

Original Research Article

RISK ASSESSMENT OF POLYCYCLIC AROMATIC HYDROCARBONS IN FOREIGN AND LOCAL RICE CONSUMED IN SOUTH EAST NIGERIA

Abstract

Polycyclic aromatic hydrocarbons are group of organic compounds known for their carcinogenic and genotoxic nature and generated from incomplete burning of organic materials. Rice is one of the staple foods commonly consumed by both genders of all ages in Nigeria. Both local and foreign rice can be obtained from every nook and cranny of the country. The study aimed at determining the quantity of polycyclic aromatic hydrocarbons in foreign and local rice consumed in South East Nigeria; estimating the daily intake amount and the health risks associated with the consumption among adult male and female individuals in South East Nigeria. The samples were analyzed of sixteen PAHs contamination levels using gas chromatography coupled flame ionization detector. The result showed that the $\sum 16$ PAHs concentrations in the foreign and local rice ranged from $(24.103 \text{ to } 26.933) \times 10^{-2} \mu\text{g/kg}$ and $(22.953 \text{ to } 32.662) \times 10^{-2} \mu\text{g/kg}$ respectively. The concentrations of $\sum 8$ carcinogenic PAHs in the foreign and local rice ranged from $(9.728 \text{ to } 12.398) \times 10^{-2} \mu\text{g/kg}$ and $(11.262 \text{ to } 11.717) \times 10^{-2} \mu\text{g/kg}$ respectively. The mean concentration levels ($\times 10^{-2} \mu\text{g/kg}$) of lower molecular weight, LMW PAHs ranged from 9.382 ± 2.474 in Indian rice to 12.448 ± 5.883 in Thailand while that of higher molecular weight, HMW PAHs ranged from 14.238 ± 6.945 in Thailand to 16.377 ± 6.316 in Royal. The dietary intakes of the 16 PAHs were estimated, using the suitable indicators: BaP, PAH2, PAH4, PAH8. The total dietary exposure of adult male individuals ($\times 10^{-2} \mu\text{g/kg bw/day}$) for all the indicators was 31.94 in foreign and 33.86 in local rice, and for adult female 37.24 in foreign and 39.52 in local rice. This showed that both individuals are more exposed by consuming local rice than foreign rice. It also indicated that adult female individuals have higher intakes and are therefore more exposed to health risk than adult male individuals. Margin of exposures, MOEs, was used to assess the health risk of dietary exposure. This was calculated using the expected daily intake, EDI and benchmark dose level, BMDL10 for BaP, PAH2, PAH4 and PAH8. The values obtained were much higher than 10,000 indicating a low concern for consumer health at the mean estimated dietary exposures. The PAHs values detected were all below $1.0 \mu\text{g/kg}$ which is the permissible limit established by EFSA.

Keywords: Polycyclic aromatic hydrocarbons, Margin of exposure, estimated daily intake, health risk, rice.

1.0 Introduction

Polycyclic aromatic hydrocarbons, PAHs are set of organic compounds comprising of two or more fused benzene rings. They are formed as a result of incomplete burning of organic matter and some chemical processes. PAHs have been reported to cause carcinogenic and mutagenic

effects and are potent immune suppressants. They can interfere with the normal function of DNA (1). PAHs containing up to four rings are referred to as light PAHs and are called low-molecular weight (LMW) polycyclic aromatic hydrocarbons, LMW-PAHs. They include naphthalene, acenaphthylene, acenaphthene, fluorine, phenanthrene and anthracene. While those that contain more than four rings are heavy PAHs and are referred to as high-molecular weight, (HMW) polycyclic aromatic hydrocarbons, HMW-PAHs and they include fluoranthene, pyrene, benzo[b]fluoranthene, benzo[k]fluoranthene, benzo[a]pyrene, indeno[1,2,3-cd]pyrene, dibenzo[a,h]anthracene, benzo[g,h,i]perylene. Heavy PAHs are more stable and more toxic than the light PAHs (2). The HMW PAHs are even more detrimental to the environment and human health. The two primary factors which contribute to the persistence of HMW PAHs in the environment are PAHs molecule stability and hydrophobicity. Human beings may be exposed to these substances at home, outside or at workplace through inhalation, ingestion or dermal contact. Oncogenic, teratogenic effects, genotoxicity, an increased level of cholesterol in the blood or reproduction defects, biochemical disruption and cell damage were observed after longterm PAHs exposure and confirmed by toxicological experiments.(3) Using margin of exposure (MOE), toxic equivalency factor (TEF), benzo [a]pyrene (BaP) and several others have been proposed as markers for the risk assessment of PAHs present in foods (4 - 5). It has been suggested that MOE approach, using PAH4 (BaA, BaP, BbF&Chy) and PAH8 (BaA, Chy, BaP, BbF, BkF, DahA, BghiP&IndP), is better for the risk assessment, since BaP alone is not a suitable indicator of occurrence and effects of PAHs in foods (4).

