International Journal of Food Studies | IJFS April 2023 | Volume 12 | pages 84–94

Evaluation of the Formation of Polycyclic Aromatic Hydrocarbons (PAHs) in Chicken Cooked by Saudi Traditional Methods and Their Dietary Risk Assessment

Abdullah A. Al Sayari[a], Mohammed A. Almutairi[a], Turki S. Alsaleem[a], Turki Abu Hamrh[a], Naseer Al Thenyian[a], Abdullah I. Al Tamim[a], and Abdullah M. Alowaifeer[a*]

[a] Reference Laboratory for Food Chemistry, Saudi Food & Drug Authority (SFDA), Riyadh 11561, Saudi Arabial
[*]Corresponding author
vet.abdullah@gmail.com

Received: 9 March 2022; Published online: 18 April 2023

Abstract

This study evaluates the formation of 13 polycyclic aromatic hydrocarbons (PAHs) in chicken thighs and breasts, cooked by different methods. These methods are: madhbi, charcoal mandi, electric oven mandi, gas flame oven mandi and shawaya. Chicken samples were collected from a restaurant in Riyadh, Saudi Arabia. Analysis of the samples was carried out using high-performance liquid chromatography with a fluorescence detector (HPLC-FLD). The data obtained showed that madhbi chicken had higher PAHs levels than other cooking styles, with the mean concentration in chicken breast of 87.72 μg/kg and thigh of 75.56 μg/kg. Phenanthrene was the compound detected at the highest concentration in different cooking methods. There was no significant difference in concentration of PAHs between the parts of chicken cooked with the same method. However, the method of cooking had a significant impact on the formation of PHAs. Therefore, the formation of PAHs in chicken meat could be reduced by choosing appropriate cooking methods. Moreover, the margin of exposure was used to assess the health risk in adults due to madhbi chicken ingestion. The results showed that there is no serious health concern.

Keywords: PAHs; HPLC; Chicken; Risk Assessment; Margin of Exposure

1 Introduction

Polycyclic aromatic hydrocarbons (PAHs) are organic compounds comprised of carbon and hydrogen, forming two or more aromatic joint rings (Codex Alimentarius Commission, 2009). The formation of PAHs occurs naturally or anthropogenically from the combustion of natural matter, therefore, they are widely dispersed throughout the globe. Industrial activities, wildfires and volcanic eruptions are among the most common PAH sources in the environment (Hokkanen et al., 2018). There are more than 100 different PAHs and, because of their chemical structures, these chemicals tend to become highly lipophilic and stable in the environment (Falcó et al., 2003; Tang et al., 2005).

Human exposure to PAHs occurs through inhalation, dermal contact or the consumption of contaminated foods, which accounts for 88–98% of exposure (Farhadian et al., 2011). Manufacturing processes or cooking methods are responsible for the presence of PAHs in food (Rose et al., 2015), which accumulates by direct

 10.7455/ijfs/12.1.2023.a6

deposition from the smoke produced by thermal treatment. The pathways of the formation of PAHs in processed food are not well-known. There are at least three possible ways that might lead to the formation of PAHs in meat. The first pathway is by pyrolysis of organic matters such as carbohydrates, proteins and particularly fats at a temperature above 200 ^{o}C. The second mechanism is by the dripping of fat over the heat source, which generates PAHs deposited on the meat (Alomirah et al., 2011; Lee et al., 2016). The incomplete combustion of charcoal is the third pathway, which can form PAHs that adhere to the surface of the food (Alomirah et al., 2011; Chen & Lin, 1997). It is known that PAHs with two or three rings have greater volatility than other PAHs (Szopińska et al., 2019).

Many factors influence the generation of PAHs in food, such as heat source and distance from it. Onwukeme et al. (2015)and Babić et al. (2017) documented 11 parameters that affected the composition and number of PAHs in smoked fish. These were source of heat, distance from heat source, type of wood, moisture content, oxygen accessibility, the temperature of smoke generation, cooking duration, natural content of PAHs in raw meat, water activity of food, fat content and the design of the food device.

The European Union (EU) and the Environmental Protection Agency (EPA) both included PAHs in their priority pollutant lists due to their carcinogenicity (Farhadian et al., 2011). The carcinogenic capacity of PAHs depends on the number of aromatic rings. According to the International Agency for Research on Cancer (IARC), compounds with four to six combined rings are considered class 1 carcinogens (Hokkanen et al., 2018) (Table 1), while the others are classified as either 2A, 2B or 3. PAHs are known to have a strong affinity to nucleic acid (DNA), which metabolically convert diolepoxides that lead to replication errors (Farhadian et al., 2011).

