

Original Research Article

LEVEL OF POLYCYCLIC AROMATIC HYDROCARBON (PAH) CONTAMINATION IN BRAISED FISH MARKETED AND CONSUMED IN EIGHT (8) COMMUNES IN THE DISTRICT OF ABIDJAN, CÔTE D'IVOIRE.

Comment [1]: Unnecessary.

ABSTRACT

Aims : The objective of this study is to evaluate the exposure of consumers to polycyclic aromatic hydrocarbons (PAHs) and to estimate the health risk related to the consumption of braised fish from eight (8) communes of the District of Abidjan in order to contribute to the protection of these consumers.

Study Design : This experimental study consisted in sampling fish cooked with charcoal embers in the collective catering circuit, followed by the determination of PAHs and the estimation of health risks.

Place and Duration of Study : The braised fish samples were collected in the Autonomous District of Abidjan and analyses were performed at the Central Laboratory of Agrochemistry and Ecotoxicology between January and March 2015.

Methodology : PAHs were extracted and purified on a Bond Eut C18 cartridge and then determined by High Performance Liquid Chromatography (HPLC) according to ISO 15753.

Results : All the eight (8) PAH molecules investigated were found in varying concentrations and numbers in the braised fish depending on their origin. Concentrations range from 0.013 µg/kg to 1.246 µg/kg for individual molecules and from 0.670 µg/kg to 3.271 µg/kg for total PAHs (TPAHs) regardless of the municipality and PAH molecule. In addition, 0-25% of the samples are non-compliant with the standard for PAHs and 0-13% for TPAHs. Also, braised fish collected in the communes of Marcory and Plateau contain eight (8) molecules while

variable numbers of molecules are found in those from other communes. Thus, seven (7) molecules were found in four communes (Abobo, Cocody, Koumassi, Port-Bouet) and six (6) molecules in two communes (Treichville, Yopougon). However, all hazard quotients (0.00000-0.00041) are less than 1 regardless of the municipality and PAH molecule.

Conclusion : These results are essential to improve the safety of food of animal origin, especially braised fish made available to consumers in the District of Abidjan. These results recommend the consideration of other sources of PAH in the diet although the hazard quotients are less than 1 because PAH molecules have a cumulative power in the body.

Keywords: communes; braised fish, PAH, HPLC, intakes, Abidjan.

1. INTRODUCTION

Polycyclic Aromatic Hydrocarbons (PAHs) are natural constituents of coal and petroleum or can be derived from the incomplete combustion of organic materials (fuel, wood, tobacco). Ils sont composés d'atomes de carbone formant des cycles benzéniques (2 à 10) et d'atomes d'hydrogène [1]. Among these molecules, sixteen (16) have been prioritized by the United States Environmental Protection Agency (US-EPA) due to their abundance in the environment and/or their extensive use by industry. The fear caused by PAHs comes from the fact that they are contaminants resulting from combustion processes and some of these molecules are considered potentially carcinogenic, including Benzo (a) Pyrene [2]. However, the World Health Organization (WHO) proposes to monitor mainly six (6) of the sixteen (16) PAH molecules that are considered as priorities because of their harmful effects on the environment and their high toxicity [3]. These are Benzo (a) Pyrene, Fluoranthene, Benzo (a) Fluoranthene, Dibenzo (ah) Anthracene, Benzo (k) Fluoranthene, Benzo (g,h,i) Perylene and Indeno (1,2,3-cd) Pyrene. These molecules are ubiquitous in the environment (air, water, soil) and food, especially fish [4][5][6][7].

Comment [2]: This sentence is equal to one in the following article:
<<https://www.m.elewa.org/Journals/wp-content/uploads/2022/06/3.Dagnogo.pdf>>. Please, rephrase it.

Comment [3]: Please, translate to english.

Fish are very varied aquatic living beings whose nutritional and sanitary qualities are recognized. They have a low fat and calorie content and help reduce cholesterol levels. Fish and fish products play an important role in the diet of West African populations [8], particularly Ivorians. Indeed, in Côte d'Ivoire, fish is the consumer's primary source of animal protein with 15.9 kg/year/inhabitant [9][10]. However, more than 80% of the fish consumed in Côte d'Ivoire is derived from artisanal processing, notably drying, frying, braising and smoking. These modes of transformation would under certain conditions produce toxic substances such as PAHs [11][12]. Thus grilling, roasting and smoking, with charcoal, can increase the concentration of PAHs in food [6]. Aké et al.[12] showed that wood smoking leads to an increase in PAH concentration in hot smoked fish. The process of smoking fish is similar in some respects to the braising of fish. Thus, braising could be a source of PAH contamination in braised fish. However, unlike smoked fish, no regulations have yet been established for braised fish at the international, regional and national levels. While Dagnogo et al [13] have shown that braised fish can be a source of PAH contamination depending on their species. Thus, the knowledge of the level of PAH contamination of braised fish according to their production and marketing area becomes essential insofar as this foodstuff is widely consumed in urban areas in many countries, particularly in Côte d'Ivoire. This study was initiated in the Autonomous District of Abidjan to evaluate the exposure to PAHs of consumers of braised fish in eight (8) communes of the District of Abidjan and to estimate the health risk related to the said consumption in order to contribute to the protection of these consumers.

2. MATERIALS AND METHODS

2.1 Sampling

The biological material consists of fish cooked with charcoal embers, called "braised fish". Thus, eighty-six (86) samples of braised fish were collected, in a random and simple way, in the collective catering circuit of the Autonomous District of Abidjan from January to March 2015 in the following proportions: ten (10) Abobo, ten (10) Cocody, ten (10) Koumassi, ten (10) Marcory, twelve (12) Plateau, twelve (12) Port-Bouet, ten (10) Treichville and twelve (12) Yopougon.

Comment [4]: How was it? What was this random and simple way ?

