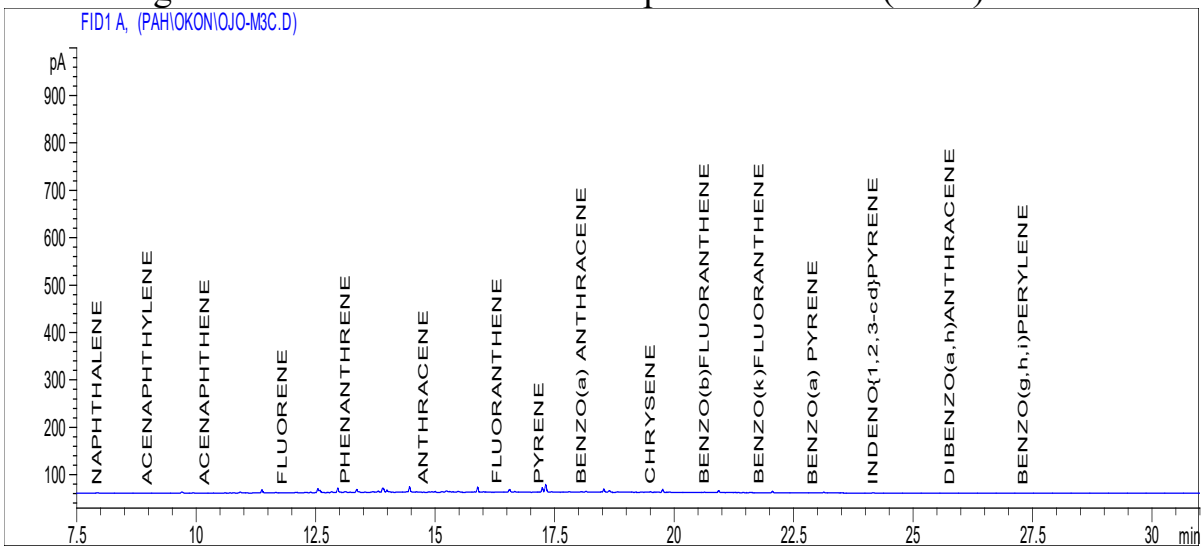
Chromatogram for Roasted beef from Sample location 3C (Jalingo)



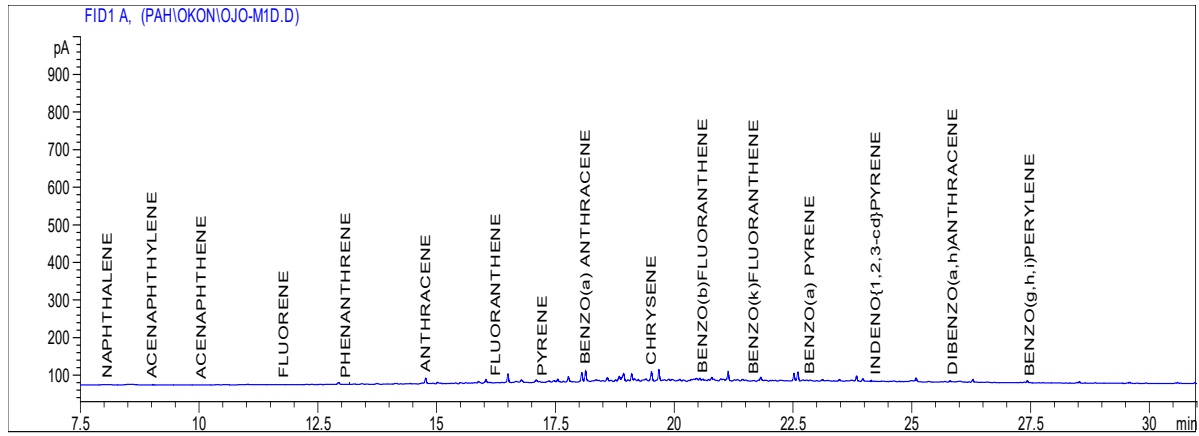
Chromatogram for Roasted beef from Sample location 4A (Lafia)



