International Journal of Applied Chemistry.
ISSN 0973-1792 Volume 19, Number 2 (2023) pp. 109-117

http://www.ripublication.com

Risk Assessment of Polycyclic Aromatic Hydrocarbons in Grilled Meat "Choukouya" and Beef Skins "Kplo" from Cooking with Rubberwood

Yaya C. [1]*, Soro D. B. [2], Hampoh A. H. [3], N'guettia K.R. [2], Aboua K. N.[2] , Diarra M.[4] , Meite L.[2], Traore K. S.[2], Dembele A.[5]

[1]UFR-Agriculture, Fisheries Resources and Agro-industry, University of SAN-PEDRO, 01 BP 1800 SAN-PEDRO 01, Côte d'Ivoire

[2]Laboratory of Environmental Sciences, UFR of Environmental Sciences and Management, NANGUI ABROGOUA University, 02 BP 801 ABIDJAN 02, Côte d'Ivoire

[3]Laboratory of Nutrition and Food Safety, UFR of Food Sciences and Technologies, NANGUI ABROGOUA University, 02 BP 801 ABIDJAN 02,Côte d'Ivoire

[4]Laboratory of Environmental Science and Technology, UFR Environment, Jean-Lorougnon-Guede University, BP 150 DALOA, Côte d'Ivoire

[5]UFR of Natural Sciences, NANGUI ABROGOUA University, 02 BP 801 ABIDJAN 02, Côte d'Ivoire

ABSTRACT

Grilled meat called "choukouya" and beef skins known as "kplo" are widely consumed in Côte d'Ivoire. However, in the District of Abidjan, their cooking is mainly done with resinous fuels, which are not very savory such as rubberwood, used tires and rubber debris. These fuels emit compounds toxic to human consumption, including polycyclic aromatic hydrocarbons (PAHs), carcinogenic or mutagenic. The objective of this study is to assess the dietary risks faced by consumers of meat and skins contaminated with PAHs. A food survey was first conducted. Subsequently, sixty (60) hide samples and 120 meat samples were collected and analyzed on an Agilent high performance liquid chromatography chain. The results showed that rubberwood and used tires are the most widely used fuels. The concentration of benzo(a)pyrene and the sum of the concentrations of the 4 indicator PAHs exceed their respective standards (2 µg/kg and 12 µg/kg). The daily exposure dose (DED) is 1.78 µgTEQ/kg bw/day. The Individual Excess Risk IERs, greater than 10^{-5}, indicate the occurrence of carcinogenic or mutagenic effects. In perspective, populations will be made aware of good culinary practices.

Keywords: meat; skins; PAHs; rubberwood; choukouya; kplo

1. INTRODUCTION

Animal products are an essential component of people's diets and livelihoods. In Côte d'Ivoire, particularly in the district of Abidjan, people appreciate grilled meat commonly known as "choukouya", beef skin called "kplo". Unfortunately, according to a field survey, unsavory fuels are used for cooking meat or pickling skins. Indeed, some craftsmen use rubber or recovered wood, available, of lower cost however resinous or with paint or varnish, for cooking meat in artisanal metal ovens. Other craftsmen use tires, plastic or rubber items, to strip animal skins. These pre-treatment and cooking processes are sources of toxic fumes. Indeed, these fumes generate toxic compounds for human consumption, including polycyclic aromatic hydrocarbons (PAHs), carcinogenic and mutagenic **[1,2]**.

Prolonged exposure to certain PAHs leads to an increased risk of developing cancer **[3]**. Similarly, it's shown that eating red meat and deli meats increases the risk of colorectal cancer. PAHs are also known to induce many other toxic effects such as hepatic **[4,5].**

In Côte d'Ivoire, there are between 15,000 and 20,000 new cases of cancer per year, 20% of which are related to food **[6]**. The preservation of foods, particularly meat and offal for efficient consumption, often requires smoking and cooking techniques that are important sources of PAHs.

The presence of these molecules in the environment, particularly in food, could pose a risk to human health. Therefore, the determination of the state of contamination of these foodstuffs becomes crucial in the preservation of the health of the populations. Thus, the general objective of this study is to assess the risks associated with the consumption of meat and skins contaminated with PAHs.

2. MATERIAL AND METHODS

2.1. Sampling

The study took place in three communes in the district of Abidjan, selected for their intense stripping and restoration activity. These are the communes of Abobo, Port-Bouët and Yopougon. Two pickling sites and six grilling sites were selected. One of the two stripping sites is located in the commune of Abobo and is marked by GPS coordinates 5°27'4"N and 3°58'29"W. The other is in the commune of Port-Bouët, with GPS coordinates 5°15'45"N and 3°58'11"W. Table 1 shows the GPS coordinates of the six grilling sites.