2.2 Determination of PAHs

PAH extraction and analysis was performed according to ISO 15753-2004 as reported by Aké et al. [14]. In a centrifuge tube containing 2.5 g of the braised fish sample grind, 10 ml of acetonitrile/acetone mixture (60/40; v/v) was introduced. The mixture was vortexed for 30 seconds and ultrasonicated for 5 minutes before being centrifuged for 5 minutes at 4000 rpm. The upper phase was collected and transferred to a tared conical tube (Falcon®). The extraction was repeated a second time with 10 ml of the acetonitrile/acetone mixture (60/40; v/v). After centrifugation, the upper phase was collected and transferred to the same tared conical tube (Falcon®) and the solvent was evaporated using a rotary evaporator at 35°C. Two (2) ml of the acetonitrile/acetone mixture (60/40; v/v) was introduced into the conical tube containing the dry extract which was vortexed for 15 seconds and then centrifuged for 30 seconds. The upper phase was transferred to a tube and the operation is repeated twice. The supernatant was transferred to a Bond Elut cartridge (C18; 500 mg/6 ml) previously conditioned with 12 ml methanol and 12 ml acetonitrile. Elution was performed with 5 ml of the acetonitrile/acetone mixture at atmospheric pressure. The eluate was concentrated, to 50 mg, using a rotary evaporator at 35°C. The purified extract was recovered in 1 ml of hexane in a tube that was crimped and stored at -18°C before analysis.

Comment [5]: "was"

A Shimadzu High Performance Liquid Chromatograph (HPLC), coupled with a UV/VIS SPD-20A detector, was used under the operating conditions described in Table 1.

Table 1 : Analytical conditions by HPLC

Column C18	40° C		
Binary gradient	Solvent A: Water Solvent B: Acetonitrile		
Flow rate	1 ml/min		
Duration of the analysis	10 min		
Wavelength	254 nm		
Elution mode	Isocratic		
PAHs searched	Abbreviation	Limit of detection (LD ; µg/Kg)	Limite de quantification (LQ ; µg/Kg)
Fluoranthene	F	0.017	0.060
Pyrene	P		
Benzo (k) Fluoranthene	Bkf		
Benzo (a) Pyrene	BaP		
Indeno (1,2,3-cd)	Ind (1,2,3 cd)		
Pyrene	Py		
Benzo (g,h,i) Perylene	B (g,h,i) Py		
Benzo (a) Anthracene	BaA		
Benzo (b) Fluoranthene	BbF		

2.3 Estimation of daily PAH intakes

A deterministic approach was adopted for the estimation of PAH intake. It consisted of multiplying a fixed value of fish food consumption by the concentration of PAHs found in the fish and dividing this product by the actual body weight of the individual [15][16][13]. In Côte d'Ivoire, the amount of fish consumed daily per inhabitant is 44.17 g [9][10]. PAH intake was calculated according to the formula:

$$\mathbf{AJE} = (C_i \times Q) / P$$

Where AJE is the estimated daily intake of PAHs ($\mu\text{g}/\text{kg bw}/\text{d}$) for a 60 kg adult individual; C_i is the average concentration of PAHs ($\mu\text{g}/\text{kg}$); Q is the amount of fish consumed per day per capita (kg) and P is the body weight (kg).

2.4 Estimation of the hazard ratio(RD)

The hazard ratio was determined as the ratio of the estimated daily intake to the toxicological reference value (TRV).

$$\mathbf{RD} = \text{AJE}/\text{VTR}$$

Where AJE is the estimated daily intake of PAHs ($\mu\text{g}/\text{kg bw}/\text{d}$) for a 60 kg adult individual; VTR is the toxicological reference value for PAHs ($\mu\text{g}/\text{kg bw}/\text{d}$).

This ratio only provides information on the potential occurrence of adverse effects and not on their importance. Thus when :

- $\text{RD} < 1$ means that the exposed population is theoretically out of danger, i.e. this exposed population is not likely to develop the health effects studied.
- $\text{RD} > 1$ means that the toxic effect can occur without being able to predict the probability of this event occurring [15][17].

2.5 Statistical analysis

All tests were performed in triplicate. Averages were calculated with standard deviations to assess the level of PAH contamination. One-way analysis of variance (ANOVA) was performed to determine the existence of statistically significant differences between the calculated means. Statistically significant differences were revealed by the Student-Newman-Keuls test at the 0.05 risk using SPSS version 20.0 software. A multivariate principal component analysis (PCA) was performed using STATISTICA Software (version 7.1) to structure the variability between PAH concentrations and fish sampling municipalities. The percentages and proportions made it possible to assess, respectively, the occurrence of PAHs in the sampling municipalities and the risks incurred by consumers in relation to the Maximum Limit and the Daily Dose Authorized.

3. RESULTS AND DISCUSSION

3.1 Results

3.1.1 Polycyclic aromatic hydrocarbon concentration in fish

The average PAH contamination levels of braised fish, according to the communes, are given in Table 2. Eight (8) PAH molecules were found in the samples taken in the communes of Marcory and Plateau, seven (7) molecules in those of Abobo, Cocody and Koumassi, and six (6) molecules in the communes of Treichville and Yopougon. The mean levels of Fluoranthene in braised fish ranged from 0.015 ± 0.019 $\mu\text{g}/\text{kg}$ (Abobo) to 1.018 ± 2.862 $\mu\text{g}/\text{kg}$ (Marcory). Statistical analysis revealed no significant difference between the different concentrations ($P = 0.62$). The Pyrene molecule has average concentrations of 0.273 ± 0.205 $\mu\text{g}/\text{kg}$, 0.920 ± 1.534 $\mu\text{g}/\text{kg}$, 0.506 ± 1.432 $\mu\text{g}/\text{kg}$, 0.650 ± 1.464 $\mu\text{g}/\text{kg}$, 0.029 ± 0.065 $\mu\text{g}/\text{kg}$, $0.756 \pm 1, 282$ $\mu\text{g}/\text{kg}$ and 0.408 ± 0.601 $\mu\text{g}/\text{kg}$ found in braised fish from the communes of Abobo, Koumassi, Marcory, Plateau, Port-bouet, Treichville and Yopougon respectively. It was not detected in the samples from Cocody and no significant difference was observed

