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Research Article
Chemical Contaminants in Traditionally Smoked Chicken Sold in The Open Markets of Lomé

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Abstract

Background and Objective: Safety is an important issue in international food trade. In Lomé (Togo) chicken meat is imported without effective control, moreover, its traditionally processed smoked chicken meat is contaminated by some chemical compounds. This study aimed to estimate some of these chemical compounds in smoked chicken meat retailed to consumers in Lomé. **Materials and Methods:** A total of 32 samples were obtained from the main open markets of Lomé and the quantification of Antibiotics Residues (ARs) was performed using a multi-class multi-residue of veterinary drugs and LC-MS/MS method while the polycyclic aromatic hydrocarbons (PAHs) analysis was carried out using a quick GC/MS method. The heavy metals were assessed using Atomic Absorption and the total phenols content were determined using spectrophotometer. The description of the data was made using Xlstat Version 2016.02.27444. **Results:** The study revealed four ARs in smoked chicken meat and ciprofloxacin was the most prevalent in the samples (100%) however their contents were within maximum residues limit (MRL). Regarding PAHs, the MRL in smoked meat products (2.92 ± 1.67 µk kg^{-1}) exceeded in about 56% of samples. Lead was present in all samples and their contents were (0.15 ± 0.17 mg kg^{-1}) far beyond MRL. Cadmium was found in 56.25% of samples and their contents (0.007 ± 0.002 mg kg^{-1}) were within MRL and total phenols ranged from 1.24-6.06 mg kg^{-1}. **Conclusion:** Consumption of traditionally smoked chicken meat sold in Lomé is not safe with respect to heavy metals and PAHs in particular and poses a potential health risk to the local consumers.

Key words: Traditionally smoked chicken, heavy metals, antibiotics residues, phenol, polycyclic aromatic hydrocarbons

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Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Meat and poultry are foods that contain important nutrients such as proteins of high- biological-value, B vitamins, minerals and trace elements and other bioactive compounds which are globally accepted[1,2]. Previous studies have reported that poultry meat is exposed to several chemical contaminants, particularly during the feeding, transportation, processing and at retail stages[1-3]. Codex Alimentarius Commission[4] defines "contaminant" as any substance not intentionally added to a food, which is present in food as a result of the production, manufacturing, processing, preparation, treatment, packaging, transportation, storage or environmental contamination. The term does not include insect fragments, rodent hairs, and other extraneous matter". Poultry meat may be contaminated with veterinary drugs, residues of environmental contaminants and substances generated during meat processing[1-3]. Antimicrobials are widely used for the prevention and treatment of disease, they maintain the health of all treated poultry, induce growth, improve meat quality and reduce production costs[2,3]. Application of veterinary drugs could result in deposition of antimicrobial residues in the final product[5]. Their presence is mainly due to non-compliance of the recommended withdrawal period before slaughtering of the treated animals and/or consuming products made from these animals[5,6]. These residues can detrimentally affect human health. These include immunopathological effects, carcinogenicity, mutagenicity, nephropathy, hepatotoxicity, reproductive disorders, bone marrow toxicity, hypersensitivity and resistance to antibiotic[5,7]. Environmental contamination is quite widespread in the world, and globalization makes it even more difficult to control. The reasons for the presence of environmental contamination in poultry meat are varied: use of contaminated ingredients in feed, lack of control of feed ingredients, improper processing, molds growth in feed grains and flours. These contaminants in meats are difficult to control because of the different routes of absorption for the animal and the diversity of the compounds to be analyzed, although the contaminants can exert toxicity in the final product[1]. Some of these substances can accumulate in the animals and human body, especially in fatty tissue with long-term harmful effects. Cadmium as a contaminant, has a negative effect on the renal, pulmonary, cardiovascular and skeletal systems and lead can damage the kidneys and the human reproductive and immune systems[1-3,8]. Among the substances generated during meat processing, polycyclic aromatic hydrocarbons (PAHs) and phenols are the main harmful compounds which contaminate smoked products during smoking process[1,9]. PAHs are generated by incomplete combustion of wood especially within a temperature range of 500-700°C with limited oxygen supply and their carcinogenic, mutagenic and bio-accumulative capacities have been established[1,10-12]. The European Union's Scientific Committee for Food assessed 33 PAHs in 2002 and identified 15 with genotoxic and carcinogenic properties as high priority. The determination of all PAHs is quite complex and the committee proposed benzo-a-pyrene (BaP) and a combination of four PAH [BaP, benzo(a) anthracene (BaA), chrysene (CHR), benzo (b) fluoranthene (BbF)] for monitoring and assessment of PAH contamination in foodstuffs[13,14]. Phenols are associated with almost all the desired technological effects of smoking (coloring, preservation and taste formation) but concerns have been reported about health hazards (carcinogenic or cocarcinogenic properties) of some of its compounds[1,9,15].