Analysis of some foodstuffs such as rice (6 - 7) beans, wheat, maize (6,8,9), vegetable,(10 - 11) edible oil (12), fish, meat (13), pasta products (14), oil bean (15), soya bean oil (16), friuts (17) have revealed their PAHs contaminations. Studies have also reported the presence of PAHs in soil (18-19), sediments (20-21), water (22).The occurrence of the PAHs in food is due to environmental and soil contamination, manufacturing and cooking processes. Processing procedures, such as smoking and drying, and cooking of food is commonly thought to be the major source of contamination of dietary components by PAHs.

Rice is the most widely consumed staple food for a large number of human population in Nigeria. It is high in complex carbohydrates, has no fat, a good source of vitamins and minerals, contains eight essential amino acids. Foreign rice are those brands imported from the outside Nigeria while Local rice are referred to those cultivated and processed within Nigeria. Both brands are obtained from every nook and cranny of Nigeria including major and minor markets, shops, companies.

PAHs are detrimental to public health due to their carcinogenic properties and bioaccumulation and this have been a worldwide concern (23).The United State Environmental Protection Agency, USEPA lists sixteen PAHs as priority pollutants present in the air, water and soil (24). This study therefore proposed to determine the levels of PAHs in both foreign and local rice grains from markets in Enugu State Nigeria and assess the risk involved in consuming them.

2.0 Materials and Methods

2.1 Equipment and Reagents

Gas chromatography/flame ionization detector (HP 6890 Powered with HP ChemStation) , rotary evaporator, borosilicate beaker, glass column, sonicator.

All reagents and solvents were of analytical grade and were purchased from Sigma Aldrich U S A. These included hexane, dichloromethane, activated alumina as well as four deuterated (surrogate) standard namely acenaphthalene d₁₀, chrysene d₁₂, phenanthrene d₁₀ and perylened₁₂.

The analysis was carried out in Multi Environmental Management Consultants Ltd, Plot 4/5 LaaraSownmade Rd, off Igbe Rd, IjedaIkorodu, Lagos, Nigeria.

2.2 Sampling

Eighteen (18) samples which included different types of rice, foreign rice (Indian rice, Royal Stallion, Thailand rice): local rice (Abakaliki, Lafia ,Adani.) were purchased from some major markets in Enugu and Anambra states of Nigeria. The markets included New market, Gariki market and Ogbete main market in Enugu East, Enugu South and Enugu North Local Government Areas of Enugu State respectively, Nsukka main market in Igboetiti Local Government Area, Awka central market in Awka South Local Government Area of Anambra State, Umunze main market in Orumba South L. G. A. of Anambra State. The samples were picked to remove sand and other impurities, ground and put in labeled amber sample bottles ready for extraction.

2.3 Extraction of Samples:

Recovery experiments to optimize PAH extraction from grain samples were carried out. Three mixed standard solutions of concentrations 100, 500 and 1000 µg/mL were prepared using four deuterated PAHs (d-PAHs). These were used to spike three 5 g portions of ground grain samples which were extracted by sonication using 3:1 dichloromethane-hexane mixture as solvent. The extracts were cleaned-up in an alumina column using the same solvent mixture (25).