between the values from the other communes ($P = 0.69$). The average concentrations of Benzo (k) Fluoranthene found in braised fish are 0.073 ± 0.048 $\mu\text{g}/\text{kg}$ (Abobo), 0.512 ± 0.997 $\mu\text{g}/\text{kg}$ (Cocody), 0.193 ± 0.238 $\mu\text{g}/\text{kg}$ (Koumassi), 0.109 ± 0.171 $\mu\text{g}/\text{kg}$ (Marcory), 0.421 ± 0.766 $\mu\text{g}/\text{kg}$ (Plateau), 1.246 ± 3.198 $\mu\text{g}/\text{kg}$ (Port-Bouet), 0.713 ± 0.736 $\mu\text{g}/\text{kg}$ (Treichville) and 0.094 ± 0.106 $\mu\text{g}/\text{kg}$ (Yopougon); no significant difference was observed between concentrations ($P = 0.54$). For the molecule Benzo (a) Pyrene, the average concentrations ranged from 0.024 ± 0.048 $\mu\text{g}/\text{kg}$ (Abobo) to 0.634 ± 1.200 $\mu\text{g}/\text{kg}$ (Cocody). Statistical analysis revealed no significant difference between the concentrations ($P = 0.13$) and this molecule was not detected in the samples taken in the communes of Port-Bouet and Treichville. The average levels of Indenol (1,2,3-cd) Pyrene are 0.051 ± 0.059 $\mu\text{g}/\text{kg}$, 0.895 ± 1.201 $\mu\text{g}/\text{kg}$, 0.406 ± 1.041 $\mu\text{g}/\text{kg}$, 0.326 ± 0.566 $\mu\text{g}/\text{kg}$, 0.013 ± 0.029 $\mu\text{g}/\text{kg}$ and 0.088 ± 0.249 $\mu\text{g}/\text{kg}$ for braised fish from the communes of Abobo, Cocody, Koumassi, Marcory, Plateau and Port-Bouet respectively. These values did not allow discrimination between communes at risk $P = 0.11$ and the said molecule was not detected in braised fish collected in the communes of Treichville and Yopougon. For Benzo (g,h,i) Perylene, the average concentrations are 0.092 ± 0.106 $\mu\text{g}/\text{kg}$ (Abobo), 0.176 ± 0.432 $\mu\text{g}/\text{kg}$ (Cocody), 0.068 ± 0.149 $\mu\text{g}/\text{kg}$ (Marcory), 0.048 ± 0.094 $\mu\text{g}/\text{kg}$ (Plateau), 0.923 ± 1.352 $\mu\text{g}/\text{kg}$ (Port-Bouet), 0.050 ± 0.141 $\mu\text{g}/\text{kg}$ (Treichville) and the highest concentration was obtained in Port-Bouet ($P = 0.01$). However, this molecule was not detected in braised fish collected in the communes of Koumassi and Yopougon. As for Benzo (a) Anthracene, it has average contents of 0.035 ± 0.054 $\mu\text{g}/\text{kg}$, 0.062 ± 0.115 $\mu\text{g}/\text{kg}$, 0.359 ± 1.016 $\mu\text{g}/\text{kg}$, 0.020 ± 0.068 $\mu\text{g}/\text{kg}$, 0.022 ± 0.062 $\mu\text{g}/\text{kg}$, 1.023 ± 1.488 $\mu\text{g}/\text{kg}$ and 0.083 ± 0.259 $\mu\text{g}/\text{kg}$ determined in braised fish from the communes of Cocody, Koumassi, Marcory, Plateau, Port-Bouet, Treichville and Yopougon respectively. The high level is obtained in Treichville ($P = 0.03$), but this molecule was not detected in the samples taken in Abobo. Benzo (b) Fluoranthene concentrations ranged from 0.119 ± 0.336 $\mu\text{g}/\text{kg}$

(Marcory) to 0.710 ± 1.469 $\mu\text{g/kg}$ (Treichville) and no significant difference was observed between these values at P -value = 0.91. The average levels of total polycyclic aromatic hydrocarbons (TPAH) are 0.670 ± 0.617 $\mu\text{g/kg}$ (Abobo), 3.150 ± 3.036 $\mu\text{g/kg}$ (Cocody), 2.421 ± 2.173 $\mu\text{g/kg}$ (Koumassi) 2.633 ± 3.944 $\mu\text{g/kg}$ (Marcory), 1.474 ± 2.152 $\mu\text{g/kg}$ (Plateau), 3.136 ± 4.728 $\mu\text{g/kg}$ (Port-Bouet), 3.271 ± 3.964 $\mu\text{g/kg}$ (Treichville) and 1.430 ± 2.984 $\mu\text{g/kg}$ (Yopougon). Statistical analysis revealed no significant difference at $P = 0.73$.

UNDER PEER REVIEW

Table 2 : Average concentrations of polycyclic aromatic hydrocarbons in braised fish according to the commune ($\mu\text{g}/\text{kg}$)

Communes	Fluo	Pyrè	BkFluo	BaPyr	IndPyr	BgPyr	BaAnt	BbFluo	TPAH
Abobo	0.015±0.019a	0.273±0.205a	0.073±0.084a	0.024±0.048a	0.051±0.059a	0.092±0.106b	< LD	0.143±0.285a	0.670±0.617a
Cocody	0.754±1.441a	< LD	0.512±0.997a	0.634±1.200a	0.895±1.201a	0.176±0.432ab	0.035±0.054b	0.293±0.333a	3.150±3.036a
Koumassi	0.244±0.425a	0.920±1.534a	0.193±0.238a	0.369±0.284a	0.406±1.041a	< LD	0.062±0.115b	0.226±0.640a	2.421±2.173a
Marcory	1.018±2.862a	0.506±1.432a	0.109±0.171a	0.128±0.243a	0.326±0.566a	0.068±0.149b	0.359±1.016ab	0.119±0.336a	2.633±3.944a
Plateau	0.105±0.111a	0.650±1.464a	0.421±0.766a	0.073±0.226a	0.013±0.029a	0.048±0.094b	0.020±0.068b	0.149±0.510a	1.474±2.152a
Port-Bouet	0.693±1.902a	0.029±0.065a	1.246±3.198a	< LD	0.088±0.249a	0.923±1.352a	0.022±0.062b	0.134±0.248a	3.136±4.728a
Treichville	0.019±0.031a	0.756±1.282a	0.713±0.736a	< LD	< LD	0.050±0.141b	1.023±1.488a	0.710±1.469a	3.271±3.964a
Yopougon	0.045±0.120a	0.408±0.601a	0.094±0.106a	0.168±0.516a	< LD	< LD	0.083±0.259b	0.633±2.194a	1.430±2.984a
Fvalue	0.763	0.681	0.865	1.619	1.793	3.230	2.416	0.388	0.633
Pvalue	0.62	0.69	0.54	0.15	0.111	0.01	0.03	0.91	0.73