Smoking is one of the chicken processing techniques in Togo using mainly imported chicken[16] and therefore subject to these same types of contamination. While heat treatments during processing can remove and inactivate pathogenic microorganisms from food, they have limited effects on chemical contaminants[17-20]. Therefore, the constant monitoring of these contaminants in ready-to-eat foods of animal origin should be an ongoing concern. West African countries like Togo are poor in legislation to regulate these contaminants in foodstuffs[6].

The objective of this study was to assess the cadmium and lead, antibiotic residues, total phenols and PAHs contents in traditionally smoked chicken meat retailed to consumers in Lomé and their public health concerns.

MATERIALS AND METHODS

Samples collection: A total of 32 smoked chicken samples were collected from the main open markets in the city of Lomé. They were wrapped in aluminum foil to avoid photo degradation of some components. In the laboratory, samples were skinned and completely homogenized in a blender and stored in a freezer at -20°C prior to analysis.

Total phenols: Total phenol content was determined according to AFNOR[21]. Briefly, phenolic compounds from samples were extracted into ethanol. A colored complex of phenols and 4-aminoantipyrine formed in the presence of potassium ferricyanide and was separated using chloroform. Absorbance and concentrations were measured at 455 nm using the UV-1280 Spectrophotometer (Shimadzu Corporation, Japan).

Heavy metals: The cadmium (Cd) and lead (Pb) contents were determined on ashes after dry calcination of sample at à 550±50°C and diluted to 1% nitric acid (HNO^3), (NF EN 14082:2003). The concentrations were then measured using graphite furnace atomic absorption spectrophotometer with palladium or orthophosphoric acid matrix modifier (Agilent 200 Series AA/ GTA 120; Germany).

Polycyclic aromatic hydrocarbons (PAHs): All reagents and solvents were HPLC grade. Acetonitrile (ACN) was from Honeywell (Muskegon, MI, USA) and acetone was from VWR International (WestChester, PA, USA). The standard PAHs used were obtained from Dr. Ehrenstorfer GmbH, Germany. The samples were extracted and cleaned up and the PAH fractions were analyzed according to Smith and Lynam[22] using the Agilent Bond Elut QuEChERS dSPE (Enhanced Lipid Matrix Removal) for the extraction of the samples. The 18 targeted PAHs studied were: Naphthalene (NAP), Acenaphthylene (ACE), Acenaphthene (ACE), Fluorene (FLE), Anthracene (ANT), Phenanthrene (PHE), Fluoranthene (FLA), Pyrene (PyR), benzo(a)anthracene (BaA), Chrysene (CHR), Benzo(a)pyrene (BaP), Benzo(b)fluoranthene (BbF), Benzo(k)fluoranthene (BkF), Indeno(1,2,3-cd)pyrene(IcP), Dibenzo(a,h) anthracene (DhA), Benzo(g,h,i)perylene (BgP), Benzo(e)pyrene (BeP), Pyrelene (PyL).