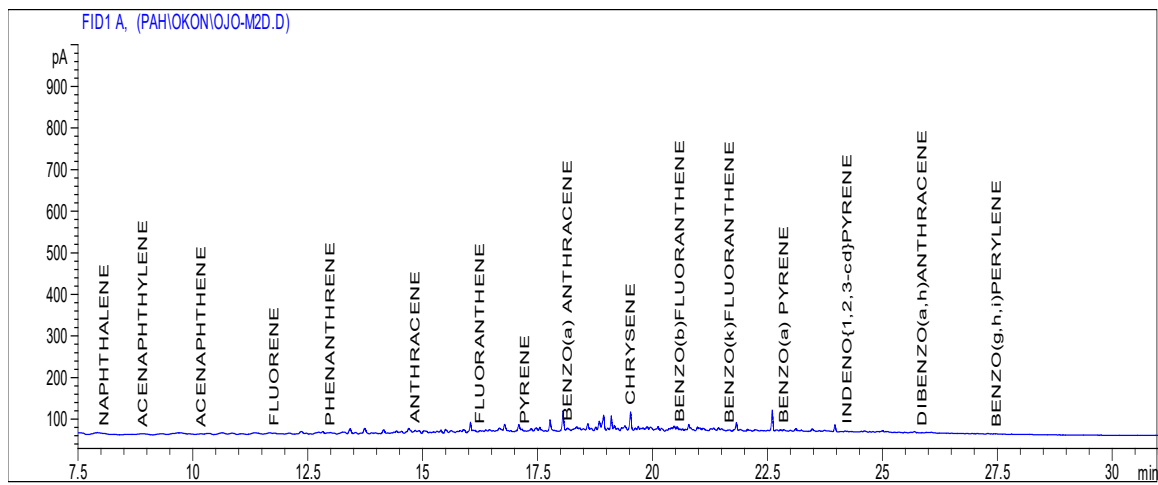
Chromatogram for Roasted beef from Sample location 4B (Lafia)



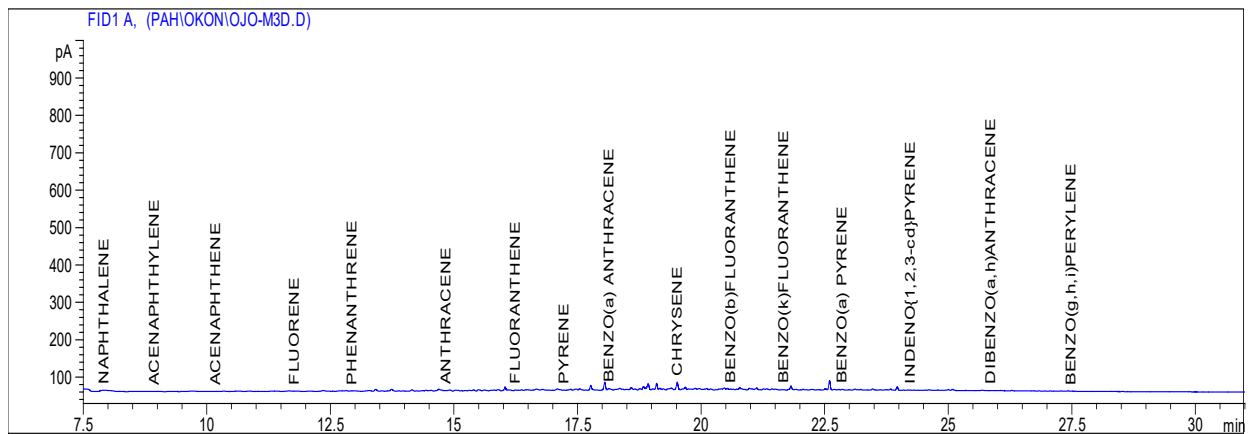
Chromatogram for Roasted beef from Sample location 4C (Lafia)



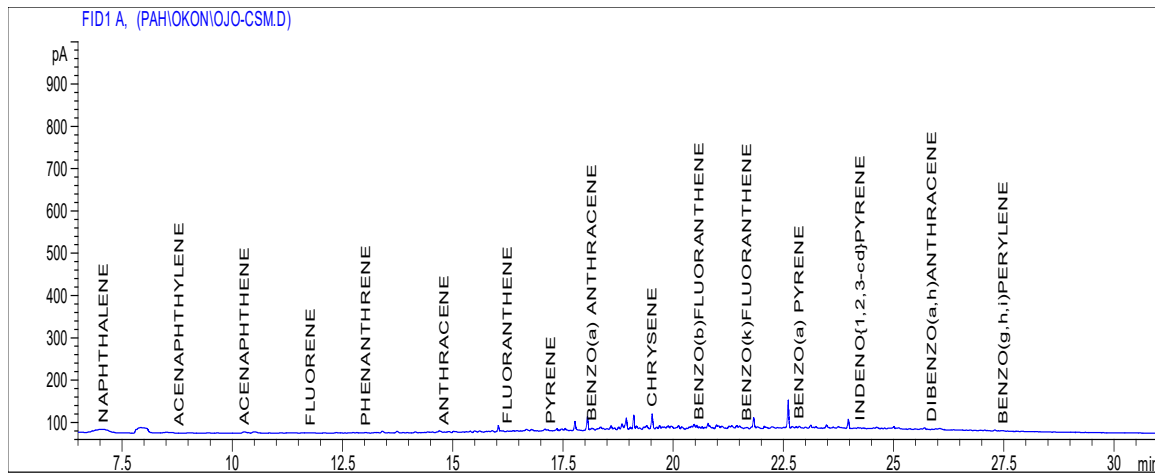
Chromatogram for Roasted beef from Sample location 5A (Makurdi)