Means in the same column with the same letter are not significantly different at risk $P = 0.05$.

Fluo : Fluoranthene, **Pyrè** : Pyrene, **BkFluo** : Benzo (k) Fluoranthene, **BaPyr** : Benzo (a) Pyrene, **IndPyr** : Indeno (1,2,3-cd) Pyrene, **BgPyr** : Benzo (g,h,i) Perylene, **BaAnt** : Benzo (a) Anthracene, **BbFluo** : Benzo (b) Fluoranthene, **TPAH** : Total Polycyclic Aromatic Hydrocarbons.

3.1.2 Proportion of PAH positive braised fish samples

The braised fish samples are positive to PAH and the proportions vary between 0% (Pyrene, Benzo (a) Pyrene, Indeno (1,2,3-cd) Pyrene, Benzo (g,h,i) Perylene, Benzo (a) Anthracene) and 88% (Benzo (k) Fluoranthene). Concerning the Fluoranthene molecule, the proportions of positive samples are 50%, 50%, 75%, 38%, 67%, 38%, 38% and 33% for braised fish from the communes of Abobo, Cocody, Koumassi, Marcory, Plateau, Port-Bouet, Treichville and Yopougon respectively. The proportions of positive braised fish samples for Pyrene are 0% (Cocody), 13% (Marcory), 25% (Port-Bouet), 38% (Koumassi), 50% (Plateau, Yopougon), 63% (Treichville) and 75% (Abobo). For Benzo (k) Fluoranthene, the proportions of positive samples of braised fish are between 33% (Cocody) and 88% (Treichville). For Benzo (a) Pyrene the proportions of positive braised fish samples are 25%, 50%, 75%, 38%, 17%, 0%, 0%, and 25% for the communes of Abobo, Cocody, Koumassi, Marcory, Plateau, Port-Bouet, Treichville and Yopougon respectively. As for Indeno (1,2,3-cd) Pyrene, it is found in 0% (Treichville, Yopougon), 13% (Port-Bouet), 25% (Koumassi, Plateau), 38% (Marcory), 50% (Abobo) and 67% (Cocody) of the braised fish samples analyzed. Concerning Benzo (g,h,i) Perylene, the rates of contaminated braised fish samples vary between 0% (Koumassi, Yopougon) and 75% (Abobo). The proportions of braised fish samples contaminated with Benzo (a) Anthracene are 0%, 33%, 38%, 13%, 8%, 13%, 38% and 17% for the communes of Abobo, Cocody, Koumassi, Marcory, Plateau, Port-Bouet, Treichville and Yopougon respectively. Benzo (b) Fluoranthene was found in 8% (Yopougon) to 67% (Cocody) of braised fish samples (Table 3).

Table 3 : Proportion of PAH positive braised fish samples by municipality (percentage)

Communes	Fluo	Pyrè	BkFluo	BaPyr	IndPyr	BgPyr	BaAnt	BbFluo
Abobo	50	75	50	25	50	75	0	25
Cocody	50	0	33	50	67	17	33	67

Koumassi	75	38	75	75	25	0	38	13
Marcory	38	13	50	38	38	25	13	13
Plateau	67	50	67	17	25	25	8	17
Port-Bouet	38	25	38	0	13	38	13	25
Treichville	38	63	88	0	0	13	38	38
Yopougon	33	50	75	25	0	0	17	8

Total number of samples: 86

Fluo : Fluoranthene, **Pyre** : Pyrene, **BkFluo** : Benzo (k) Fluoranthene, **BaPyr** : Benzo (a) Pyrene, **IndPyr** : Indeno (1,2,3-cd) Pyrene, **BgPyr** : Benzo (g,h,i) Perylene, **BaAnt** : Benzo (a) Anthracene, **BbFluo** : Benzo (b) Fluoranthene.

3.1.3 Proportion of braised fish samples not conforming to quality standards

Table 4 shows the proportions of braised fish samples with PAH concentrations above the standards of 2 µg/kg (individual molecule) or 12 µg/kg (total PAH). Concerning the municipality of Abobo, the proportion of non-compliant braised fish samples is 0% for all PAH molecules. No non-compliance was detected for total PAHs for this commune. As for the commune of Cocody, it has proportions of non-compliance of 17% (Fluoranthene, Benzo (k) Fluoranthene, Benzo (a) Pyrene, Indeno (1,2,3-cd) Pyrene) with 0% for total PAH. The proportions of non-compliant braised fish samples in Koumassi are 13% and 25% respectively for Pyrene and Indeno (1,2,3-cd) Pyrene with 0% for total PAH. The proportion of non-compliance obtained in Marcory is 13% for Fluoranthene, Pyrene and Benzo (a) Anthracene with 0% for total PAH. It is 8% at Plateau for Pyrene and Benzo (k) Fluoranthene with 0% for total PAH. For the commune of Port-Bouet, the non-compliance rates are 13% (Fluoranthene, Benzo (k) Fluoranthene), 25% (Benzo (g) Pyrene) and 13% (total PAH).