GC conditions: A 7890B GC system (Agilent Technologies, USA) equipped with a 7693 autosampler (Agilent Technologies, USA) and connected to a 7000C triple Quad System mass spectrometer (Agilent Technologies, USA) was used for analysis of PAHs. Chromatographic separation was achieved with a fused silica capillary column (0.7 m×150 µm×0 µm) and the GC was also fitted with a pressure-controlled tee (PCT) post-column for automatic back flush. Helium (99.999%) was used as the carrier gas with a constant flow rate of 1.2 mL min^{-1}. The oven temperature was programmed as follows: initial temperature at 70°C held for 2 min, increased from 25°C min^{-1} to 150°C, then increased by 3°C min^{-1} at 200°C, 8°C min^{-1} at 280°C maintained for 12 min. The injection temperature was set at 325°C with a pulsed splitless injection at a volume of 5 µL. The calibration curves were obtained by plotting the response factor as a function of the analyte concentration (0.5; 1.5; 10 and 50 µg mL^{-1}). All calibration curves showed excellent linearity with R^2 >0.99 for all compounds. Agilent Mass Hunter software was used to quantify the different PAHs.

Antibiotics residues: All reagents and solvents were HPLC or analytical grade. Acetonitrile (ACN) was from Honeywell (Muskegon, MI, USA). The veterinary drug standards were from Dr. Ehrenstorfer GmbH (Augsburg, Germany). Reagent-grade formic acid (FA) was from BDH Laboratory Supplies (England). Ammonium acetate (NH4OAc) was from Fisher Chemicals (Fair Lawn, NJ, USA).

The antibiotics investigated were Amprolium Hydrochloride, Danofloxacin Mesylate, Metronidazole, Sulfadiazine, Oxytetracycline Hydrochloride, Chlortetracycline Hydrochloride, Doxycycline Hyclate and Ciprofloxacin Hydrochloride. The smoked chicken were extracted, cleaned up and analyzed according to Zhao and Lucas[23].

UHPLC conditions: The separation was carried out using a model 1290 UHPLC system, consisting of a binary pump, an autosampler and a thermostated column compartment (Agilent Technologies, Santa Clara, CA, USA), equipped with a 150×2.1 mm Poroshell 120 EC-C18 column, 2.7 µm particle size and a 5×2.1 mm Poroshell 120 guard column, 2.7 µm particle size (Agilent, Newport, DE, USA). The UHPLC system was coupled to a model 6490 triple quadrupole mass spectrometer system (Agilent Technologies, Santa Clara, CA, USA) equipped with Jet Stream electrospray and iFunnel technology. MassHunter workstation software was used for data acquisition and analysis. Chromatography separation was based on the following settings. Gradient at 0.2 mL min^{-1} from 10% solvent B for 0-0.5 min, then 100% solvent B by 8 min and hold 100% solvent B until 12 min, followed a post column equilibrium time for 3 min. The total cycle time was 15 min for each injection. The column was maintained at 40°C and the autosampler was at 4°C. The injection volume was 6 µL. Needle wash was used to flush the injection needle and valve. The MS system was in positive and negative ion mode using the following parameters: gas temperature: 300°C; gas flow: 13 L min^{-1}; capillary voltage: 3000 V; nebulizer pressure: 40 psi; gas heating duct: 400°C; sheath gas flow rate: 12 L min^{-1}; nozzle voltage: 0 V for both positive and negative ion mode. The iFunnel parameters were: 90 V high pressure RF for positive and negative ion mode and low-pressure RF of 70 V for positive and 60 V for negative ion mode.

The calibration standards prepared above corresponded to 5, 10, 20, 50, 100, 200 ng g^{-1} in the matrix blank samples and the calibration curves were obtained by plotting the response factor as a function of the analyte concentration. Calibration curves for all analytes were linear within the range given with correlation coefficients at least 0.99.