The Scientific Committee on Food (SCF) advised the monitoring of 15 PAHs in food (benz(a)anthracene, benzo(b)fluoranthene, benzo(j)fluoranthene, benzo(k)fluoranthene, benzo(ghi)perylene, benzo(a)pyrene, chrysene, cyclopenta(cd)pyrene, dibenzo(a,h)anthracene, dibenzo(a,e)pyrene, dibenzo(a,h)pyrene, dibenzo(a,i)pyrene, dibenzo(a,l)pyrene, indeno(1,2,3-cd)pyrene and 5-methylchrysene) because they have shown clear carcinogenic effects in experimental animals (Zelinkova & Wenzl, 2015). However, others have suggested the measurement of benzo(a)pyrene as a marker for the occurrence of carcinogenic PAHs (Zelinkova & Wenzl, 2015). While the European Food Safety Authority (EFSA) considered PAH4 (sum of four different polycyclic aromatic hydrocarbons, named benzo[a]anthracene, chrysene, benzo[b]fluoranthene and benzo[a]pyrene) as an appropriate indicator (Lee et al., 2016). Based on the Commission Regulation of the EU, the maximum levels (MLs) of benzo(a)pyrene and PAH4 in smoked meat are 2 and 12 μg/kg, respectively (EU EC No 835/2011).

Most PAH investigations in the literature were conducted on smoked, grilled or fried meat but as far as we know no research has been carried out to evaluate the formation of PAHs by traditional Arabian cooking methods. Mandi, an Arabian steam cooking method that requires the chicken to be concealed with the heat source, is a very common cooking method in the Arab world. The heat source of mandi is usually generated by electricity, gas flame or charcoal. In this study, the formation of 13 PAHs in chicken cooked by mandi, shawayah (chicken grilled in gas or chicken rotisserie oven) and madhbi (chicken grilled on stones overlaid on charcoal) methods are assessed. The health risk assessment for adults exposed to PAHs from chicken consumption is also estimated.

2 Materials and Methods

2.1 Sampling and Sample Preparations

Five cooked chicken samples for each method (charcoal mandi, gas flame oven mandi, electric oven mandi, shawayah and madhbi) were collected from local restaurants around the city of Riyadh. Each cooked chicken sample was divided into two portions (breast and thigh). The samples were packed in aluminum foil and placed in polyethylene bags before they were trans-

Table 1: List of PAH compounds, showing which belong to PAH4 and PAH8, and their IRAC classification

PAH compounds	PAH 4	PAH 8	IRAC group
Fluorene			3
Phenanthrene			3
Anthracene			3
Fluoranthene			3
Pyrene			3
Benz(a)anthracene	✓	✓	2B
Chrysene	✓	✓	2B
Benzo(b)fluoranthene	✓	✓	2B
Benzo(k)fluoranthene		✓	2B
Benzo(a)pyrene	✓	✓	1
Dibenz(a,h)anthracene		✓	2A
Benz(g,h,i)perylene		✓	3
Indeno(1,2,3, -c, d)pyrene		✓	2B

IARC (International Agency for Research on Cancer) classification: group 1 = carcinogenic to humans, group 2A = probably carcinogenic to humans, group 2B = possibly carcinogenic to humans, group 3 = not classifiable as to carcinogenicity to humans.

ported to the lab (COMMISSION DIRECTIVE 2005/10/EC). The samples were stored at 4 ^{o}C until the day of analysis. Samples were homogenized in a Retsch GM 200 for 2-3 minutes.

2.2 Reagents and Chemicals

All solvents used in the extraction and analysis were HPLC grade. Acetonitrile was purchased from Merck (Darmstadt, Germany). Sodium Acetate with a purity of 99.99% was obtained from Merck (Darmstadt, Germany). Magnesium Sulphate, extra pure, was purchased from Scharlab (Barcelona, Spain). A PAH mixture analytical standard (PN 8500-6035) was obtained from Agilent (Foster City, CA, USA).