Concerning the commune of Treichville, the proportions of non-compliant braised fish samples are 13% (Benzo (k) Fluoranthene, Benzo (b) Fluoranthene, total PAH) and 25% (Pyrene, Benzo (a) Anthracene). No non-compliance was observed in Yopougon for any PAH molecule except for Benzo (b) Fluoranthene (8%).

Table 4 : Proportion of braised fish samples not compliant with quality standards (percentage).

Communes	Fluo	Pyrè	BkFluo	BaPyr	IndPyr	BgPyr	BaAnt	BbFluo	TPAH
Abobo	0	0	0	0	0	0	0	0	0
Cocody	17	0	17	17	17	0	0	0	0
Koumassi	0	25	0	0	13	0	0	0	0
Marcory	13	13	0	0	0	0	13	0	0
Plateau	0	8	8	0	0	0	0	0	0
Port-Bouet	13	0	13	0	0	25	0	0	13
Treichville	0	25	13	0	0	0	25	13	13
Yopougon	0	0	0	0	0	0	0	8	0
Standards (µg/kg)	2								12

Total number of samples : 86

Fluo : Fluoranthene, **Pyrè** : Pyrene, **BkFluo** : Benzo (k) Fluoranthene, **BaPyr** : Benzo (a) Pyrene, **IndPyr** : Indeno (1,2,3-cd) Pyrene, **BgPyr** : Benzo (g,h,i) Perylene, **BaAnt** : Benzo (a) Anthracene, **BbFluo** : Benzo (b) Fluoranthene, **TPAH** : Total Polycyclic Aromatic Hydrocarbons.

3.1.4 Variability of municipalities in relation to PAH molecules.

Figure 1 shows the projection of communes and PAH molecules in the principal component analysis plane. The projection in the comp1-comp2 plane expresses 66.56% of the variability. The communes of Treichville, Plateau, Abobo and Yopougon and the molecules of Benzo (a) Anthracene, Benzo (b) Fluoranthene and Pyrene are positively correlated to component 1 whereas the commune of Cocody and the molecules of Indeno (1,2,3-cd) Pyrene and Fluoranthene are negatively correlated. On the other hand, the communes of Koumassi, Marcory and the Benzo (a) Pyrene molecule are positively correlated to component 2, while the commune of Port Bouet as well as the Benzo (k) Fluoranthene and Benzo (g,h,i) Perylene molecules are negatively correlated to component 2.

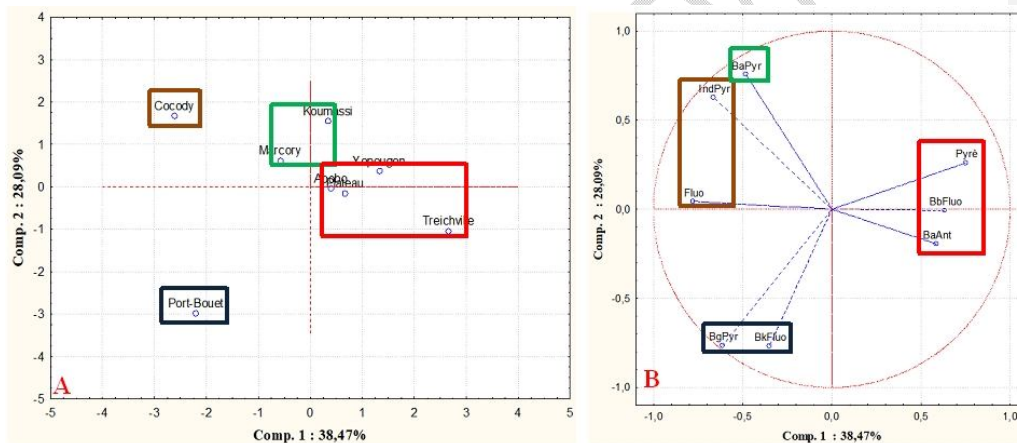


Figure 1: Projection of municipalities (A) and PAH molecules (B) in the Principal Component Analysis design.

Fluo : Fluoranthene, **Pyrè** : Pyrene, **BkFluo** : Benzo (k) Fluoranthene, **BaPyr** : Benzo (a) Pyrene, **IndPyr** : Indeno (1,2,3-cd) Pyrene, **BgPyr** : Benzo (g,h,i) Perylene, **BaAnt** : Benzo (a) Anthracene, **BbFluo** : Benzo (b) Fluoranthene.