Data analysis: The description of data was carried out using Xlstat version 2015.6.08 (Addinsoft, Paris, France). Descriptive

statistics were calculated and the results were expressed in micrograms per kilogram (μg kg^{-1}) wet weight for PAHs and antibiotics residues and milligrams per kilogram (mg kg^{-1}) wet weight for total phenols and heavy metals. The values were compared to Maximum Residue Limit (MRL) if necessary (Compliance tests-comparison to a theoretical value), at $p>0.05$.

RESULTS

Total phenols and PAH fractions in traditionally smoked chickens: The phenolic compounds of the smoked chicken ranged from 1.24-6.06 mg kg^{-1} with an average of 2.92±1.67 mg kg^{-1} (Table 1). Analysis of all the PAHs showed that five fractions (NAP, ANT, PyR, BaA, BaP) were the most abundant in the samples (100%, n = 32), followed by ACA, ACE, FLU, FLT, CHR, BeP and IcP (87.5%, n = 28); PyL was present in 78.12% of the sample (n = 25); DaA was present in 56.25% of the sample (n = 18); BkF was 34.37% (n = 11); BgP and BbF was 2.5% (n = 4) of the sample. No trace of PHE was found in the samples. The simultaneous presence of BaP and CHR (HAP2) was detected in 87.5% (n = 28), HAP4 (BaP, CHR, BbF, BaA) and HAP8 (BaP, CHR, BbF, BaA, BkF, BgF, DaA, IcP) was found in 12.5% of the samples (Table 1).

NAP was not only the most common PAHs (100%) in smoked chicken but its content was also the highest (40.74 μg kg^{-1}) while the CHR content (10.62 μg kg^{-1}) was the highest in the PAH8 group (Table 1). Average content of NAP and CHR was 14.68±13.22 μg kg^{-1} and 2.53±1.18 μg kg^{-1} respectively. For all samples analyzed, the BaP content ranged from 0.92-4.99 μg kg^{-1} with an average of 2.53±1.18 μg kg^{-1}. Compliance tests did not reveal any similarity of the samples according to the regulations ($p>0.05$). Of the 32 samples analyzed, 18 (56.25%) had an exceeding level of the MRL for BaP (2 μg kg^{-1}). The BaA and BbF contents varied from 2.05-8.20 μg kg^{-1} with an average of 4.40±2.19 μg kg^{-1} and not detected to 1.56 μg kg^{-1} with an average of 0.17±0.49 μg kg^{-1}. The maximum level of PAH4 was 25.36 μg kg^{-1} with a mean value of 12.58±6.71 μg kg^{-1} and there was no significant difference ($p>0.05$) in the MRL for HAP4 (12 μg kg^{-1}). The number of samples with PAH4 levels above the regulatory limit was 14 (43.75%). The ACA, BkF, DaA, BgP, Icp, ACE, FLU, FLT, BeP and PyL fractions varied from non-detected to 15.06, 2.89, 0.81, 3.24, 1.55, 38.46, 30.92, 29.59, 13.93, and 7.15 μg kg^{-1} respectively. The ANT ranged from 0.98-18.33 μg kg^{-1} and PyR ranged from 0.89-22.69 μg kg^{-1} (Table 1). A significant correlation between BaP and others PAH8 and PAH18 ($|r|>0.65$; $p<0.05$) was observed (67%) (Table 2). The correlation coefficients between BaP and PAH2, PAH4 and PAH8 were 0.90, 0.88 and 0.88, respectively while those between PAH2 and PAH4, PAH8 were 0.99 and those between PAH4 and PAH8 were approximately 1.

Table 1: Totals phenols and heavy metals content in smoked chicken (mg kg^{-1})

	Range	Mean±SD	MRL
Total phenols	1.24-6.10	2.32±1.30	-
Lead (Pb)	0.03-0.63	0.15±0.17	0.10
Cadmium (Cd)	6.10^{-5}-0.10	0.007±0.002	0.05

Heavy metals traces and antibiotics residues in smoked chicken: The average concentration of smoked chicken samples for cadmium was significantly lower ($p>0.05$) than its MRL while lead content was above its MRLs (0.05 mg kg^{-1} and 0.1 for Cd and Pb respectively). The lead contents ranged from 0.04-0.63 mg kg^{-1} with an average of 0.15±0.17 mg kg^{-1} and Cd contents ranged from non-detected to 0.01 mg kg^{-1} with an average of 0.007±0.002 mg kg^{-1} (Table 3).

