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COMPARISON OF POLYCYCLIC AROMATIC HYDROCARBONS CONTAMINATION LEVELS IN ROASTED FOODS (PLANTAIN, MAIZE & YAM) AND BEVERAGES (MILO, MYCHOCO, LIPTON & NESCAFE) CONSUMED IN ANAMBRA STATE

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ABSTRACT

Polycyclic aromatic hydrocarbons (PAHs) are a class of hazardous organic chemicals consisting of three or more fused benzene rings, which can exist in linear, angular, and group arrangements. This study aimed to determine and compare PAH contamination levels and emission sources in roasted foods and beverages from road vendors and markets, respectively, in major cities in Anambra State, Nigeria. A total of 14 PAH samples were extracted using a Soxhlet extractor, and the extracts were cleaned up. The PAHs analysis was performed using gas chromatography coupled with a flame ionization detector (GC-FID). The results showed that the ∑16 PAHs (× 10-2 µg/kg) in roasted foods ranged from 1.500±1.873 in roasted yams to 9.074±8.120 in plantains, and in beverages, ∑16 PAHs ranged from 36.25±0.01 in Nescafé to 90.04±0.01 in Mychoco. The ∑HMW PAHs in roasted foods ranged from 0.028±0.000 in yams to 6.090±4.142 in maize, while for beverages, ∑HMW PAHs varied from 25.48±0.01 at 70.3% in Nescafé to 66.48±0.03 at 73.8% in Mychoco. The ∑LMW PAHs in roasted foods varied from 2.824±1.892 in yams to 9.759±7.861 in maize, and in beverages, they ranged from 9.94±0.01 at 15.0% in Lipton to 23.56±0.02 at 26.2% in Mychoco. The ∑PAH8 contamination level (× 10-2 µg/kg) in beverages ranged from 22.98±1.00 in Nescafé to 63.38±1.03 in Mychoco, while in roasted foods it ranged from 0.028±0.000 in yams to 3.090±4.124 in maize. The PAHs detected in the analyzed samples were below the permissible limit of 1.0µg/kg by EFSA for cereal and cereal-based products and are considered safe for consumption.

Keywords: Benzo[a]pyrene; Carcinogenic; Food; Gas Chromatography; Mychoco; Polycyclic Aromatic Hydrocarbons.

1.0. Introduction

More than 200 chemical substances with two or more fused aromatic rings are classified as polycyclic aromatic hydrocarbons (PAHs) (Domingo et al., 2015) and can be categorized as light compounds (two to three rings) or heavy compounds (four to six rings) based on the number of aromatic rings

(Purcaro et al., 2013). In addition to natural sources such as forest fires and volcanic emissions, environmental PAHs can also come from anthropogenic sources such as coal burning, car exhaust emissions, engine lubricants and cigarette smoke (Amirdivani et al., 2019).

The pyrolytic process that produces PAHs involves three fundamental factors: high temperatures, reduced oxygen levels, and organic matter resulting from incomplete combustion. This process typically yields a complex mixture of PAHs that can accumulate in the environment, affecting water, air, and soil, and ultimately entering the food chain (Zelinkova et al., 2015; Abdel-Shafy et al., 2016). PAHs are among the genotoxic, mutagenic, and carcinogenic compounds formed during the thermal processing of foods (Abid et al. 2014; Rengarajan et al., 2015). Recent epidemiological studies with humans and animals have indicated that the increase in cancer prevalence can be partially attributed to PAH exposure (EU Commission, 2011; Purcaro et al., 2013; Domingo et al., 2015).

Cancer is becoming the second most common non-infectious disease worldwide due to significant changes in the population and lifestyle (Ferlay et al., 2021). Cancer is the second most common cause of mortality worldwide, after heart disease. In 2020, the World Health Organisation (WHO), an intergovernmental agency, and the International Agency for Research on Cancer (IARC) released a report detailing 10.0 million cancer-related deaths and 19.30 million new cases (Ferlay et al., 2021).

Drinks that are used for enjoyment, nutrition, and hydration are known as beverages. (Hewlings and Kalman, 2017). To accommodate a range of tastes and preferences, they are available in various shapes, flavours, and textures. Because they offer enjoyment, sustenance, and social interaction, beverages are an essential component of human society (Hewlings and Kalman, 2017). They comprise soft drinks, juices, tea, coffee, milk, energy drinks, cheese, wine, and beer. They help maintain body fluid and electrolyte balance, provide essential vitamins, minerals, and antioxidants, and contribute to daily caloric intake.

