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Polycyclic Aromatic Hydrocarbons Kolanuts (*Cola nitida* Schott & Amp; Endl) Daily Intake Exposure Risk from Côte d’Ivoire, West Africa

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Authors’ contributions

This work was carried out in collaboration among all authors. Author KKR designed the study, wrote the protocol, fitted the data and wrote the first draft of the manuscript. Author NY checked the first draft of the manuscript and achieved the submitted manuscript. Authors BGH, AA, KY and AC performed the statistical analysis, managed the literature and assisted the experiments implementation. Author BGH expertized the results interpretations. All authors read and approved the final manuscript.

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ABSTRACT

Polycyclic aromatic hydrocarbons (PAHs) are pervasive environmental pollutants of high toxicity. Due to their lipophilic characteristics, they tend to accumulate in the food chain. Human exposure to PAHs seems inevitable and the main route of exposure is food. The presence of PAHs in kolanut could cause serious health problem for consumers and slow down the export to new markets, which would constitute a significant shortfall for all actors in the kola sector.This study aimed to

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detrmined the Polycyclic Aromatic Hydrocarbons (PAHs) levels in kolanuts and estimate the exposure risks of nuts consumption by Ivorian population. Samples were collected from farmers, rural collectors, urban stores in districts (Mountains, Comoe, Lagoons, Down-Sassandra) and big storage centers of Anyama and Bouake cities. Concentrations of 9 PAHs (B[a]P, B[b]F, B[a]A, CHR, FLA, B[k]F, D[ah]A, B[ghi]P and IcdP) were measured using an Adept brand High Performance Liquid Chromatograph (HPLC) equipped with an ultraviolet (UV) / visible CE 4200 (CECIL) detector. Data showed the average concentration of PAHs in kolanuts at 1.22 ± 0.86 µg/kg. The PAHs concentrations expressed in BaP equivalent (BaPeq) ranging from 0.0009 ± 0.0003 µg/kg-BaPeq to 0.88 ± 0.24 µg/kg-BaPeq for B[ghi] P and D[ah]A. Based on the concentrations and the daily consumption of kolanuts estimated at 0.6 g/person in Côte d'Ivoire, the intakes values estimated of PAHs vary from 6.10^{-5} µg-BaPeq /day to $6.48.10^{-4}$ µg-BaPeq/day with an average of $3.06.10^{-4}$ ± $6.6.10^{-5}$ µg-BaPeq/day. The exposure daily doses (EDD) are all lower than the toxicity reference values (5 ng-BaPeq/kg BW/d). Thus, the occurrence of a toxic effect from PAHs after kolanuts consumption is very unlikely since the hazard quotient (HQ) are all less than 1. The risk of developing cancer is less than one case per 1,000,000 people.

Keywords: Cola nitida; PAHs; HPLC; consumption; health risk; Ivorian.

1. INTRODUCTION

Polycyclic aromatic hydrocarbons (PAHs) constitute a large class of chemical compounds containing two or more fused aromatic rings constituted of carbon and hydrogen atoms [1,2]. They are pervasive environmental pollutants of high toxicity [3,4]. HAPs are formed and released during the incomplete combustion or pyrolysis of organic matter, during industrial processes and other human activities [5,6,7]. Indeed, PAHs can also be derived from natural sources such as forest fires and volcanic eruptions. However, the majority of PAHs found in the environment are from anthropogenic sources such as emissions from motor vehicular exhaust; burning of coal, wood and wastes; and industrial sources such as thermoelectric power generation, cooking operations, production and combustion processes. [8,4]. Due to the lipophilic characteristics of PAHs, they tend to accumulate in the food chain [9]. Thus, several studies have revealed the presence of PAHs in certain foods such as fresh and smoked fish [10], smoked meat Products [11], Kolanut [6,7], raw cocoa [12,13], tubers [2], vegetable oils [14], coffee [15], honey [5], seafood, tea, rice, tomato, potato, fruit and infant foods [3]. Human exposure to PAHs seems inevitable and the main route of exposure is food [1,3]. PAHs are known as strict and toxic contaminants for living beings. Indeed, they are particularly studied because of their carcinogenic, teratogenic and mutagenic properties [16,17]. For example, sixteen [16] PAHs were listed as priority pollutants by the United States Environmental Protection Agency (USEPA) [4]. However, available experimental data have shown that certain PAHs have hematological, immunological and osteoarticular effects [18,19]. The toxicity of all well-known PAHs was previously shown [20]. However, the EFSA panel found four major PAHs named benzo[a]anthracene (BaA), chrysene (Chr), benzo[b]fluoranthene (BbF), and benzo[a]pyrene (BaP) are considered as PAH markers in foodstuffs [21,22].