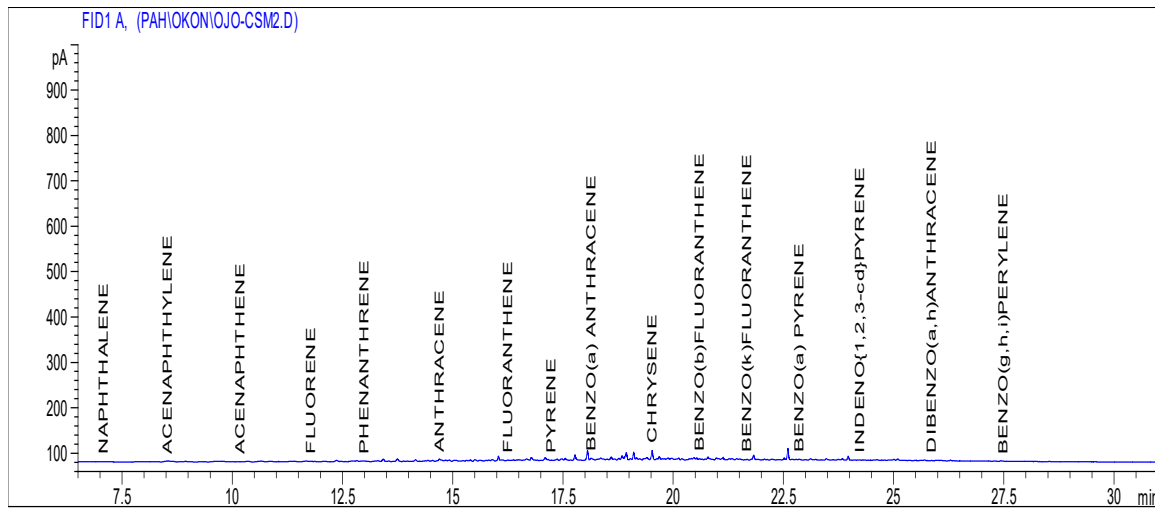
Chromatogram for Roasted beef from Sample location 5B (Makurdi)



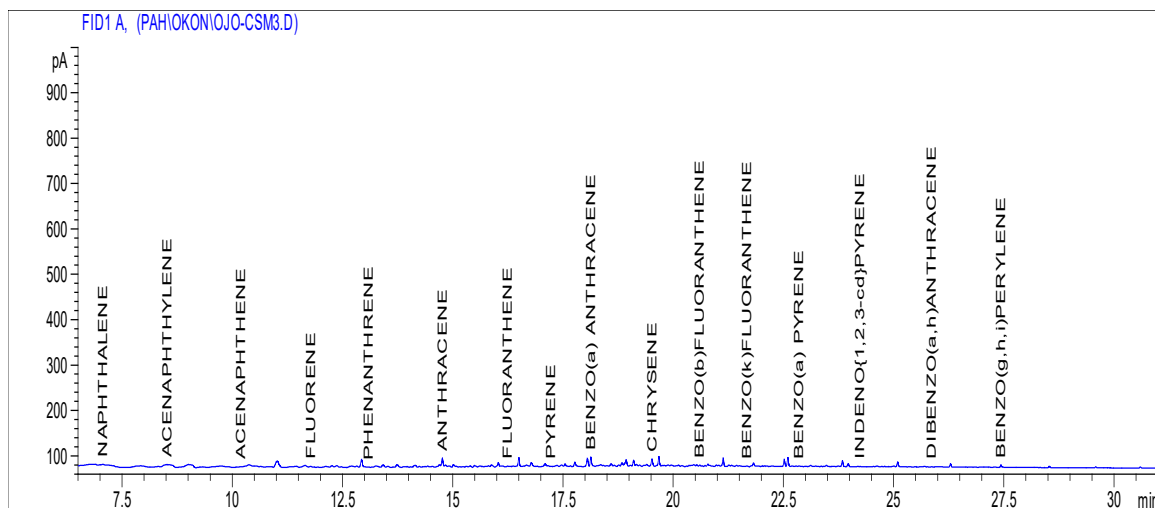
Chromatogram for Roasted beef from Sample location 5C (Makurdi)



Chromatogram for non-roasted beef from Sample Location 1 (Wukari)