The study revealed that out of the eight antibiotics four were found in the sample of smoked chicken; four antibiotics were not found at the detectable limit. These were Amprolium, Danofloxacin Mesylate, Metronidazole and Sulfadiazine. The four antibiotics found in the samples belong to two groups of antibiotics: Fluoroquinolones (Ciprofloxacin) and Tetracycline (Oxytetracycline, Chlortetracycline and Doxycycline). Ciprofloxacin was the most prevalent in the samples (100%) and its content ranged from 3.05-7.17 μg kg^{-1} with an average of 4.85±1.36 μg kg^{-1}. Oxytetracycline was found in 34.37% of the samples. Its content ranged from not detected to 13.28 μg kg^{-1} and the average was 8.48±3.39 μg kg^{-1}. Doxycycline and chlortetracycline were found in almost 22% of the samples. Their contents ranged from not detected to 18.43 μg kg^{-1}, with a mean value of 11.60±6.83 μg kg^{-1} and from not detected to 21.41 μg kg^{-1} with a mean value of 17.33±4.08 μg kg^{-1} respectively (Table 4). The results showed that the levels of the different antibiotics residues recorded were lower than ($p>0.05$) their MRLs .

DISCUSSION

Since there are no national regulations regarding contaminants in foodstuffs in Togo, regulations set by the European Union were used in this study[24-26]. Phenols are the main compounds associated with the desired technological effects (coloring, preservation and taste formation) of smoking[9], they are referred to as indicator of the degree of smoking[27]. The low levels recorded in this study (maximum of 6.1 mg kg^{-1}) showed that chicken were not heavily smoked

Table 2: Hydrocarbons polycyclic aromatics analysis of the smoked chicken (μg kg^{-1})

PAHs	Effective (%)	Range	Mean±SD	LMR (μg kg^{-1})	Samples >LMR (%)
Naphthalene	100 (n = 32)	0.75-40.74	14.67±13.22	-	-
Acenaphthylene	87.5 (n = 28)	0-38.46	15.78±11.73	-	-
Acenaphthene	87.5 (n = 28)	0-15.06	6.13±4.85	-	-
Fluorene	87.5 (n = 28)	0-30.92	13.10±10.12	-	-
Anthracene	100 (n = 32)	0.98-18.33	10.24±4.51	-	-
Phenanthrene	ND	-	-	-	-
Fluoranthene	87.5 (n = 28)	0-20.59	10.87±5.80	-	-
Pyrene	100 (n = 32)	0.89-22.69	12.15±6.12	-	-
Benzo(a)anthracene	100 (n = 32)	2.05-8.2	10.80±2.53	2	56.25
Chrysene	87.5 (n = 28)	0-10.62	5.47±3.26	-	-
Benzo(a)pyrene	100 (n = 32)	0.92-4.99	2.52±1.18	-	-
Benzo(b)fluoranthene	12.5 (n = 4)	0-1.56	0.17±0.49	-	-
Benzo(e)pyrene	87.5 (n = 28)	0-13.93	3.80±4.84	-	-
Pyrelene	78.12 (25)	0-7.15	1.95±2.49	-	-
Benzo(k)fluoranthene	34.37 (n = 11)	0-2.89	1.55±0.88	-	-
Indeno(1.2.3-c.d)pyrene	87.5 (n = 28)	0-1.55	0.79±0.51	-	-
Dibenzo (a.h) anthracene	56.25 (n = 18)	0-0.81	0.24±0.340	-	-
Benzo(g.h.i)pyrelene	12.5 (n = 4)	0-3.24	1.56±0.92	-	-
HAP2	87.5 (n = 28)	0.92-15.61	8.00±4.28	-	-
HAP4	12.5 (n = 4)	2.97-25.36	12.57±6.71	12	43.75
HAP8	12.5 (n = 4)	2.97-30.66	14.83±8.15	-	-
HAP18	-	12.45-286.2	135.83±80.32	-	-