According to Aikpokpodion *et al*. [6], PAHs have been detected in kolanut. Yet, kolanut plays a vital and leading role in the economy of Côte d'Ivoire as well as many African households and public authorities, which is reported to be the world leader of kolanut production. For example, Ivorian production reaches about 260,000 tons of fresh kolanuts per year [23,7]. About 90% of this production is consumed daily by the population and this, in many sociocultural rituals such as weddings, baptisms, expressions of friendship, funerals and rituals of sacrifice [24,25]. The importance of kolanuts is relative to its chemical composition. Indeed, it is a source of bioactive and functional compounds such as polyphenols and caffeine which reflects an increasingly growing interest for industries [26,27]. Then, nowadays, one of the serious problems in terms of safety of kalonut is the presence of PAHs [6,7]. Indeed, technological post-harvest treatments, storage condition and pollution caused by combustion engines present disadvantages for the kolanuts quality [6]. The consumption of kola could cause a serious health problem for consumers if the toxicity due to PAHs were proven.

Toxicity Equivalent Factor (TEF) method of risk assessment is used to evaluate the toxicity and assess the risks associated with a given compound. TEF is an estimate of the relative toxicity of a chemical compound to a reference chemical [28]. For a mixture of PAHs, the reference chemical is Benzo(a)pyrene (B[a]P). BaP is chosen because the toxicity of the chemical is well characterized. The toxicity equivalent factor of each PAH is an estimate of the relative toxicity of the PAH compound compared to B[a]P [29,2].

The presence of PAHs could also slow down the export of kolanut to new markets, which would constitute a significant shortfall for all actors in the kola sector.

The aim of this study was to determine PAHs levels in kolanuts produced in Côte d'Ivoire in order to estimate the health risk for consumer.

2. MATERIALS AND METHODS

2.1 Investigation Site

The study was conducted in the main areas of kolanuts production, big storage and distribution centers in Côte d'Ivoire. The investigated regions are located between 2°30' and 8°30' of West longitude and between 4°30' and 10°30' of North latitude. Thus, the mountain district (pole 1), the districts of Comoe and lagoons (pole 2) and the district of Bas-Sassandra (pole 3) were selected as production areas while the cities of Anyama and Bouake represent the storage and distribution centers (Fig. 1).