3.1.5 Estimated intake of PAHs.

The estimated intakes related to the various standards are 1.262 ng/kg Bw/d (PAH molecules) and 7.572 ng/kg Bw/d for all the PAH (TPAH). Concerning the commune of Abobo, the contributions are included between 0,000 ng/kg Bw/d (Benzo (a) Anthracene) and 0,172 ng/kg Bw/d (Pyrene) with 0,423 ng/kg Bw/d (PAH). As for the commune of Cocody, it has PAH intakes of 0.476 ng/kg Bw/d (Fluoranthene), 0.000 ng/kg Bw/d (Pyrene), 0.323 ng/kg Bw/d (Benzo (k) Fluoranthene), 0.400 ng/kg Bw/d (Benzo (a) Pyrene), 0.565 ng/kg Bw/d (Indeno (1,2,3-cd) Pyrene), 0.111 ng/kg Bw/d (Benzo (g,h,i) Perylene), 0.022 ng/kg Bw/d (Benzo (a) Anthracene), 0.185 ng/kg Bw/d (Benzo (b) Fluoranthene), and 1.988 ng/kg Bw/d (PAH). The intakes obtained in the commune of Koumassi are 0.154 ng/kg Bw/d, 0.581 ng/kg Bw/d, 0.122 ng/kg Bw/d, 0.233 ng/kg Bw/d, 0.256 ng/kg Bw/d, 0.000 ng/kg Bw/d, 0.039 ng/kg Bw/d, 0.143 ng/kg Bw/d and 1, 528 ng/kg Bw/d for Fluoranthene, Pyrene, Benzo (k) Fluoranthene, Benzo (a) Pyrene, Indeno (1,2,3-cd) Pyrene, Benzo (g,h,i) Perylene, Benzo (a) Anthracene, Benzo (b) Fluoranthene and TPAHs respectively. For the commune of Marcory, the estimated PAH intakes are between 0.043 ng/kg Bw/d (Benzo (g,h,i) Perylene) and 0.642 ng/kg Bw/d (Fluoranthene) with 1.661 ng/kg Bw/d for TPAHs. Concerning the Plateau commune, the estimated PAH intakes are 0.066 ng/kg Bw/d (Fluoranthene), 0.410 ng/kg Bw/d (Pyrene), 0.266 ng/kg Bw/d (Benzo (k) Fluoranthene), 0.046 ng/kg Bw/d (Benzo (a) Pyrene), 0.008 ng/kg Bw/d (Indeno (1,2,3-cd) Pyrene), 0.030 ng/kg Bw/d (Benzo (g,h,i) Perylene), 0.013 ng/kg Bw/d (Benzo (a) Anthracene), 0.094 ng/kg Bw/d (Benzo (b) Fluoranthene), and 0.930 ng/kg Bw/d (TPAH). The commune of Port-Bouet recorded PAH intakes of 0.437 ng/kg Bw/d, 0.018 ng/kg Bw/d, 0.786 ng/kg Bw/d, 0.000 ng/kg Bw/d, 0.056 ng/kg Bw/d, 0.582 ng/kg Bw/d, 0.014 ng/kg Bw/d, 0.085 ng/kg Bw/d and 1, 979 ng/kg Bw/d for Fluoranthene, Pyrene, Benzo (k) Fluoranthene, Benzo (a) Pyrene, Indeno (1,2,3-cd) Pyrene, Benzo (g,h,i) Perylene, Benzo (a) Anthracene, Benzo (b) Fluoranthene and TPAHs respectively. PAH intakes in the commune of Treichville vary between 0.012 ng/kg Bw/d

(Fluoranthene) and 0.646 ng/kg Bw/d (Benzo (a) Anthracene) with 2.064 ng/kg Bw/d (TPAH). The commune of Yopougon recorded PAH intakes of 0.028 ng/kg Bw/d (Fluoranthene), 0.257 ng/kg Bw/d (Pyrene), 0.059 ng/kg Bw/d (Benzo (k) Fluoranthene), 0.106 ng/kg Bw/d (Benzo (a) Pyrene), 0.000 ng/kg Bw/d (Indeno (1,2,3-cd) Pyrene; 0.000 ng/kg Bw/d Benzo (g,h,i) Perylene), 0.052 ng/kg Bw/d (Benzo (a) Anthracene), 0.399 ng/kg Bw/d (Benzo (b) Fluoranthene), and 0.902 ng/kg Bw/d (TPAH; Table 5).

Table 5 : Estimated PAH intakes for an adult Ivorian (ng/kg Bw/d).

Communes	Fluo	Pyrè	BkFluo	BaPyr	IndPyr	BgPyr	BaAnt	BbFluo	TPAH
Abobo	0.009	0.172	0.046	0.015	0.032	0.058	0.000	0.090	0.423
Cocody	0.476	0.000	0.323	0.400	0.565	0.111	0.022	0.185	1.988
Koumassi	0.154	0.581	0.122	0.233	0.256	0.000	0.039	0.143	1.528
Marcory	0.642	0.319	0.069	0.081	0.206	0.043	0.227	0.075	1.661
Plateau	0.066	0.410	0.266	0.046	0.008	0.030	0.013	0.094	0.930
Port-Bouet	0.437	0.018	0.786	0.000	0.056	0.582	0.014	0.085	1.979
Treichville	0.012	0.477	0.450	0.000	0.000	0.032	0.646	0.448	2.064
Yopougon	0.028	0.257	0.059	0.106	0.000	0.000	0.052	0.399	0.902
Standards	1.262								7.572

Fluo : Fluoranthene, **Pyrè** : Pyrene, **BkFluo** : Benzo (k) Fluoranthene, **BaPyr** : Benzo (a) Pyrene, **IndPyr** : Indeno (1,2,3-cd) Pyrene, **BgPyr** : Benzo (g,h,i) Perylene, **BaAnt** : Benzo (a) Anthracene, **BbFluo** : Benzo (b) Fluoranthene, **TPAH** : Total Polycyclic Aromatic Hydrocarbons.

3.1.6 Estimated risk for PAHs.

The estimated risks for individual molecules (0.00000-0.00013) are lower than that estimated for the maximum allowable limit (0.00025) and they also remain lower (0.00008-0.00041) than that obtained with the maximum allowable limit for total PAHs (0.00151) regardless of molecule and municipality. Moreover, all the estimated risks are lower than 1 whatever the municipality and the molecule searched (Table 6).

Table 6 : Estimated risk for an adult Ivorian.

Communes	Fluo	Pyrè	BkFluo	BaPyr	IndPyr	BgPyr	BaAnt	BbFluo	TPAH
Abobo	0.00000	0.00003	0.00001	0.00000	0.00001	0.00001	0.00000	0.00002	0.00008
Cocody	0.00010	0.00000	0.00006	0.00008	0.00011	0.00002	0.00000	0.00004	0.00040
Koumassi	0.00003	0.00012	0.00002	0.00005	0.00005	0.00000	0.00001	0.00003	0.00031
Marcory	0.00013	0.00006	0.00001	0.00002	0.00004	0.00001	0.00005	0.00002	0.00033
Plateau	0.00001	0.00008	0.00005	0.00001	0.00000	0.00001	0.00000	0.00002	0.00019
Port-Bouet	0.00009	0.00000	0.00016	0.00000	0.00001	0.00012	0.00000	0.00002	0.00040
Treichville	0.00000	0.00010	0.00009	0.00000	0.00000	0.00001	0.00013	0.00009	0.00041
Yopougon	0.00001	0.00005	0.00001	0.00002	0.00000	0.00000	0.00001	0.00008	0.00018
Standards	0.00025								0.00151

Fluo : Fluoranthene, **Pyrè** : Pyrene, **BkFluo** : Benzo (k) Fluoranthene, **BaPyr** : Benzo (a) Pyrene, **IndPyr** : Indeno (1,2,3-cd) Pyrene, **BgPyr** : Benzo (g,h,i) Perylene, **BaAnt** : Benzo (a) Anthracene, **BbFluo** : Benzo (b) Fluoranthene, **TPAH** : Total Polycyclic Aromatic Hydrocarbons.