The foods are cooked by roasting, which uses dry heat and usually temperatures between 300°F (150°C) and 425°F (220°C). Dry heat is used to improve the taste, texture and nutritional value of roasted meals, which can be prepared in an oven, on a grill or over an open flame. Roasting enhances a food's flavour profile, brings out its inherent sweetness, and gives it a delightful crunch. PAHs in food can originate from contaminated environments or form during thermal processing, such as when meat is grilled or smoked. Studies confirmed that PAHs are present in thermally processed meat, smoked cheese, coffee and tea, edible oils, toasted bread, rice, beans, bambara nut, pigeon peas, guinea corn, (Odika et al.2018; Odika et al. 2022) pasta (Ihedioha et al., 2019) roasted maize, yam and plantain (Olabemiwo et al., 2013; Amos-Tautua et al., 2013; Eze et al., 2019), roasted fish, suya beef (Amos-Tautua et al., 2013) chocolate (Kumari et al., 2012; Sadowska-Rociek et al., 2015) milk (Shariatifar et al., 2019), cocoa beans (Ciecierska, 2020)

2.0. Materials and Methods

2.1. Equipment and Reagents

Gas chromatography/flame ionization detector (HP Agilent 7820A Powered with HP ChemStation), borosilicate beaker, chromatographic column, Analytical balance, Conical flask, Soxhlet extraction kit, Rotary evaporator (Model: Heidolph HB

Digital, 20-1800 °C, coupled with BUCHI, Switzerland. All analytical-grade reagents and solvents were obtained from Sigma-Aldrich U.S.A. This included hexane, dichloromethane, sodium sulphate, ethanol, and activated alumina, as well as four deuterated (surrogate) standards, namely acenaphthalene d10, chrysene d12, phenanthrene d10, and perylene $d_{12.}$. The analysis was carried out at Docchy Analytical Laboratories and Environmental Services Limited, No. 30 Onwura Street, Adjacent Rock Foundation Hospital, Awka, Anambra State, Nigeria.

2.2. Sampling

Twenty-four (24) samples, which included different brands of beverages (Milo, Mychoco, Lipton, and Nescafé), were purchased from major markets- Eke-Awka and Onitsha main market in Anambra state, Nigeria. Roasted foods (roasted corn, roasted plantain, and roasted yams) were purchased from roadside vendors in two different cities, Awka and Onitsha, in Anambra State, Nigeria. The samples were collected, stored in labelled polyethene bags, and then taken to the laboratory.

2.3. Extraction

By spiking mixed PAHs standard solutions of varying concentrations into samples that were extracted using a soxhlet extractor with dichloromethane and n-hexane, recovery studies were conducted to maximise the extraction of PAHs from the samples. The solvent mixture was used to clean the extracts in an alumina column. (Oyekunle et al. 2011) All samples were extracted and cleaned up using the same procedure and solvent mixture after the good recovery result.

2.4. PAHs Determination in the Samples

All samples were each analyzed of PAHs using GC/FID under the described instrument parameters. The initial oven temperature programme was 60 ° C for 5 min, then increased to 250 ° C at 15 ° C / min, maintained at this temperature for 14 min, and finally increased to 10 ° C / min for 5 min. Nitrogen gas was used as the carrier gas at a flow rate of 1 cm3/min at a pressure of 30 psi. Retention time was used to characterize each PAH.

3.0. Results and Discussion

Table 1.0: PAHs Concentration levels ($\times 10^{-2}$ µg/kg) in Beverages

S/N	PAH	MILO	MYCHOCO	NESCAFE	LIPTON
1	Naphthalene	5.08±0.01	5.16±0.01	5.07±0.01	5.89±0.01
2	Acenapthlene	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00
3	Acenaphthene	0.00±0.00	18.40±0.00	0.00±0.00	0.00±0.00
4	Flourene	5.20±0.00	0.00±0.00	0.00±0.00	0.00±0.00
5	Phenanthrene	0.00±0.00	0.00±0.00	5.70±0.01	4.05±0.01
6	Anthracene	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00
7	Fluoranthene	0.00±0.00	0.00±0.00	0.00±0.00	1.60±0.01
8	Pyrene	0.00±0.00	3.10±0.01	2.50±0.01	24.00±0.01