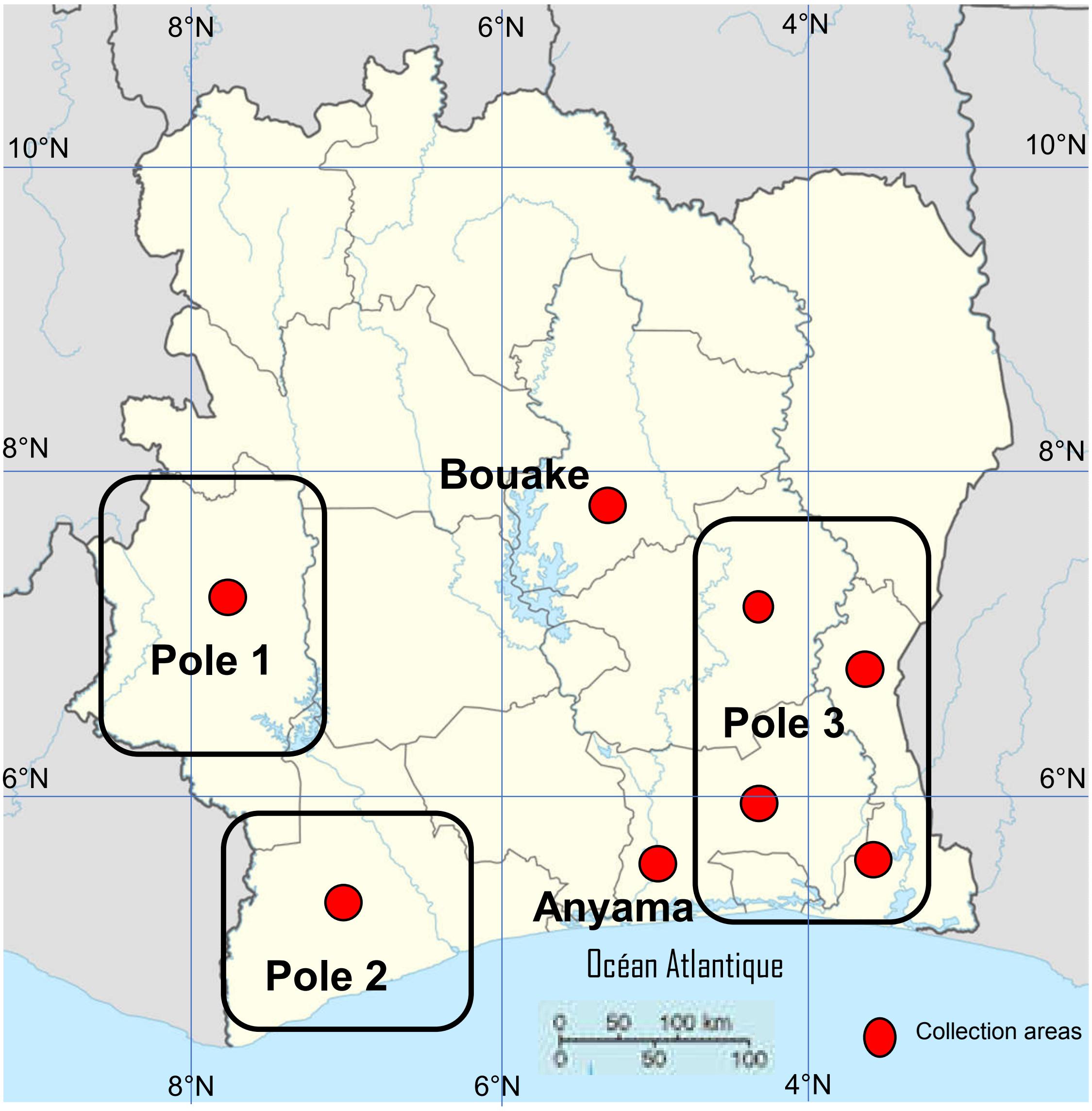

Fig. 1. Map showing kolanut samples collection sites

2.2 Plant Material and Sampling

The biological material of this study consists of fresh *Cola nitida* Vent. (Schott & Endl) nuts collected from farmers, rural collectors, urban stores and big storage centers, in accordance with the Regulation No 333/2007 of the European Commission [30]. So, 27 Samples were collected by storage centers (Anyama and Bouake cities) and by production pole namely 9 samples per type of actors. In total, 270 fresh kolanuts samples, weighing 2 kg each, were used for this study. Kolanuts was authenticated by N'Guessan botanist in the National Floristic Center (CNF) in Abidjan, Côte d'Ivoire, Training and Research Unit of Biosciences, Felix HOUPHOUËT-BOIGNY University where a voucher specimen was documented.

2.3 Mineralization and Analysis of PAHs

The analyzes of PAHs was conducted by using the method perfectly mastered and validated in accordance with the work of Kouadio *et al*. [7]. Kolanuts were treated according to the method described by Nyamien *et al*. [31].

2.4 Estimation of the Risk of Exposure to PAHs from Kolanuts Consumption

The risks considered in this study derived solely from the consumer exposure through ingestion of kolanuts contaminated with PAHs. The assessment methodology was conducted according to the model of Codex Alimentarius about risk assessment [32]. This procedure follows four main steps [33] including the hazard identification, the hazard characterization, the exposure assessment and risk characterization.

The concept of toxic equivalency factor (TEF) was used in this study. Health risk associated with the PAHs in kolanuts were evaluated using the toxicity equivalency factor (TEF) method [28]. TEF for each PAH was an estimate of the relative toxicity of the PAH compounds compared to B[aP]. The total equivalent concentration was expressed as BaP equivalent (BaPeq) BaPeq for individual PAH which was estimated using the equation 1.

$$BaPeq = \Sigma Cn \times TEFn \quad \text{(Eq 1)}$$

BaPeq is the BaP equivalent, Cn is the concentration of individual PAH sample, and TEFn is the toxic equivalency factor of the individual PAH in the sample matrix.