Table 1. GPS coordinates of the grilling sites

Port-Bouët	Yopougon
Site I :5°15'34''N and 3°58'5''W	Site IV : 5°21'18''N and 4°4'45''W
Site II :5°15'35''N and 3°58'9''W	Site V : 5°21'14''N and 4°4'27''W
Site III : 5°15'33''N and 3°58'8''W	Site VI : 5°20'47''N and 4°3'54''W

Sampling of Skins

Pickled beef pelts with flaming rubberwood were purchased at the site. Figure 1 shows beef skins being stripped with rubberwood. After stripping, they were cleaned and looked like Figure 2. After cooking and softening with potash, the samples were conditioned. Figure 3 shows samples of skins during packaging.

Figure 1: Beef skins during a pickling session

Figure 2: Appearance of skin after cleaning

Figure 3: Skin packaging

Meat Sampling

Fresh meat was purchased and divided into two parts. A first part was used to make the samples of fresh meat and a second part was grilled with rubberwood. After cooking and cooling, the grilled meat samples were packaged. Figures 4, 5 and 6 show fresh meat, grilled meat and the packaging of grilled meat samples, respectively.

Figure 4 : Fresh meat

Figure 5 : Grilled meat

Figure 6 : Packaging of meat samples

A total of sixty (60) hide samples were collected at the 2 pickling sites and 120 meat samples were collected at the 6 grilling sites for analysis.

2.2. Analysis of samples

The determination of PAHs was done according to ISO 15753:2016 which describes the method of determination of these molecules in fats of animal and vegetable origin. In the present study, the following 9 PAHs molecules were assayed: benzo(a)pyrene; benzo(a)anthracene; benzo(a,h)anthracene; benzo(b)fluoranthene; benzo(k)fluoranthene; benzo(g,h,i)perylene; chrysene; fluoranthene; indeno(1,2,3-c,d)pyrene.

The method of analysis of the samples is the chromatographic method **[7].**

2.3. Risk assessment methodology

The risk considered arises solely from the ingestion exposure of foods that are "kplo" skin, grilled meat "choukouya" and meat to cook. To assess risks, the FAO/WHO standardized methodology **[8]** was used. The Daily Exposure Dose (DED), the Hazard Ratios (HR) and the Individual Excess Risk (IER) were calculated using the following formulae:

DED=EDI/bw with EDI the estimated daily intake; bw the body weight;

HR=DED/TRV with TRV the Toxicological Reference Value;

IER =DED×(T/Tp)× EUR with EUR the Excess Unit Risk ; exposure time T=5 years; the weighting time Tp = 70 years and bw the body weight determined by the dietary survey.

3. RESULTS AND DISCUSSION

3.1. Contamination levels and exposure assessment

Among PAHs, chrysene has the highest average concentrations in skins in Abobo and Port-Bouët. In samples of grilled meat (choukouya) from Port-Bouët and Yopougon, chrysene and fluoranthene have the highest average concentrations. The average concentrations of benzo(a)pyrene and the sum of the average concentrations of the 4 indicator PAHs of the European Council are above the MRLs set at 2 µg/kg and 12 µg/kg respectively **[9].**

It should be noted that the 4 indicator PAHs are: benzo(a)pyrene; benzo(a)anthracene; benzo(b)fluoranthene and chrysene. The high concentration of chrysene could be explained on the one hand by its presence in the resin from rubberwood, fuels most frequently used for pickling skins and cooking meat. Indeed, some plants such as conifers can create, for example, phenanthrene or chrysene present in the resin **[10]**. Similarly, its formation during the combustion of rubber, the substance constituting the tires used for pickling, is also at the origin of its abundance **[11].**

The survey showed that the average daily consumption of hides (kplo), fresh meat and grilled meat (choukouya) is respectively 70 g, 20 g and 80 g in the commune of Port-Bouët. This commune, with the most complex ration consisting of the three matrices, was retained for the calculation of exposures. Also, the average body weight (bw) is 65 kg.

To calculate the EDI and the DED, the toxic equivalent factor (TEF) for each molecule was taken into account **[12]**. As the TEF of B(A)P is 1, it has been kept as a reference. The results obtained are shown in table 2.