MRL: Maximum residue limit, Samples >LMR: number of samples exceeding the MRL, ND: Not detected, HAP2: (BaP, CHR), HAP4: (BaP, CHR, BbF, BaA), HAP8: (BaP, CHR, BbF, BaA, BkF, BgP, DhA, IcP). BaP: benzo-a-pyrene, BaA: benzo(a)anthracene, CHR: chrysene, BbF: Benzo (b) fluoranthene, BkF: Benzo(k)fluoranthene, BgP: Benzo (g,h,i) perylene, DhA: Dibenzo (a,h) anthracene, IcP: Indeno (1,2,3-cd) pyrene

Table 3: Correlational analysis of PAHs contents in the smoked chicken

PAHs	BaA	CHR	BaP	BbF	BkF	IcP	DaA	BgP	HAP2	HAP4	HAP8	HAP18
BaA	1.00											
CHR	0.97	1.00										
BaP	0.79	0.82	1.00									
BbF	0.61	0.56	0.74	1.00								
BkF	0.76	0.76	0.58	0.37	1.00							
IcP	0.79	0.86	0.67	0.64	0.72	1.00						
DaA	0.52	0.57	0.62	0.27	0.66	0.37	1.00					
BgP	0.61	0.56	0.74	1.00	0.37	0.64	0.27	1.00				
HAP2	0.95	0.99	0.90	0.63	0.74	0.84	0.61	0.63	1.00			
HAP4	0.98	0.98	0.88	0.67	0.74	0.84	0.58	0.67	0.99	1.00		
HAP8	0.97	0.98	0.88	0.69	0.78	0.87	0.59	0.69	0.99	1.00	1.00	
HAP18	0.94	0.96	0.90	0.66	0.66	0.82	0.54	0.66	0.98	0.98	0.97	1.00

Bold values are significant (|r>| 0.65; $p < 0.05$)

Table 4: Antibiotics residues content in smoked chicken

		Content (μg kg^{-1})		
Analyte	Effective (%)	Range	Mean±SD	MRL
Doxycycline	21.87 (n = 7)	ND-18.43	2.58±5.80	100
Chlortetracycline	21.87 (n = 7)	ND-21.41	3.85±7.46	100
Oxytetracycline	34.37 (n = 11)	ND-13.28	2.83±4.45	100
Ciprofloxacin	100 (n = 32)	3.05-7.17	4.85±1.36	100

and this confirms the relatively short duration reported by Akakpo *et al.*[16] in the traditional processing of smoked chicken in Lomé. Different levels of phenols have been reported in several types of African smoked meats. Alonge[28] found phenol levels ranging from 5-137 mg kg^{-1} with an average of 88 mg kg^{-1} in smoked meat, while Ratsimba *et al.*[29] found 28±18 mg kg^{-1} and Poligné[30] reported a level of 29.5±0.3 mg kg^{-1}. Elsewhere, varying concentrations of phenolic were also reported[15]. The variability in the reported contents is due to the rate of smoke deposition and penetration which depends on several factors including temperature, humidity, volatility and smoke velocity[12,31,32]. For phenolic compounds, no MRLs have been fixed by the European commission[26] in foods probably due to their ability to be excreted from the body through the urine within a few days of ingestion. However, Agency for Toxic Substances and Disease Registry[15] reported that in USA, a minimal lethal oral dose for adults is approximately 70 mg kg^{-1}.