Assessment risk leads to the calculation of the Estimated Daily Intake (EDI) and the Exposure Daily Dose (EDD) from the average amount of 0.6 g per day of kolanuts consumed by an Ivorian adult [34,27]. The exposure scenarios where the individual is the most exposed have been used (maximalist assumption). EDI and EDD of PAHs linked to the consumption of kolanuts were determined from equations 2 and 3:

$$EDI = C \times Q \quad \text{(Eq 2)}$$

EDI is the estimated daily intake (µg BaPeq/d) ; C the Concentration of PAHs in kolanut (µg/kg BaPeq) and Q the Daily consumption of kolanut (kg/d).

$$EDD = C \times Q \times F/P \quad \text{(Eq 3)}$$

EDD is the exposure daily dose (µg BaPeq /kg/d) ; C the Concentration of PAHs in kolanut (µg/kg BaPeq) ; Q the Daily consumption kola nut (kg/d) ; F the Frequency of exposure (F = 1) and P the body weight of an Ivorian adult.

* The average body weight of an adult is conventionally equal to 70 kg according to the American Environmental Protection Agency (US EPA) [35].

The risk characterization for threshold effects was expressed by the hazard quotient (HQ). It was calculated for the oral route of exposure from equation 4:

$$HQ = EDD/TRV \quad \text{(Eq 4)}$$

HQ is the hazard quotient ; EDD the exposure daily dose (µg/kg/d) ; TRV the Toxicity Reference Value fixed by the Netherlands National Institute for Public Health and the Environment (RIVM: Rijksinstituut voor Volksgezondheid) [36].

If HQ <1, the occurrence of a toxic effect is very unlikely.

If HQ> 1, the appearance of a toxic effect cannot be excluded.

2.5 Statistical Analysis

Data has been captured with Excel Spreadsheet and were statistically treated using Statistical Program for Social Sciences (SPSS 20.0, SPSS for windows, USA) at 5% significance. The statistical test consisted in a one-way analysis of variance (ANOVA) with the origin of kola nuts. The statistical differences have been highlighted by the test of Duncan test at the 5% level of significance.

3. RESULTS

3.1 PAHs Markers Contents Extracted from Kolanuts Collected

The concentrations of B[a]P, B[a]A, B[b]F and CHR in kolanuts samples are presented in Table 1. The results indicate that only B[a]P concentrations are below the limit quantification (LOQ) for all samples analyzed. The concentrations of PAHs ranging from 0.14 ± 0.01 µg/kg to 3.05 ± 0.49 µg/kg, 0.01 ± 0.001 µg/kg to 3.63 ± 0.29 µg/kg and 0.01 ± 0.002 µg/kg to 2.31 ± 0.28 µg/kg for B[a]A, B[b]F and CHR, respectively. Statistical analysis revealed no significant difference ($p \geq 0.05$) between different B[a]A and CHR levels determined in the kolanuts whatever the origins unlike B[b]F ($p<0.05$) contents.

Table 2 presents the average values of Σ4PAHs (Σ4PAHs = sum of benzo(a)pyrene, benzo(a)anthracene, benzo(b)fluoranthene and chrysene) contents in kolanuts collected from actors.

In general, the concentrations of Σ4PAHs decrease while passing from wholesale stores, farmers, communal storage sites and rural hawkers. Thus, the highest content was found from the wholesale stores (1.87 ± 0.95 µg/kg); whereas the rural hawkers showed the lowest Σ4PAHs content (0,73 ± 0,31 µg/kg). The mean concentration of PAHs markers was 1.22 ± 0.86 µg/kg.

Based on the Regulation No 835/2011 of the European Commission [22] setting maximum levels for Σ4PAHs in foodstuffs for human consumption, the samples analyzed revealed lower levels than the maximum value set (10 µg/kg). Thus, the overall satisfaction of the sanitary quality of kolanut analyzed was 100%.

B[b]F dominance in kolanuts samples collected from farmers (0.33 µg/kg). As for collectors, urban stores and storage centers, B[a]A is the dominant PAH with contents of 0.36 µg/kg, 0.31 µg/kg and 0.82 µg/kg, respectively (Fig. 2).