TABLE 2. Estimated Daily Intake (EDI) and Daily Exposure Dose (DED)

Food matrix	**Average food consumption (g/pers/day)**	**EDI (µgTEQ/kg/day)**		**DED (µgTEQ/kg bw/day)**	
		B(A)P	**∑PAHs**	**B(A)P**	**∑PAHs**
Skins	70	5.643	16.754	0.087	0.258
Fresh meat	20	0.245	1.650	0.004	0.025
Grilled meat	80	3.777	97.305	0.058	1.497
Daily Exposure Dose DED=EDI/bw (µgTEQ/kg.bw/day)				0.149	1.780

Results indicate an overall exposure to PAHs of 1.78 (µgTEQ/kg bw/day) above the virtually safe dose (VSD) of 0.14 µg TEQ/kg bw/day **[13].**

It consists of exposure from the consumption of hides and skins (0.258 µgTEQ/kg bw/day), grilled meat (1.497 µgTEQ/kg bw/day) and fresh meat (0.025 µgTEQ/kg bw/day). It appears that grilled meat has the highest contribution with 84% followed by skins (15%) and fresh meat (1%).

3.2. Food risk characterisation

Case of threshold effects: hazard ratio (HR) for PAHs

For threshold effects, Table 3 presents the hazard ratios for B(A)P and the cumulation of the 4 PAHs.

Table 3. Hazard Ratio (HR) for PAHs

	B(A)P	**∑4HAPind**
Toxicological Reference Value TRV (µg/kg/day)	0.14	0.5
Daily Exposure Dose : DED	0.149	119.97
Hazard Ratios HR=DED/TRV	1.064	239.94

It appears that the hazard ratios are greater than 1 for benzo(a)pyrene and the cumulation of the 4 indicator PAHs. The occurrence of PAHs related side effects is possible. These effects can be abdominal pain, vomiting neurological disorders (headaches, hallucinations, convulsions). They can be hepatic, haematological or immunological **[14,15].**

Case of effects without threshold: individual excess risk (IER*)*

To characterize the risks due to chronic toxicity without threshold, the IERs were calculated taking into account the benzo(a)pyrene EURs and the cumulative 4 PAH indicators. For B(a)P the EURo established by RIVM is 2 (mg/kgbw/day) $^{-1}$. This value is derived from a virtually safe dose VSD of 50 ng/kgbw/day for an excess cancer risk of 10^{-5} **[16]**.

Applying the TEF to the cumulation of the 4 indicator PAHs, the corresponding Excess Unit Risk EUR is 2.574 mg/kg bw/day. Hence the calculated IER shown in Table 4.

Table 4. IER calculated due to B(a)P and cumulation of 4PAHs

PAHs	**Excess Unit Risk EUR (mg/kg bw/day)$^{-1}$**	**Daily Exposure Dose DED (mg/kg bw/day)**	**Individual Excess Risk IER**
B(a)P	2	0.000149	2.128×10^{-5}
$\sum PAH_{ind}$	2.574	0.11997	2.205×10^{-2}

The results show that for non-threshold effects, the calculated individual risk excesses (IER) are above the norm 10^{-5}. Therefore, carcinogenic or mutagenic effects have occurred.

The manifestation of these pathologies could be explained essentially by the structure and physico-chemical properties of PAH molecules which are the 4 indicator PAHs. Indeed, benzo(a)pyrene and benzo(b)fluoranthene molecules have 5 rings, thus classifying themselves among heavy PAHs with low water solubility (0.0038 mg/L and 0.0015 mg/L) **[17]** and a relatively high octanol/water partition coefficient (log Kow= 6.04 and log Kow= 5.8) **[18].**

They are therefore inclined to cross biological membranes which are lipid bilayers, and thus to accumulate in living organisms, thus describing the process of bioaccumulation.

The presence of these molecules in living organisms could promote exchanges with DNA. Indeed, the existence of "bay" regions in their structure is a determining parameter of carcinogenic potential**.** In addition, the benzo(a)anthracene and chrysene molecules are also lipophilic, with 4 rings also presenting "berry" regions.The regions (bay and fjord) are made up of angular structures formed by benzene rings and by their shape represent a steric congestion. The presence of one of these regions makes degradation of compounds possible by promoting oxidation reactions **[19].**

4. CONCLUSION

Consumers are exposed to PAHs. Both threshold effects (neurological disorders) and non-threshold effects (carcinogenic or mutagenic) related to PAHs have occurred. In perspective, it will be a question of conducting awareness campaigns on good culinary practices and on the use of rubberwood as timber for work and cabinetmaking.

ACKNOWLEDGMENT

Thanks are due to the National Laboratory for Agricultural Development Support for the analyses (LANADA). We express our gratitude to Nangui Abrogoua University for some of the material. We would also like to thank the artisans and consumers who were the subject of the food survey.

REFERENCES

[1] INERIS, Métaux-mercure. Rapport final. DRC-02-39266-AIRE /étude 10-782 fmr., 87p, 2002.