9	Benzo[a]anthracene	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00
10	Chrysene	8.92±0.00	0.00±0.00	0.00±0.00	0.00±0.00
11	Benzo[b]Fluoranthene	21.90±0.01	49.67±0.01	13.68±0.01	14.70±0.01
12	Benzo[k]Fluoranthene	0.00±0.00	0.00±0.00	2.40±0.01	0.00±0.00
13	Benzo[a]pyrene	0.00±0.00	13.71±0.01	0.00±0.00	0.00±0.00
14	Indeno[1,2.3-cd]pyrene	0.00±0.00	0.00±0.00	4.70±0.01	0.00±0.00
15	Dibenz[a,h]anthracene	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00
16	Benzo[g,h,i]perylene	0.00±0.00	0.00±0.00	2.20±0.01	16.00±0.01
	∑16 PAHs	41.10±0.01	90.04±0.01	36.25±0.01	66.24±0.01
	∑LMW PAHs	10.08±0.01	23.56±0.01	10.77±0.01	9.94±0.01
	∑HMW PAHs	30.82±0.01	66.48±0.01	25.48±0.01	56.30±0.01
	∑8PAH	30.82±0.01	63.38±0.01	22.98±0.02	30.70±0.02

Table 2.0: The PAH Concentrations (× 10-2 µg/kg) in the Analyzed Roasted Foods

	PAH	**Plantain**	**Maize**	**Yam**
1	Naphthalene	ND	ND	ND
2	Acenaphthylene	ND	8.046±0.000	ND
3	Acenaphthene	2.379±3.365	0.000±0.000	ND
4	Fluorene	1.433±2.027	1.592±2.234	2.796±0.000
5	Phenanthrene	2.310±3.218	0.091±0.040	0.016±0.001
6	Anthracene	1.406±1.936	0.028±0.023	0.072±0.086
7	Fluoranthene	0.301±0.393	0.026±0.000	ND
8	Pyrene	0.003±0.000	ND	ND
9	Benzo[a]anthracene	1.036±1.445	0.114±0.000	0.020±0.000
10	Chrysene	0.027±0.028	0.112±0.000	0.008±0.000
11	Benzo[b]fluoranthene	0.067±0.000	0.116±0.082	ND

12	Benzo[k]fluoranthene	0.133±0.170	0.845±0.000	ND
13	Benzo[a]pyrene	0.029±0.000	ND	ND
14	Indeno[1,2,3-cd]pyrene	ND	0.034±0.000	ND
15	Dibenzo[a,h]anthracene	ND	4.844±0.000	ND
16	Benzo[g,h,i]perylene	ND	ND	ND
	∑16 PAHs	9.074±8.120	8.839±3.719	1.500±1.873
	∑LMW PAHs	7.529±6.492	9.759±7.861	2.824±1.892
	∑HMW PAHs	1.595±1.627	6.090±4.142	0.028±0.000
	PAH2	0.041±0.008	0.112±0.000	0.008±0.000
	PAH4	1.110±1.406	0.229±0.077	0.028±0.000
	PAH8	1.244±1.236	3.090±4.124	0.028±0.000

ND = Not detected

The accuracy and precision of the determination were assessed using three spiking concentrations, enabling the plotting of calibration curves with straight lines. The average recoveries ranged from 69.4% to 99.2%, indicating that the method was very efficient and precise, since the recommended recoveries by the United States Environmental Protection Agency (USEPA, 1990) range from 50% to 120%.

Some PAHs were detected in the analyzed samples. Table 1.0 shows the detection of PAHs in certain beverage samples. There were no detections of acenaphlene, anthracene,benzo[a]anthracene, diben[a,h] anthracene in the beverages. The ∑16 PAHs concentration ($\times 10^{-2}$ µg/kg) obtained in the analysed beverages ranged from 36.25±0.01 in Nescafé to 90.04 ±0.01 in mychoco. Benzo[b]fluoranthene showed the highest abundance in mychoco, with a PAHs concentration of 49.67±0.01. Mean concentration levels ($\times 10^{-2}$ µg/kg) of the eight probable carcinogenic PAHs, ∑8 PAHs, ranged from 22.98±0.02 in Nescafé to 63.38±0.01 in Mychoco. Based on the results obtained, Mychoco has the highest concentration of PAHs, especially the most probable carcinogenic ones, while Nescafé has the lowest. This study is not in agreement with the study of Sadowska-Rociek et al. (2015), but is partially consistent with the studies of Kumari et al. (2012) and Shariatifar et al. (2019)

Sadowska-Rociek et al. examined the PAHs content in white (n = 6), milk (n = 7) and dark (n = 7) chocolate samples and reported that 10% of the samples exceeded PAH4 levels stipulated by European Regulation 835/2011 (Sadowska-Rociek et al., 2015). Samples were mostly contaminated with light PAHs, with phenanthrene representing the most frequent compound (13.5–96.6 µg/kg). This is not in line with the present study, as benzo[b]fluoranthene recorded the highest PAHs concentration in Mychoco at 49.67 µg/kg.