3.2 Trends of 9 PAHs Concentrations Recommended for Risk Assessment in Kolanuts

The concentrations of the 9 PAHs recommended for risk assessment in kolanuts samples collected from actors are presented in Table 3. The PAHs contents expressed in BaP equivalent (BaPeq) ranging from 0.0009 ± 0.0003 µg/kg-BaPeq to 0.88 ± 0.24 µg/kg-BaPeq for B[ghi]P and D[ah]A, respectively. Statistical analysis revealed significant difference ($p<0,05$) between cumulative mean concentrations of 9 PAHs levels determined in the kolanuts whatever the actors.The highest content was found from the big storage centers (1.08 ± 0.27 µg/kg-BaPeq); whereas the communal storage sites showed the lowest PAHs cumulative content (0.10 ± 0.02 µg/kg-BaPeq). The mean concentration of Σ9PAHs was 0,51 ± 0,11 µg/kg-BaPeq.

3.3 Human Health Risk Assessment

The regularly exposed populations are those adults who daily consume kolanut. Table 4 presents the data of the model of quantitative evaluation of the risks related to the consumption of kolanut. The estimated daily intakes (EDI) were calculated for the levels of HAPs in kolanuts collected. The results showed that the level of PAHs evolves according to their concentrations in the matrix. Indeed, the estimated daily intakes of PAHs were ranged from 6.10^{-5} µg BaPeq/d to $6.48.10^{-4}$ µg BaPeq/d with a mean value of $3.06.10^{-4} \pm 6.6.10^{-5}$ µg BaPeq/d.

The exposure daily doses (EDD) are all lower than the Toxicity Reference Value (TRV) fixed by the Netherlands National Institute for Public Health and the Environment (RIVM: Rijksinstituut voor Volksgezondheid) at 5 ng BaPeq/kg b.w/d [36, 29]. In fact, mean EDD was $4.37.10^{-3} \pm 9.43.10^{-4}$ ng BaPeq/kg b.w/d. Therefore, the average risks of oral exposure to PAHs from the consumption of the kolanut are all less than 1 ($HQ = 8.74.10^{-4} < 1$).

According to the different exposure scenarios, the occurrence of a toxic effect from PAHs due to the kolanut consumption is very unlikely.

For an exposure to PAHs less than 5 ng BaPeq/kg b.w/d, the risk of cancer occurrence is one per 1,000,000 people. Whatever the EDD in our study, the risk of developing cancer is less than one case per 1,000,000 people (Table 5).

Table 1. Means concentrations of PAHs markers in kolanuts samples

Origin of the kola samples	TB[a]P (µg/kg)	B[a]A (µg/kg)	*Coefficient of variation*	B[b]F (µg/kg)	*Coefficient of variation*	CHR (µg/kg)	*Coefficient of variation*
F1	<LD	0.37 ± 0.05^{a}	13.51	0.49 ± 0.05^{bc}	10.20	0.34 ± 0.02^{a}	5.88
F2	<LD	1.41 ± 0.20^{a}	14.18	3.63 ± 0.29^{b}	7.99	1.82 ± 0.28^{a}	15.38
F3	<LD	0.16 ± 0.03^{a}	18.75	0.36 ± 0.03^{bc}	8.33	0.14 ± 0.01^{a}	7.14
C1	<LD	3.05 ± 0.49^{a}	16.06	1.03 ± 0.12^{bc}	11.65	0.01 ± 0.002^{a}	20
C2	<LD	0.48 ± 0.04^{a}	8.33	0.12 ± 0.02^{c}	16.67	0.24 ± 0.01^{a}	4.16
C3	<LD	0.14 ± 0.01^{a}	7.14	0.20 ± 0.02^{c}	10	0.13 ± 0.01^{a}	7.69
S1	<LD	0.37 ± 0.02^{a}	5.40	0.01 ± 0.001^{c}	10	0.11 ± 0.01^{a}	9.09
S2	<LD	0.26 ± 0.01^{a}	3.85	0.29 ± 0.05^{bc}	17.24	0.06 ± 0.01^{a}	16.67
S3	<LD	0.31 ± 0.03^{a}	9.68	0.16 ± 0.01^{c}	6.25	0.02 ± 0.004^{a}	20
Center 1	<LD	1.95 ± 0.2^{a}	10.26	0.77 ± 0.04^{a}	5.19	2.31 ± 0.28^{a}	12.12
Center 2	<LD	1.96 ± 0.10^{a}	5.10	<LD	<LD	0.98 ± 0.12^{a}	12.24