2.4 Determination of PAHs

PAHs concentrations were determined with a gas chromatography equipped with flame ionization detector, GC-FID, (HP 6890). The initial oven temperature programme was 60 °C for 5 min, raised to 250 °C, first at a rate of 15 °C/min and maintained at this temperature for 14 min and at second rate of 10 °C/min for 5 min. Nitrogen gas was used as the carrier gas at the flow rate of 1 cm³/min at a pressure of 30 psi. Recovery experiment was first carried out. Following recoveries of 94.0 to 99.2%, the grain samples were extracted and PAHs determined by the same procedure (25).

2.5 Statistical Analysis

Analysis of variance and Pearson Correlation Coefficient at 95% confidence level were carried out using SPSS version 16.00 on the data obtained.

2.6 Daily Estimated Intake

The dietary intakes of the 16 PAHs were estimated, using a deterministic approach. A fixed value for the consumption of an individual food was multiplied by a fixed value for the contaminant concentration in that food. (26) . The total exposure was obtained by summing the intakes from all foods, using the following equation:

$$\text{Estimated daily Intake (EDI)} = \frac{\sum \text{Consumption rate} \times \text{Occurrence}}{\text{Body Weight}}$$

Body weight for adult female = 60 Kg and body weight for adult male = 70 Kg were assumed (27)

2.7 Risk Assessment

For risk assessment, the margin of exposure approach (MOE) as adopted by the EFSA Scientific Committee in the Opinion related to substances which are both genotoxic and carcinogenic (28). The uncertainty in the assessment objectives is considered to be negligible. Margins of exposures (MOEs) were calculated by dividing the lowest BMDL₁₀ values among the models with acceptable fits by the mean and high level estimates of dietary exposure to benzo[a]pyrene, PAH₂, PAH₄ and PAH₈. However, for high level consumers the MOEs are close to or less than 10,000, which as proposed by the EFSA Scientific Committee (28) indicates a potential concern for consumer health and a possible need for risk management action. But for lower level consumers, the MOEs of 10,000 or higher would be of low concern for human health and might be considered low priority for risk management actions. (29)

The risk was estimated using the Margin of Exposure (MOE) approach according to the following equation (30):

$$\text{MOE} = \frac{\text{BMDL}_{10}}{\text{EDI}}$$

Where BMDL₁₀ is the benchmark dose lower confidence limit at 10% incidence level. Considering a BMDL₁₀ of 0.07, 0.17, 0.34 and 0.49 all in mg/kg bw per day for BaP, PAH₂, PAH₄ and PAH₈, respectively, for adult and children scenario, where:

BaP= Benzo[a] pyrene

PAH₂ = Benzo [a]pyrene and chrysene

PAH₄ = Benzo [a]anthracene, benzo[a] pyrene, benzo [b] fluoranthene and chrysene

PAH₈ = The sum of eight carcinogenic PAHs: benzo [a] anthracene; benzo [b]fluoranthene; benzo [k] fluoranthene; benzo[g,h,i]perylene; benzo [a]pyrene; chrysene; dibenz[a,h]anthracene; and indeno[1,2,3-C,d] pyrene. (4)

3.0 RESULT AND DISCUSSION

The results of the PAHs concentration levels of the Foreign and Local rice were shown in the tables 1 and 2 respectively. The sixteen PAHs were detected in all the analyzed rice samples. From Table 1, the $\sum 16$ PAHs concentration levels ($\times 10^{-2}$ $\mu\text{g}/\text{kg}$) detected in the Foreign rice studied ranged from 24.103 \pm 6.100 in Indian rice to 26.933 \pm 12.478 in Royal. The mean concentration levels of