Kumari et al. (2012) investigated the levels of PAHs in chocolate candy and reported

detecting 16 PAHs at concentrations ranging from 2.70 to 235.91 ng/g (mean, 67.62 ng/g). Their results showed that BaP concentrations ranged from 0.35 to 12.76 ng/g (mean, 1.62 ng/g) and that 8% of candy samples exceeded the 5 µg/kg limit stipulated by European Union regulation for products containing cocoa butter (Kumari et al., 2012). But in this study, not all 16 PAHs were detected in the analyzed samples; the total mean PAHs concentration ranged from 36.25 to 90.04 µg/kg. BaP was detected only in Mychoco at a concentration of 13.71 µg/kg. But the present study showed BaP concentration exceeding 5 µg/kg. The present study does not fully agree with Kumari et al. (2012).

Shariatifar et al. (2019) evaluated PAHs in milk and milk powder samples obtained in Iran. They found that powdered milk had the highest mean level of 16 PAHs (2.28 ± 0.39 µg/kg), with some samples exceeding the limit of the European Commission (EU, 2011). Meanwhile, pasteurised milk showed the lowest PAHs levels (0.87 ± 0.18 µg/kg). Importantly, the authors noted a clear seasonal influence on these compound levels, with samples collected in winter showing the highest PAH levels. This finding highlights the impact of biomass-burning pollution on milk contamination. In addition, some PAH concentrations detected in this study exceeded the European Commission's limit.

Table 2.0 showed the detection of PAHs in roasted foods, plantains, maize, and yam. The ∑16 PAHs concentration (× 10^{-2} µg/kg) obtained in analyzed roasted foods ranged from 1.500±1.873 in roasted yam to 9.074±8.120 in plantain. Acenaphthene was recorded in the highest abundance in maize, with a PAH concentration of 8.046±0.00. The mean concentration levels (×10^{-2} µg/kg) of the eight probable carcinogenic PAHs, ∑8 PAHs ranged from 0.028±0.00 in roasted yam to 3.090±4.124 in roasted maize. The total mean concentration of ∑LMW PAHs ranged from 2.824±1.892 in yam to 9.759±7.861 in maize, while that of ∑HMW PAHs varied from 0.028±0.00 in yam to 6.090±4.142 in maize. Based on the results obtained, maize has the highest PAH concentrations, while yams have the lowest. This study agreed with the study of Eze et al. (2019) and Ciecierska (2020) but disagreed with the study of Amos-Tautua et al. (2013)

Eze et al. (2019) measured PAHs concentrations in roasted yams, ripe plantains, and corn. Six out of sixteen PAHs, acenaphthene, fluorene, phenanthrene, anthracene, fluoranthene and benzo[a]pyrene, were not detected in any sample. The highest mean concentration was pyrene at 3.872 mg/kg in corn, which contributed the most to total PAHs, followed by chrysene at 2.716 mg / kg in the same sample; acenaphthylene had the lowest value at 0.0057 mg / kg. The total PAHs were highest in corn (6.8073 mg/kg) and lowest in the yam (0.0158 mg / kg). In general, PAHs concentrations were highest in maize and lowest in yams, and some PAHs, such as anthracene, were consistently undetected.

Ciecierska (2020) examined the levels of 19 PAHs in several varieties of cocoa beans (Forastero, Trinitario, Criollo, and Nacional) from different countries as well as in their processed products. Ciecierska observed the highest levels of PAHs in roasted cocoa beans, cocoa mass, and cocoa butter (16.69–74.15 µg/kg of fat). In addition, the samples were predominantly contaminated with light PAHs. All raw cocoa beans exhibited relatively low contamination by heavy PAHs, with combined levels considerably lower than those permitted under the European Commission Regulation 835/2011 (EU 2011). The author concluded that the origin of these beans was associated with low levels of environmental pollution and that sun

drying was performed safely with respect to PAH contamination. This is in compliance with the present study, which showed predominant contamination of light or low molecular weight, ∑LMW-PAHs (2.824±1.892 to 9.759±7.861) in the roasted plantain, maize, yam and low contamination level of heavy or high molecular weight ∑HMW-PAHs (0.028±0.00 to 6.090±4.142).

Amos Tautau et al. (2013) evaluated PAHs and heavy metals in roasted food snacks. They reported no detection of PAHs in roasted plantains and other food items. However, the present study detected 11 out of 16 PAHs concentrations, ∑ with 16PAH = 9.074±8.120 in the analysed roasted plantain.

4.0. Conclusions

The PAHs detected in analyzed samples were much less than permissible limit and are considered safe for consumption. This can serve as a useful baseline for continuous monitoring of PAHs in Nigerian grains to protect human health in the country.

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