Means with the same letters exponentiating in the same column are not different at 5% according to Duncan test ; LD : Limits of detection; TB[a]P: Benzo(a)pyrene ; TB[a]A: Benzo(a)anthracene ; B[b]F: Benzo(b)fluoranthene ; CHR: Chrysene ; F1, C1, S1: Farmers, Collectors, Stores of mountain district; F2, C2, S2: Planters, Collectors, Stores of districts of Comoe and lagoons; F3, C3, S3: Planters, Collectors, Stores of district of Bas-Sassandra ; Center: big storage and distribution centers (1: Anyama ; 2: Bouake)

Table 2. Concentrations of PAHs markers in kolanuts according to the actors

Actors	Average (µg/kg)	Min - Max
Farmers	0.87 ± 0.51^{B}	[0.21 – 1.65]
Collectors	0.73 ± 0.31^{B}	[0.25 – 1.19]
Urban Stores	0.76 ± 0.36^{B}	[0.17 – 1.32]
Centers	1.87 ± 0.95^{A}	[0.27 – 4.61]
Average	1.22 ± 0.86	[0.17 – 4.61]
FML ($\mu g.kg^{-1}$)	10	
Sample (%) < FML	100	
Sample (%) ≥ FML	0	
Overall satisfaction	100	

FML: Fixed Maximum Level by the European Commission [22]; Min: minimum; Max: maximum; Means with the same letters exponentiating in the same column are not different at 5% according to Duncan test

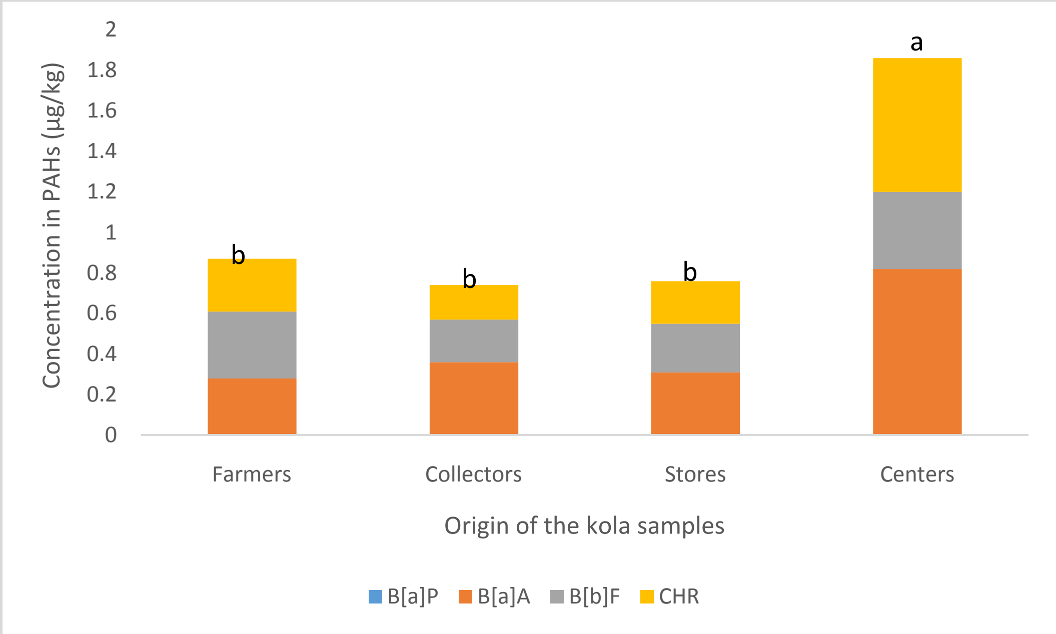

Fig. 2. Cumulative mean concentrations of PAHs markers from the kola nuts studied
values differ statistically at P=5% according to the lowercase letter