3.2 Discussion

The study made it possible to highlight the presence of eight (8) molecules of Polycyclic Aromatic Hydrocarbons sought in the samples of "braised fish" collected in the communes of

Marcory and Plateau of the Abidjan District. However, variable numbers of molecules were found in the other communes, notably seven (7) molecules in four communes (Abobo, Cocody, Koumassi, Port-Bouet) and six (6) molecules in two communes (Treichville, Yopougon). The rate of positive samples varies from 0% to 88% but the average concentrations are all below the standard of 2 $\mu\text{g}/\text{kg}$ set by the European Union Regulation (EC) [20] regardless of the commune and the molecule. On the other hand, variable proportions of samples that do not comply with the standards are obtained in the communes and they vary from 0% to 25% (individual molecule) and from 0% to 13% (TPAH). No non-compliance is obtained in the commune of Abobo whatever the PAH molecule. This variability in contamination could be explained by certain factors related to the origin and fatty or non-fatty nature of the fish, the type of charcoal used and the mode of fish processing [18][11][12][13]. According to Aké et al. [11], PAH concentration is a function of the amount of fat in the processed fish. Also Dagnogo et al. [13] showed in their study that the mackerel fish species is more contaminated with PAH than the other species carp, pike and tilapia. Indeed, the mackerel species is also richer in fat than the other three species. This variability of contamination between the communes is also confirmed by the Principal Component Analysis for the distribution of molecules according to the communes. The communes of Marcory and Koumassi contain the greatest quantity of one molecule (Benzo (a) Pyrene), two molecules in the communes of Cocody (Fluoranthene, Indeno (1,2, 3-cd) Pyrene) and Port-Bouet (Benzo (g,h,i) Perylene, Benzo (k) Fluoranthene) and three molecules in the communes of Abobo, Plateau, Treichville and Yopougon (Pyrene, Benzo (a) Anthracene, Benzo (b) Fluoranthene). These contamination levels are not specific to "braised fish" as higher PAH concentrations are determined in several foodstuffs including smoked meats and fish [19]. This regulation indicates that smoked meat and fish from some EU countries cannot comply with the low levels of PAHs despite the application of good smoking practices. This situation

led to a double amendment of the regulation (EC) 1881/2006 by the regulations (EU) 835/2011 and (EU) 2020/1255 [19][20][21]. PAH intake from the smoking process and the fish collection site independently of the fish type was highlighted by Aké et al. [12]. Thus, these authors found variable levels of Benzo (a) Pyrene in fresh fish (14.65 µg/kg) and smoked fish (64.97 µg/kg). They also obtained variable levels of Benzo (a) Pyrene depending on the sampling sites including Macaci (24.76 µg/kg) and Port-Bouet (101.64 µg/kg). So the variability in contamination observed in this study could also be explained by the braising process of the vendors and the fish collection site. However, the Benzo (a) Pyrene levels obtained in this study are largely lower than those obtained by Aké et al. [11][12].

The standard for PAHs is respected whatever the commune visited. All concentrations (0.670 µg/kg-3.271 µg/kg) are below 12 µg/kg, which is the European Union standard for PAHs [19][20][21]. However, 13% of braised fish samples from the communes of Port-Bouet and Treichville have TPAH levels that do not comply with this standard. This situation could be the result of the high presence of mackerel among the braised fish from these communes. Dagnogo et al. [13] have shown that the presence of mackerel fish contributes to exceeding the TPAH standard.

In addition, consumption of "braised fish" from the communes visited provides between 0.000 ng PAH/kg Bw/d and 0.786 ng PAH/kg Bw/d to an adult Ivorian, regardless of the PAH molecule and the commune. All these individual intakes are lower than 1.262 ng PAH/kg Bw/d obtained from the 2 µg/kg standard. Thus, all of these intakes are lower than those obtained by Aké et al. [12] regardless of the condition (fresh or smoked) of the fish (0.09-52.01 ng/kg Bw/d) or smoked (0.47-32.51 ng/kg Bw/d). This difference could be related to the difference in fish treatment and the type of fish. Indeed, the fish used by Aké et al. [12] are sardines that have undergone smoking, while those used in this study group together several species including Tilapia, carp, mackerel and pike that have undergone braising. Apart

from the type of fish, "braised fish" and smoked fish would not be governed by the same cooking conditions or criteria (duration of exposure of the fish to rising smoke, temperature and duration of cooking). In fact, smoked fish would be exposed for 2 to 5 hours to rising wood smoke at a temperature of more than 60°C while braised fish would be cooked after 30 minutes at a temperature supposed to be the same or close to 60°C.

Concerning the health risk, all the determined danger ratios (0.00000-0.00041) are lower than 1 whatever the commune and the PAH molecule searched, which would mean that the Ivorian adult consumers of braised fish, coming from the eight (8) communes, are out of danger. However, this situation should not obscure the consideration of other diffuse sources of intake of PAH molecules in the diet. Because these molecules would have a very important cumulative power in the body.

Comment [6]: This affirmation must have reference!

4. CONCLUSION

The chemical analysis of "braised fish" samples collected in eight (8) communes of the District of Abidjan revealed the presence of eight (8) PAH molecules. The molecules are distributed differently in number according to the commune of origin of the braised fish samples. Braised fish collected in the communes of Abobo, Plateau, Treichville and Yopougon contain three (3) molecules in relatively large quantities, while those collected in the communes of Cocody and Port-Bouet contain two (2). On the other hand, braised fish from the communes of Koumassi and Marcory contain one molecule in large quantities. A priori, the consumption of "braised fish" from the eight (8) municipalities visited could not present a major risk for the consumer because all the determined danger ratios are lower than 1. Thus, these results recommend the consideration of other sources of PAHs in the risk assessment of consumers of braised fish from the eight (8) municipalities in this study.