The suitability of BaP as a biomarker of carcinogenic PAHs was checked (Table 2). The results showed that PAH2 and PAH4 was found to be correlated with PAH8 and all of the PAHs analyzed. In this study, PAH2 and/or PAH4 would be the most appropriate indicator to assess the presence of genotoxic and carcinogenic PAHs in the smoked chicken

samples[24]. The MRL for smoked meat and meat products fixed by the European Commission is 2 μg kg^{-1} for BaP and 12 μg kg^{-1} for PAH4[13,24]. Despite these restrictions, as in the case of this study, high levels of BaP are still found in smoked meat products around the world. In Africa, (Nigeria) Alonge[33] reported that BaP levels ranged from 10.5-66.9 μg kg^{-1} in traditionally smoked meat, while Akpambang *et al.*[34] reported BaP levels of around 10.1 μg kg^{-1} and more recently Adeyeye[35] reported maximum average levels of BaP (6.81±0.24 μg kg^{-1}) in Suya (a spicy grilled beef) usually grilled over a wood fire. In a similar product, Coulibaly[36] obtained a BaP content ranged from 5.73-12.35 μg kg^{-1}. Ratsimba *et al.*[29] reported high level of BaP content (59 μg kg^{-1}) in a traditional Malagascan meat product. Widely varying levels of BaP have also been reported by Ledesma *et al.*[37]. The results showed that NAP was the most prevalent and abundant PAHs in smoked chicken (100% of samples, 40.74 μg kg^{-1}). This is not in line with the European Food Safety Agency[11] which indicated that chrysene was the most prevalent and abundant in food contaminated by PAH. According to Alonge[33], the NAP levels ranged from 2.2-30.8 μg kg^{-1} and Adeyeye[35] reported that the NAP levels was 4.01±0.18 μg kg^{-1} while Coulibaly *et al.*[36] reported much higher levels (9810.924 μg kg^{-1}). In general, there is evidence that the contamination of meat with PAH during smoking is influenced by several factors. These factors include the smoke generation conditions such as the temperature, the nature of the wood, the oxygen control and the smoke removal procedures applied immediately after smoke generation[11,12,37-39]. The rate and extent of smoke deposition also depend on the reactivity properties of the surface and deep layers of the smoking material[12,40]. Finally, the Codex Alimentarius Commission[41] in the "Code of Practice for the Reduction of Contamination of Foods Containing PAHs from Direct Smoking and Drying Processes" (CAC/RCP 68/2009) states that the contamination of foods by PAH should be minimized by controlling 10 variables which are fuel type, smoking method (direct or indirect), smoke generation process, distance/position between food and heat source, fat content of foods, smoking time, temperature during smoking, cleanliness/maintenance of equipment and design of smoking chamber and equipment used for air-smoke mixing. The relevance of BaP as a biomarker of possible and probable carcinogenic PAHs was also verified between the contents of BaP and PAH8 (Table 2). PAH2 and PAH4 were correlated with all the PAHs analyzed therefore PAH2 and/or PAH4 would be the most appropriate indicator to assess the presence of genotoxic and carcinogenic PAHs in the smoked chicken samples analyzed in this study[24].

Lead levels in all smoked chicken were above the MRL set by the European commission[25]. Over time these levels of lead may induce bioaccumulation in the tissues following constant consumption and causes toxicity complications for consumers[1-3,8]. The high levels of metals in poultry products originate mainly from contaminated feeds, water and soil[1,2]. Compared to previous studies, the Pb and Cd contents recorded in this study were substantially low. Frederick *et al.*[42] reported levels of Pb and Cd ranged from 0.41-0.70 mg kg^{-1} in raw and grilled guinea fowl meat in Tamale Metropolis, Ghana. According to Kayode *et al.*[43] the concentration of Pb was 13.2±0.1 mg kg^{-1} and 0.8±0.2 mg kg^{-1} for Cd in chicken meat imported from Nigeria and Iwegbue *et al.*[44] recorded the range of Pb from 0.01-4.60 and Cd from 0.01-1.27 mg kg^{-1} in Nigerian chicken meat. Variable contents were recorded by Ogu and Akinnibosun[45] worldwide. Differences in levels of heavy metal contamination in meat are related to variations in exposure levels and their concentration in animal tissues[42,44]. According to Panisset *et al.*[46] there is no threshold value for the level of lead below which it would not have a toxic effect. Therefore, all measures must be taken to reduce their contamination in the environment[46].