eight probable carcinogenic PAHs, $\sum 8$ PAHs, varied from 9.728 ± 4.569 in Thailand to 12.398 ± 4.160 in Royal. The mean concentration levels of lower molecular weight, LMW PAHs ranged from 9.382 ± 2.474 in Indian rice to 12.448 ± 5.883 in Thailand while that of higher molecular weight, HMW PAHs ranged from 14.238 ± 6.945 in Thailand to 16.377 ± 6.316 in Royal. From Table 2, the $\sum 16$ PAHs concentration levels ($\times 10^{-2}$ $\mu\text{g}/\text{kg}$) detected in the Local rice analyzed varied from 22.935 ± 4.295 in Lafia rice to 32.662 ± 0.683 in Abakaliki rice. The mean concentration levels ($\times 10^{-2}$ $\mu\text{g}/\text{kg}$) of the eight probable carcinogenic PAHs, $\sum 8$ PAHs ranged from 11.262 ± 2.848 to 11.717 ± 0.846 in Lafia and Abakaliki rice respectively. While the mean concentration of LMW and HMW PAHs respectively varied from 7.339 ± 0.633 to 16.623 ± 2.911 and 15.614 ± 4.922 to 16.038 ± 2.228 in Lafia and Abakaliki rice. Local rice (Abakaliki rice) contained the highest level of PAHs. But the PAHs concentration levels detected in all the samples were very much lower $1.0 \mu\text{g}/\text{kg}$ which is the maximum allowed level set by EFSA for cereal grains and cereal based products.

The PAHs values detected in this study were very low compared to that reported by (18) and (7). (18) reported determination of 18 PAHs concentration levels in rice cooked by some common cooking methods in Ghana using GC- FID analysis. According to Essumang, the rice used as a control in the research showed various levels of PAHs, notably among them were pyrene (0.24 mg/kg), benzo[b]fluoranthene (0.66), dibenzo[a,h]anthracene (2.54 mg/kg), 2-methyl naphthalene (0.16 mg/kg). This was attributed to the increased cooking temperature. Also the study by Akan et al. (2018) on polycyclic aromatic hydrocarbons in different varieties of rice from Yobe state revealed detection of 16 PAHs also in low concentrations. The authors reported the predominancy of the lower molecular weight PAHs over the higher molecular weight PAHs in the studied rice samples, naphthalene, fluorine and pyrene having the highest values of 2.25×10^5 , 4.26×10^5 and 3.23×10^5 mg/kg respectively.

From the analysis of variance of all the analyzed grains, $p > 0.05$ indicating that there was no significant difference between the PAH concentrations of the grains. The Pearson correlation coefficient analysis indicated strong positive correlation among the PAH concentrations of the analyzed grains.

TABLE 1: MEAN CONCENTRATION ($\times 10^{-2}$ $\mu\text{g}/\text{kg}$) LEVELS OF PAHs IN DIFFERENT BRANDS OF FOREIGN RICE

PAHs	INDIAN RICE	ROYAL	THAILAND
	Mean±SD	Mean±SD	Mean±SD
Naphthalene	0.034±0.020	0.031±0.015	0.038±0.006
Acenaphthylene	0.054±0.019	0.051±0.001	0.140±0.147
Acenaphthene	2.61±1.909	2.916±2.718	1.905±2.200
Fluroene	0.261±0.285	0.322±0.380	0.197±0.253
Phenanthrene	1.778±2.119	2.150±1.059	5.574±3.881
Anthracene	4.649±2.437	5.086±4.140	4.594±0.603
Fluoranthene	1.890±1.568	2.117±1.972	1.221±0.997
Pyrene	1.634±0.109	1.862±0.184	3.287±3.375
Benzo[a]anthracene	3.648±1.203	2.956±0.917	3.001±3.082
Chrysene	2.508±2.748	3.660±4.211	1.512±1.844
Benzo[b]fluoranthene	0.605±0.533	0.713±0.651	0.465±0.025
Benzo[k]fluoranthene	0.576±0.345	0.666±0.436	0.446±0.071
Benzo[a]pyrene	3.266±2.090	3.746±2.384	4.031±3.197
Indeno[1,2,3-cd]pyrene	0.118±0.115	0.135±0.140	0.053±0.035
Dibenzo[a,h]anthracene	0.069±0.053	0.078±0.071	0.027±0.003
Benzo[g,h,i]perylene	0.406±0.160	0.444±0.117	0.195±0.125
∑16 PAHs	24.103±6.100	26.933±12.478	26.686±1.062
∑LMW PAHs	9.382±2.474	10.556±6.162	12.448±5.883
∑HMW PAHs	14.721±4.525	16.377±6.316	14.238±6.945
PAH2	5.773±0.657	7.406±1.828	5.542±1.353
PAH4	10.027±2.394	11.075±3.395	9.007±4.412
PAH8	11.197±3.067	12.398±4.160	9.728±4.569