[2] Aliapur, Extrait du document de référence ALIAPUR R&D 2009 sur l'utilisation des pneus usagés comme combustible alternatif : valeurs de référence et protocoles de caractérisation. Aliapur – R&D – juillet 2009, 15p, 2009.

[3] Okona-Mensah K. B., Battershill J., Boobis A., Fielder R., An approach to investigating the importance of high potency polycyclic aromatic hydrocarbons (PAHs) in the induction of lung cancer by air pollution. Food and Chemical Toxicology, 43, pp. 1103-1116, 2005

[4] Bastide N., Fer héminique et cancérogénèse colorectale : étude des mécanismes et recherche de stratégies préventives. Thèse de Doctorat, Université de Toulouse, France, 198p, 2012.

[5] CSS,Viande rouge, charcuterie à base de viande rouge et prévention du cancer colorectal. Résumé. Avis du conseil supérieur de la santé numéro 8858, 4p, 2013.

[6] ALIAM/AILC, Alliance des ligues francophones africaines et méditerranéennes. Association Ivoirienne de Lutte Contre le Cancer. Interview intitulé « manger de la viande braisée est un risque de cancer», 6p, 2014.

[7] Yaya C., Soro D. B., Aboua K. N., Kouadio D. L., Diarra M., Ehouman A. G. S., Meite L., Traore K. S. and Dembele A., Levels of contamination of meat and offal (skins) by polycyclic aromatic hydrocarbons during grid cooking or following pre-treatment of tire stripping, Journal of Chemical, Biological and Physical Sciences, Section B, Vol. 9, No. 3; pp.372-379, 2019a.

[8] FAO/OMS, Organisation des Nations Unies pour l'alimentation et l'agriculture et l'Organisation mondiale de la Santé. Environmental Health Criteria; 240, Principles and methods for the risk assessment of chemicals in food. ISNB 978 9241572408. (http://whqlibdoc.who.int/ehc/WHO_EHC_240_6_eng_Chapter 3.pdf),2009.

[9] CE. Règlement (CE) n o 835/2011 de la Commission du 19 août 2011 modifiant le règlement (CE) no 1881/2006 en ce qui concerne les teneurs maximales pour les hydrocarbures aromatiques polycycliques dans les denrées alimentaires. Journal officiel de l'Union européenne L 215, 8p, 2011.

[10] Hansen D., Di Toro D., McGrath J., Swartz R., Mount D., Burgess R., Ozretich R., Bell H., Linton T., Procedures for the derivation of equilibrium partitioning sediment benchmarks (ESBs) for the protection of benthic organisms: PAH

mixtures. USEPA, Narragansett, RI: Duluth, MN: Newport, Or. 175p, 2003.

[11] INERIS, Caractérisation des émissions de polluants engendrés par l'incendie de cinq produits types. Rapport d'Etude DRC-09-93632-01522A. 48p, 2009.

[12] Nisbet I.C.T. and LaGoy P.K., Toxic equivalency factor (TEFs) for polycyclic aromatic hydrocarbons (PAHs). Regulatory Toxicology and Pharmacology, 16, pp. 270-300, 1992.

[13] Brune H., Deutsch-Wenzel, R. P., Habs M., Ivankovic S. and Schmähl D. Investigation of the tumourigenic response to benzo(a)pyrene in aqueous caffeine solution applied orally to Sprague-Dawley rats. Journal of Cancer Research and Clinical Oncology, 102, , pp.153-157, 1981.

[14] Bismuth C., Baud F., Conso F., Dally S., Fréjaville J.-P., Garnier R. and Jaeger A. "Toxicologie clinique ". Flammarion Médecine-Sciences, 5ème édition, Paris, 1092p , 2000.

[15] OFSP, Office Fédéral de la Santé Publique, unité de direction protection des consommateurs Juillet 2012. Les hydrocarbures aromatiques polycycliques (HAP) , 3p, 2012.

[16] INERIS, « Hydrocarbures aromatiques polycycliques (HAPs), Evaluation de la relation dose réponse pour des effets cancérigènes. Approche substance par substance : FET et approche par mélange. » Rapport final, 18 décembre 2003, 64p, 2003.

[17] Ballerini D., Gatellier C. et Vogel T., Techniques de traitement par voie biologique des sols pollués, ADEME Editions, 198 p, 1998.

[18] Mackay D., Shiu W.Y. et Ma K.C**.** , Illustrated handbook of physical-chemical properties and environmental fate for organic chemicals. Lewis Publishers, 2, 925p,1992.

[19] Tarantini A., Modulation de la génotoxicité des hydrocarbures aromatiques polycycliques (HAP) en mélanges. Thèse, Université Joseph-Fourier – Grenoble, France, 168p, 2009.