Therefore, further studies are desired to evaluate the effect of fuel type (charcoal), fish species and braising method on the PAH contamination level of braised fish.

REFERENCES

1. Hylland K. Polycyclic Aromatic Hydrocarbon (PAH) Ecotoxicology in Marine Ecosystems. *Journal of Toxicology and Environmental Health*. 2006 ; Part A 69 : 109-123.
2. Garziandia L. Polycyclic Aromatic Hydrocarbons (PAH), in ambient air. Report INERIS-LCSQA-Convention 41/2000. 2000 ; 50p.
3. Bour O. Polycyclic aromatic hydrocarbons; Methodological guide. INERIS. 2005 ; volume 1, N°66244-DESP-R01, 18p.
4. Buet A. Biological impact of PAHs in the European eel. Definition and validation of in situ biomarkers. Thèse de Doctorat, Université Paris-Sud XI. 2002 ; 194 p.
5. Normand J. Literature review: Toxic residues generated during the cooking of beef. 2007 ; Volume I, N° 170732024, 61 p.
6. EFSA. Polycyclic aromatic hydrocarbons in food. Scientific opinion of the Panel on Contaminants in the Food Chain. (Question No EFSA-Q-2007-136). 2008 ; http://www.efsa.europa.eu/EFSA/efsa_locale-1178620753812_1211902034842.htm.
7. Vignet C. Alteration of the physiology of fish exposed to polycyclic aromatic hydrocarbons (PAH): Behavior and Reproduction. D. thesis: specialty of physiology, biology of organisms, populations, interaction. University of Rochelle, GUY LUSSAC. 2014 ; 360p.
8. FAO. The State of World Fisheries and Aquaculture 2020. Sustainability in action. Rome. 2020. <https://doi.org/10.4060/ca9229fr>.
9. FAO (Food and Agriculture Organization of the United Nations). The State of World Fisheries and Aquaculture. 2012 ; 239p, ISSN 1020-5497.

10. MIRA H. Strategic Plan for the Development of Livestock, Fisheries and Aquaculture in Côte d'Ivoire, PSDEPA, Côte d'Ivoire. 2014 ; 87p
11. Ake Y, Biego GHM, Koffi KM, Kouame P. Validation de la méthode de détermination du Benzo(a)pyrène dans les poissons frais et fumés vendus et consommés en Côte d'Ivoire. *Revue Africaine de Santé et de Productions Animales*. 2010 ; 8 (53) : 54- 58 p.
12. Aké AY, Anon N, Kouamé P, Bonfoh B, Biego GHM. Assessment of the exposure to Benzo(a)Pyrene (BaP) contained in sardines (*Clupeidae*) consumed by the ivorian adult in the area of Abidjan. *International Journal of Science and Research*. 2014 ; 3 (11) : 2455-2461. Paper ID: OCT141136.
13. Dagnogo K, Coulibaly A, Kallo V, Doumbia M, Dongo CA, Boka OM. Et al. Health risk related to Polycyclic Aromatic Hydrocarbons (PAH) contamination of four species of fish braised and consumed in the District of Abidjan, Côte d'Ivoire. *Journal of Applied Biosciences*. 2022 ; 174: 18056-18068. doi.org/10.35759/JABs.174.3.
14. Ake AY, Biego GHM, Sess AD, Koffi KM, Kouame P, Bonfoh B et al. Validation of a method for the quantification of polycyclic aromatic hydrocarbons in fish. *European Journal of Scientific Research*. 2012 ; 74 (1) : 69-78.
15. Coulibaly A, Dagnogo K, Sidibe D, Silue N, Dembele A, Biego GH. Daily intake of aflatoxins from cocoa (*Theobroma cacao*) product in Côte d'Ivoire. *International Journal of Science and Research*. 2016; 5 (5): 1517-1522. Paper ID: NOV163664.
16. Dagnogo K, Coulibaly A, Kaba V, Kallo V, Dongo AC, Soro K. et al. Determination of the level of contamination and the intake of tylosin (macrolide) residues in eggs for consumption in the District of Abidjan (Côte d'Ivoire). *Journal of Applied Biosciences*. 2018 ; 129 :13067–13074. doi.org/10.4314/jab.v129i1.10.
17. Bamba S, Coulibaly A, Sidibe D, Nyamien BY, Biego GHM. Assessment of the risk of exposure to aflatoxins found in maize (*Zea mays L.*) produced in Côte d'Ivoire in ivorian

adults. Asian Food Science Journal. 2021 ; 20(7) : 72-81. DOI: 10.9734/AFSJ/2021/v20i730323.

18. Arias AH, Spetter CV, Freije RH, Marcovecchio JE. Polycyclic aromatic hydrocarbons in water, mussels (*Brachidontes sp.*, *Tagelus sp.*) and fish (*Odontesthes sp.*) from Bahía Blanca Estuary, Argentina. Estuarine, Coastal and Shelf Science. 2009 ; 85 : 67-81.

19. UE. Commission Regulation (EU) 2020/1255 of 7 September 2020 amending Regulation (EC) No 1881/2006 as regards the maximum levels of polycyclic aromatic hydrocarbons (PAHs) in traditionally smoked meat, meat products, fish and fishery products and setting a maximum level of PAHs in food powders of plant origin used for the preparation of beverages. 2020 ; L 293 : 4p.

20. CE. Commission Regulation (EC) No 1881/2006 of 19 December 2006 setting maximum levels for certain contaminants in foodstuffs. 2006 ; L 364 : 20p.

21. UE. Commission Regulation (EU) No 835/2011 of 19 August 2011 amending Regulation (EC) No 1881/2006 as regards maximum levels for polycyclic aromatic hydrocarbons in foodstuffs. Official Journal of the European Union. Brussels. 2011.