SD= Standard Deviation

TABLE 2: MEAN CONCENTRATION ($\times 10^{-2}$ $\mu\text{g}/\text{kg}$) LEVELS OF PAHs IN DIFFERENCE BRANDS OF LOCAL RICE

PAHs	ABAKALIKI	LAFIA	ADANI
	Mean±SD	Mean±SD	Mean±SD
Naphthalene	0.019±0.003	0.048±0.015	0.040±0.001
Acenaphthylene	0.077±0.034	0.051±0.024	0.041±0.016
Acenaphthene	3.958±1.135	0.694±0.534	0.689±0.494
Fluroene	0.601±0.047	0.044±0.034	0.041±0.0262
Phenanthrene	5.642±6.046	2.957±0.396	3.419±0.586
Anthracene	6.327±2.079	3.546±0.370	3.163±0.199
Fluoranthene	2.813±0.881	0.688±0.126	0.755±0.151
Pyrene	1.508±0.501	3.573±2.333	3.848±2.720
Benzo[a]anthracene	2.168±1.583	3.974±2.562	3.945±2.063
Chrysene	5.230±1.732	0.413±0.161	0.456±0.150
Benzo[b]fluoranthene	0.932±0.299	0.368±0.096	0.349±0.107
Benzo[k]fluoranthene	0.783±0.250	0.447±0.005	0.429±0.025
Benzo[a]pyrene	1.845±0.038	5.767±0.283	5.851±0.383
Indeno[1,2,3-cd]pyrene	0.190±0.052	0.047±0.018	0.047±0.018
Dibenzo[a,h]anthracene	0.103±0.024	0.030±0.004	0.030±0.003
Benzo[g,h,i]perylene	0.465±0.110	0.306±0.033	0.289±0.005
∑16 PAHs	32.662±0.683	22.953±4.295	23.381±4.758
∑LMW PAHs	16.623±2.911	7.339±0.633	7.383±0.151
∑HMW PAHs	16.038±2.228	15.614±4.928	15.998±4.910
PAH2	7.076±1.694	6.180±0.122	6.308±0.171
PAH4	10.176±0.410	10.522±2.781	10.601±2.342
PAH8	11.717±0.846	11.262±2.848	11.396±2.340

SD= Standard Deviation

Table 3 and 4 reported the estimated daily intake of both foreign and local rice among adult male and female Nigerian. Table 3 presented the daily intakes of suitable indicators: BaP, PAH2, PAH4, PAH8 of the exposed adult male individuals who consume both foreign and local rice. It was reported thus that the total dietary exposure of adult male ($\times 10^{-2}$ $\mu\text{g}/\text{kg}$ bw/day) for BaP in foreign and local rice were respectively 3.77 and 4.63; for PAH2, 6.42 and 6.72 in foreign and local rice respectively; for PAH4, 10.33 and 10.72 respectively in foreign and local rice and PAH8 contributing 11.42 in foreign rice and 11.79 in local rice. All the indicators dominated in the local rice, adult male individuals are more exposed by consuming local rice than foreign rice.

Table 4 presented the daily intakes of suitable indicators: BaP, PAH2, PAH4, PAH8 of the exposed adult female individuals who consume both foreign and local rice. The total dietary

exposure of adult female ($\times 10^{-2}$ $\mu\text{g}/\text{kg}$ bw/day) for BaP in foreign and local rice were 4.4 and 5.4 respectively: for PAH2, PAH4 and PAH8 were 7.48 and 7.84, 12.04 and 12.52, 13.32 and 13.76 all respectively in foreign and local rice. Here also, all the indicators dominated in the local rice brands indicating that adult female individuals are also more exposed by consuming local rice than foreign rice.

Comparing tables 3 and 4, adult female individuals have higher intakes, in other words they are more exposed to health risk when compared to adult male individuals. The total dietary exposure of male (65.80 $\mu\text{g}/\text{kg}$ bw/day) was less than that of female (76.76 $\mu\text{g}/\text{kg}$ bw/day). This result varied from the study of Wu et al. (2016) which reported that the dietary exposure of male (9064ng/day) in Nanjing China was more than that of female (8308ng/day) .