Table 3. Concentrations of 9 PAHs in kolanuts

PAHs	TEF (INERIS, 2003)	Farmers	Collectors	Stores	Centers
B[a]P	1	<LD	<LD	<LD	<LD
B[a]A	0.1	0.028 ± 0.003^{A}	0.036 ± 0.003^{A}	0.031 ± 0.002^{A}	0.082 ± 0.007^{A}
B[b]F	0.1	0.033 ± 0.002^{AB}	0.021 ± 0.002^{B}	0.024 ± 0.003^{B}	0.038 ± 0.005^{A}
CHR	0.01	0.0026 ± 0.0002^{AB}	0.0017 ± 0.0001^{B}	0.0021 ± 0.0003^{B}	0.0066 ± 0.0005^{A}
FLA	0.01	0.002 ± 0.0003^{A}	0.0039 ± 0.0004^{A}	0.0044 ± 0.0003^{A}	0.0056 ± 0.001^{A}
B[k]F	0.1	0.061 ± 0.01^{A}	0.027 ± 0.003^{A}	0.009 ± 0.002^{A}	0.063 ± 0.017^{A}
D[ah]A	1	0.36 ± 0.07^{A}	0.24 ± 0.04^{A}	0.03 ± 0.01^{A}	0.88 ± 0.24^{A}
B[ghi]P	0.01	0.0127 ± 0.0018^{A}	0.0052 ± 0.0006^{B}	0.0029 ± 0.0005^{B}	0.0009 ± 0.0003^{B}
IcdP	0.1	0.017 ± 0.002^{A}	0.03 ± 0.005^{A}	<LD	<LD
∑ 9 HAP		0.52 ± 0.09	0.36 ± 0.05	0.10 ± 0.02	1.08 ± 0.27
Average		0.51 ± 0.11			

Concentrations in µg/kg-BaPeq. Means with the same letters exponentiating in the same line are not different at 5% according to Duncan test ; LD : Limits of detection, TEF : Toxic Equivalency Factor

Table 4. Quantitative evaluation of the exposure of PAHs

Measured parameters	Min.	Avg.	Max.
Concentrations of PAHs (µg/kg-BaPeq)	0.10	0.51 ± 0.11	1.08
EDI (µg BaPeq/d)	6.10^{-5}	$3.06.10^{-4} \pm 6.6.10^{-5}$	$6.48.10^{-4}$
EDD (ng BaPeq/kgBW/d)	$8.57.10^{-4}$	$4.37.10^{-3} \pm 9.43.10^{-4}$	$9.26.10^{-3}$
TRV (ng BaPeq/kgBW/d)		5	
HQ = R	$1.71.10^{-4}$	$8.74.10^{-4}$	$1.85.10^{-3}$

Min. : Minimun ; Avg. : Average ; Max. : Maximum ; EDI : Estimated Daily Intake ; EDD : Exposure Daily Dose ; TRV : Toxicity Reference Value ; HQ : Hazard Quotient ; R : Risk ; BW : body weight d : day

Table 5. Risk characterization linked to the consumption of Kolanuts contaminated with PAHs

Exposure Daily Dose (ng BaPeq/Kg.BW/d)	SF (relative to the population)
$8.57.10^{-4}$	Less than 1 in 1,000,000 cases
$4.37.10^{-3} \pm 9.43.10^{-4}$	Less than 1 in 1,000,000 cases
$9.26.10^{-3}$	Less than 1 in 1,000,000 cases

SF : Slope Factor by oral route

4. DISCUSSION

The PAHs targeted in this study (B[a]P, B[b]F, B[a]A, CHR, FLA, B[k]F, D[ah]A, B[ghi]P and IcdP) are part of those recommended by the European Food Safety Authority (EFSA) for risk assessment [36]. Quantitative analysis of the different samples revealed the presence of these PAHs in the kolanut with variable levels according to the collected area and the type PAH. Furthermore, EFSA indicates that a system of four specific molecules (B[a]P, B[a]A, B[b]F, CHR) would be the most appropriate indicator of the presence of PAHs in food [37]. Results showed the average contents of PAHs markers lower than the accepted maximum limits fixed at 10 µg/kg [22]. PAHs concentrations of the kolanut samples analyzed comply with international specifications.The concentrations observed are slightly higher than those of work of Aikpokpodion *et al*. [6] on kolanuts collected from the markets of Oyo, Ogun and Osun in the area of Nigeria where the PAHs markers levels of 0.13 µg/kg, 0.21 µg/kg and 0.28 µg/kg were obtained, respectively. The difference levels observed could be due to the various conditions of post-harvest processing, storage and transportation of kolanuts [38]. Data obtained indicated that the average content of PAHs markers in the kolanut among storage centers is higher than the levels recorded from other actors (planters, collectors and stores). Indeed, the high content of PAHs in storage centers can be explained by the geographical position of these centers located in urban areas. The results from the present study were similar to those observed by Dobrinas *et al*. [5] who reported that the concentrations in PAHs of honey produced in urban areas are higher than those from rural areas. Anyama and Bouake concentrate most of the national manufacturing productive park with a high population density and an intense daily road traffic is subject to more pollution than rural zone. Thus, the concentrations of PAHs in kola nuts can be attributed to the high atmospheric PAH pollution [5,8,7]. In fact, the daily passage of gasoline engines or diesel engines of the various trucks in transit, combined with soot and smoke of all origins, from the exhaust gases of the combustion engines to the smoke of cigarette could justify this PAH content [39,40,4].