The presence of the two antibiotics (ciprofloxacin and tetracycline) in smoked chicken samples can be explained by their extensive use in veterinary medicine and several authors have already reported that they belong to the main classes of antibiotics used in animal husbandry[5,6,47]. Ciprofloxacin was the most prevalent in smoked chicken. This result is consistent with a previous study conducted by Omotoso and Omojola[48] who found that ciprofloxacin was the most abundant antibiotic residues in imported frozen chickens in Nigeria. But, according to Darwish *et al.*[5], tetracyclines were the most predominantly prescribed antibiotics and of all antibiotic-associated residues they represent 41% of cases, followed by 18% β-lactams in locally produced animal foods in several African countries. These results highlight the harmful effects of antibiotic used in animal products, in particular microbial resistance even far from the place of use. The contents of ciprofloxacin observed in this study are comparable to those reported by Sahu and Saxena[47] who found the concentration of ciprofloxacin (approximately 6.03 μg kg^{-1}) in chicken muscle. However, these values are lower than those reported by Ahmed and Gareib[49] who found the concentration of ciprofloxacin from not detected to 90 μg kg^{-1} for chicken breast and not detected to 100 μg kg^{-1} for ready-to-eat chicken luncheon. The levels of oxytetracycline residues reported in previous studies ranged from 156-900 μg kg^{-1} in a sample of raw broiler fillet[50]; 110-1089 μg kg^{-1} in

chicken breast[51]; 8.25-15.16 μg kg^{-1} in chicken meat[47] and 70 μg kg^{-1} in chicken muscle[52]. Hussein *et al.*[53] did not detect oxytetracycline residues in chicken luncheon. Quite variable levels of chlortetracycline and doxycycline residues have also been reported[47,51,52]. The variability of these drug residues recorded in different studies is generally attributed to a various reasons: drug abuse, non-compliance with withdrawal periods before slaughter of animals, lack of understanding of drug use also contribute to the food contamination[5,6,54]. The low levels of the antibiotic residues recorded in this study (Table 5) could be attributed to the effect of heat treatment applied to the chicken during processing. Indeed, many authors have indicated that sufficient temperature, heating conditions, cooking method and time can reduce some antibiotics residues[17,19,53,55] but this generally does not provide an additional safety margin for consumers[53]. In previous research Hussein and Khalil[50] have pointed out that regardless of the reduction percentages in antibiotic residues, the product is not safe for humans because antibiotic may discharge harmful metabolites. Therefore heat treatment could not be an alternative to control the use of antibiotic. Only the applications of strict measures to keep flocks on the farm until the withdrawal period has elapsed and the prevention of the misuse of antibiotics in poultry farms could solve the problem of human exposure to residues of antibiotic.

CONCLUSION

The present study highlighted a potential risk to the human health in shape of chemical contaminants present in smoked chicken consumed in Lomé (Togo). The lead concentrations were above the maximum limit while the residues of cadmium and antibiotics were acceptable according to their MLRs. Veterinary authorities should control the use of antibiotics in poultry farms and limit their use. Regulations on the use of antibiotics should be followed to ensure appropriate withdrawal periods before slaughter and marketing. The use of alternatives to antibiotics, such as antimicrobial and plant-based probiotics, can be a great option. Regarding imported poultry meat, a control policy should be implemented to ensure the compliance of international standards for antibiotics residues and heavy metals in food. The smoked chicken also contained relatively low level of phenolic compounds and varying levels of PAHs. Although not all samples have concentration levels above the regulatory limit, they still pose a risk to the health of consumers. This contamination can be controlled and minimized through the use of appropriate equipment and the selected fuels for smoking.

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