2.3 Sample extraction and clean-up

Sample extraction procedures were based on the method reported by Gratz et al. (2010). Five grams of homogenized sample was weighed into a 50 mL polypropylene tube. Then, 5 grams of water was added and the mixture shaken for one minute. A volume of 15ml of acetonitrile was then added to the mixture and the tube was shaken for one minute. Six grams of Magnesium Sulphate and 1.5 grams of Sodium Acetate were added to the mixture, followed by shaking for another minute. Then, the sample was centrifuged for 10 minutes at 3000 x g. The supernatant was filtered through a 0.2 μm PTFE syringe filter and transferred into the analysis vial.

2.4 Liquid Chromatography with FLD Analysis

Agilent 1200 Series liquid chromatography equipment, comprising autosampler, degasser, fluorescence detector, binary pump and column compartment, was used to analyze standards and samples. Chemstation software was used to control the operation of the equipment. PAHs were separated on a Zorbax Eclipse PAH analytical column (250mm × 4.6 mm, 5 μm particle size) at a 1.3 ml/min flow rate (Figure 1). Acetonitrile and water were used to make up the mobile

Table 2: HPLC-retention time, fluorescence detector parameters and LOQ values

PAH compounds	HPLC-retention time (min)	FLD-excitation (nm)	FLD-emission (nm)	FLD-PMT gain	LOQ (μg/kg)
Fluorene	17.685	272	335	14	1.08
Phenanthrene	19.84	248	380	12	5.87
Anthracene	22.127	248	380	12	2.15
Fluoranthene	24.516	280	470	14	8.27
Pyrene	26.158	270	385	14	2.13
Benz(a)anthracene	31.912	270	385	14	1.43
Chrysene	33.156	270	385	14	1.36
Benzo(b)fluoranthene	36.634	256	446	12	3.37
Benzo(k)fluoranthene	38.219	256	446	12	1.24
Benzo(a)pyrene	39.466	292	417	12	1.81
Dibenz(a,h)anthracene	42.205	292	417	12	4.61
Benz(g,h,i)perylene	43.728	292	417	12	5.43
Indeno(1,2,3, -c, d)pyrene	45.558	274	510	14	4.93

phase, which ran in a gradient condition. The column temperature was set at 25 ^{o}C, and the injection volume was 20 μL. The following parameters were used for the fluorescent detection: detection of multiwavelength emission (355, 380, 385, 417, 446, 470, 510 nm), excitation multiwavelength (248, 256, 270, 272, 274, 280, 292 nm) and general screening PMT gain of 12 and 14 (Table 2).

2.5 Quality control

Spiked samples were prepared and analyzed with each sample batch to ensure high accuracy and reliability. The recovery range of PAHs in the spiked samples was found to be between 70 - 120 %, and the expanded uncertainties of all measured compounds were $\leq$ 20%. For the calibration curves, the minimum acceptable correlation coefficient (r^2) was set at >0.9995 for a minimum of 3 levels. Limit of quantification (LOQ) was determined as 10 times the standard deviation of blank samples. Our laboratory is accredited under ISO 17025 for the determination of PAHs in chicken using HPLC-FLD.

2.6 Risk Assessment for dietary exposure to polycyclic aromatic hydrocarbons

Different PAH compounds have different abilities to generate a toxic effect. Therefore, toxic equivalency factors were used to calculate the toxicity equivalency quotient of benzo(a)pyrene (TEQ_{BaP}) to assess the carcinogenic risk. TEQ_{BaP} was calculated based on equation 1 (Food and Agriculture Organization of the United Nations & World Health Organization, 2006).

$$TEQBaP = \sum_{(i=1)}^{n} [Ci] \times TEFi \tag{1}$$

Where Ci is the concentration of individual PAH compounds in chicken and TEFi is the toxicity factor recommended by Nisbet and LaGoy (1992).
The chronic daily intake (CDI) was measured based on equation 2 (United States Environmental Protection Agency, 2001).

$$CDI(TEQBaP\mu g/kg/day) = \frac{Ci \times IRi \times ED}{BW \times AT} \tag{2}$$

Where C_i is the total TEQ level of PAH4 in the chicken samples (µg/kg) and IR_i is the average