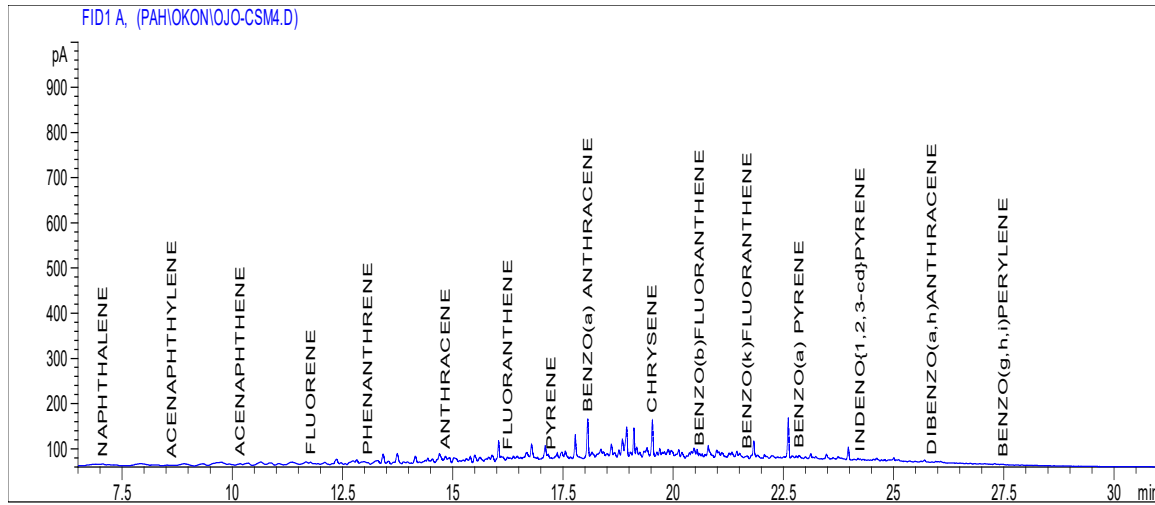


Chromatogram for non-roasted beef from Sample Location 2 (Jalingo)

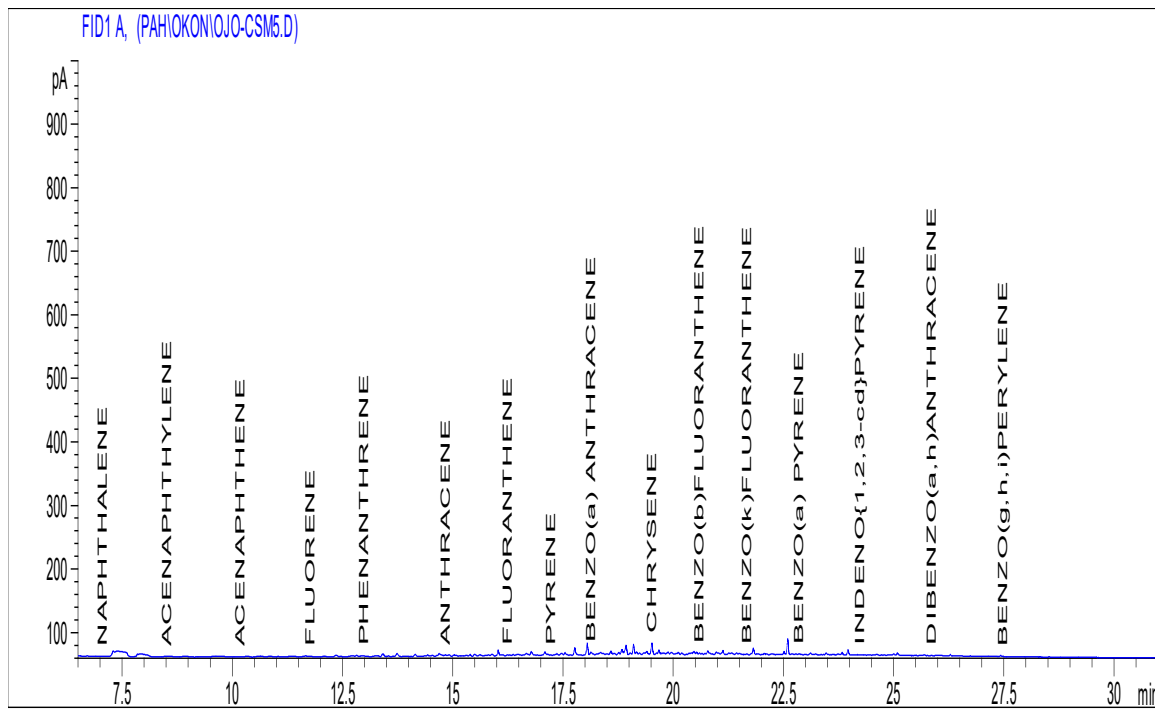


Chromatogram for non-Roasted beef from Sample Location 3 (Lafia)





Chromatogram for non roasted beef from Sample Location 4(Lokoja)



Chromatogram for non smoked beef from Sample Location 5(Makurdi)