TABLE 3: ESTIMATED DAILY INTAKES ($\times 10^{-2}$ $\mu\text{g}/\text{kg}$ bw/day OF ADULT MALE INDIVIDUALS ON FOREIGN AND LOCAL RICE FROM MARKETS IN NIGERIA

FOREIGN RICE	BaP	PAH2	PAH4	PAH8
Indian	1.13	1.99	3.43	3.84
Royal	1.27	2.54	3.81	4.25
Thailand	1.37	1.89	3.09	3.33
LOCAL RICE				
Abakaliki	0.62	2.43	3.49	4.01
Lafia	1.99	2.13	3.6	3.87
Adani	2.02	2.16	3.63	3.91

*Body weight (BW) of Male Individual = 70kg

**TABLE FOUR:
ESTIMATED DAILY INTAKES ($\times 10^{-2}$ $\mu\text{g}/\text{kg}$ bw/day OF ADULT FEMALE INDIVIDUALS ON FOREIGN RICE FROM MARKETS IN NIGERIA**

FOREIGN RICE	BaP	PAH2	PAH4	PAH8
Indian	1.32	2.32	4	4.48
Royal	1.48	2.96	4.44	4.96
Thailand	1.6	2.2	3.6	3.88

LOCAL RICE				
Abakaliki	0.72	2.84	4.08	4.68
Lafia	2.32	2.48	4.2	4.52
Adani	2.36	2.52	4.24	4.56

*Body weight (BW) of Female Individual = 60kg

The values of margin of exposures, MOEs obtained for all the indicators were much higher than 10000 which according to EFSA indicate low concern for human health and considered low priority for risk management actions. The MOEs values of the indicators in this study varied between 11290 and 127604 for males and 29661 and 126288 for females. This compare well with the studies by Ihedioha et al. (2018), Lee et al. (2019). The study by Ihedioha et al. on Risk assessment of polycyclic aromatic hydrocarbons in pasta products consumed in Nigeria revealed that the MOEs values for adult consumers were far higher than 10,000 indicating that the daily intake of PAHs was of low concern. Also the study by Lee et al. on the Occurrence and Risk characterization of PAHs of edible oils reported the values of MOEs (between 66094 and 1729776) were over 1.0×10^4 indicating that the risk of 4 PAHs in edible oils were of low concern from a public health point of view

TABLE 5: MARGIN OF EXPOSURE (MOE) FOR MALE EXPOSED INDIVIDUALS ON FOREIGN AND LOCAL RICE

FOREIGN RICE	BaP	PAH2	PAH4	PAH8
Indian	61947	85427	99125	127604
Royal	55118	66929	89239	11529
Thailand	51095	89947	11003	14715
LOCAL RICE				
Abakalilki	11290	69958	97421	12219
Lafia	35175	79812	94444	12662

TABLE 6: MARGIN OF EXPOSURE (MOE) FOR FEMALE EXPOSED INDIVIDUALS ON FOREIGN AND LOCA RICE

FOREIGN RICE	BaP	PAH2	PAH4	PAH8
Indian	53030	73275	85000	109375
Royal	47297	57432	76576	98790
Thailand	43750	77272	94444	126288
LOCAL RICE				
Abakalilki	97222	59859	83333	104701
Lafia	30172	68548	80952	108407
Adani	29661	67460	73113	107456

4.0 Conclusion

The samples were contaminated by the 16 PAHs but the contamination level was much lower than the permissible limit 1.0 µg/kg established by EFSA. Long time accumulation of these PAHs in the body can be hazardous to human health. From the estimated daily intake, total dietary exposure of male was less than that of female indicating that female daily intake of rice is higher. It can be concluded that female individuals are more exposed to PAHs than the male.

The values of margin of exposures, MOEs obtained for all the indicators were much higher than 10000 which according to EFSA indicate low concern for human health and considered low priority for risk management actions. The safety of consuming both local and foreign rice obtained from market in South East Nigeria can be ascertained. The result of this study provided

base values for future monitoring of contamination levels of rice grains. I am recommending that the environmental substances like foods, soil, water and air should be on regular analysis to ensure their safety with respect to PAHs.

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