The results show that the contents of B[b]F and B[a]A are the most abundant individual PAH compounds in the kolanuts. According to Léotz [41]. Those PAHs come mainly from the operation of gasoline engines or diesel engines. In addition, on-farm, contamination by PAHs comes from agricultural incinerators [42]. According to N'Guessan *et al*. [23] the kolanuts are packaged and presented to the consumer with a precarious concern for hygiene. The whole of the bad practices of treatment of the kolanuts applied, to the store of storage is source of PAHs contamination. In addition, no kola nut processing and storage center, surveyed cities comply with the recommendations of the Prerequisite Program.

In rural areas, PAHs contamination comes from the smoke produced by the wood fire lit during the preparation of family meals, during the storage of kolanuts in peasant kitchens [43,44]. The presence of these PAHs in kolanuts is due to the migration of PAHs from smoke from wood fires. Indeed, several studies had previously described, the incomplete combustion processes of organic matter, especially wood, generates the formation of various types of PAHs [45].

PAHs are retained on the kolanut by water vapor [6]. The decrease in PAHs concentrations from planters, passing through rural collectors and urban would be due to the repeated washing of kolanuts during the transit or storage process.

Contamination levels of kolanut in 9 PAHs recommended for risk assessment were higher than those observed from fruits in France, where the mean value was 0.17 µg/kg-BaPeq [36]. According to Azza [46], PAH levels in uncooked food largely depends on the origin of the food and can be subjected to regional variation. In addition, the difference in PAH concentrations between food matrices could be due to their bioaccumulation capacity. Also, It can be due to production conditions such as the growing soil. The genesis of PAHs in the soil is the result of biological processes transforming various aromatic precursors into PAHs, under the action of enzymes derived from anaerobic bacteria [47].

Otherwise, estimated daily intakes (EDI) increase with the concentrations in PAHs present in the kolanut. This result corroborates that of Kouadio *et al*. [27] which stipulates that the exposure level of consumer increases with concentration of the heavy metals t in the kolanut. The exposure daily doses (EDD) obtained are all lower than the Toxicity Reference Value (TRV) fixed by the Netherlands Institute for Public Health and the Environment (RIVM) to 5 ng BaPeq/kg body weight [29]. This situation indicates that kolanuts represent a low health risk of development of adverse effects (cancer) for human and would be safe for people comsumption.

On the other hand, the regular consumption of a quantity of kolanuts leading to an EDD higher than the TRV would represent a danger for the consumer's health. Taking into account the bioaccumulative, carcinogenic and genetoxic properties of PAHs, the toxicological risk exists, and even chronic exposures to relatively low concentrations can generate cancer (chronic carcinogenic effect) as the molecules studied are molecules with no threshold effect [10].

Since kolanut is largely consumed fresh, it is therefore important to intensify efforts to reduce the presence of these PAHs mainly by raising awareness among actors on good practices for the conservation of kolanuts. These good practices pass by the storage of kolanuts outside of the smoke-producing sources.

5. CONCLUSION

This study revealed the presence of PAHs at varying levels in kolanuts. PAHs markers concentrations are generally lower than the maximum limits recommended by the the Regulation No 835/2011 of the European Commission. The main source of PAH in the investigated kola nuts originated from incomplete combustion processes. Indeed, the safety of kolanuts depends on factors such as the storage condition near smoke sources, pollution caused by combustion engines and bad condition of hygiene. Estimated daily doses in PAHs from kolanuts, always remains below the toxicological reference value. Consequently, kolanuts represent a low health risk of development of cancer and would be safe for the consumer. However, this satisfaction must not forget the bad practices of post-harvest processing of kolanuts. Thus, the implementation of efficient technical during conservation, and marketing will be able to guarantee better sanitary quality for kolanuts.

DISCLAIMER

The products used for this research are commonly and predominantly use products in our area of research and country. There is absolutely no conflict of interest between the authors and producers of the products because we do not intend to use these products as an avenue for any litigation but for the advancement of knowledge. Also, the research was not funded by the producing company rather it was funded by personal efforts of the authors.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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