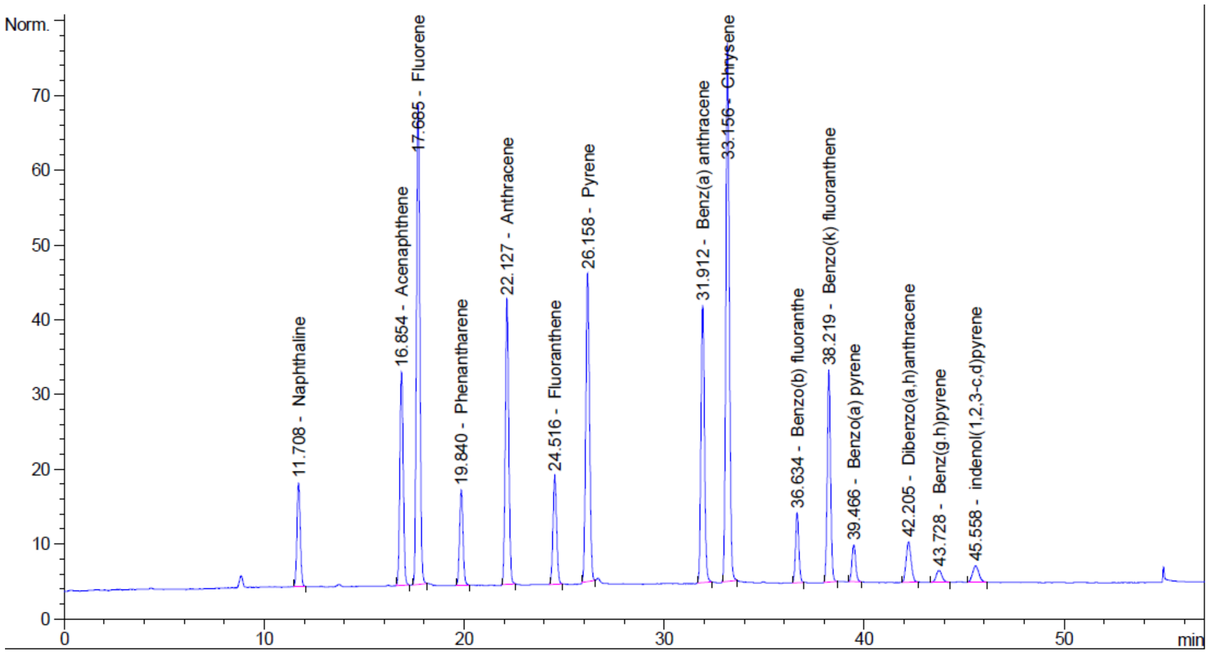

Figure 1: The chromatogram of separated PAHs by HPLC-FLD

daily intake of chicken (g/day). IR_i is estimated to be 109.6 g/day for Saudi Arabia's per capita poultry consumption in 2019 based on a report by the United States Department of Agriculture (USDA)/ Foreign Agriculture Service. BW stands for average body weight (70 kg), ED is exposure duration for adults (54 years) and AT is the average exposure time (365 day/ year ×75 years) (Almutairi et al., 2021).

The margin of exposure approach (MOE) is a tool used to assist risk assessors in evaluating the safety concern related to compounds present in food. This approach is followed when the compounds of interest are considered both genotoxic and carcinogenic (European Food Safety Authority, 2005). MOE was estimated based on equation 3, which is the ratio between benchmark dose lower confidence limit ($BMDL_{10}$) and chronic daily intake (CDI) (Food and Agriculture Organization of the United Nations & World Health Organization, 2009).

$$MOE = \frac{BMDL}{CDI} \tag{3}$$

Where the $BMDL_{10}$ value for PAH4 is 0.34 mg/kg bw per day (European Food Safety Authority, 2008). If the value of MOE is 10,000 or higher, this indicates a low concern for public health and a low priority for risk management actions (European Food Safety Authority, 2005).

In our study, Monte Carlo simulation was implemented to account for the uncertainty and variability of estimates (United States Environmental Protection Agency, 2001). Simulations were performed at 10,000 iterations, and health risk calculation values were reported at the 95% percentile. The analysis of variance (ANOVA), Monte Carlo simulation and other treatment of data were conducted using Microsoft Office Excel 2016.

3 Results

13 PAH compounds, namely, fluorene, phenanthrene, anthracene, fluoranthene, pyrene, benz(a)anthracene, chrysene, benzo(b)fluoranthene, benzo(k)fluoranthene, benzo(a)pyrene, dibenz(a,h)anthracene,

benz(g,h,i)perylene and indeno(1,2,3, -c, d)pyrene, were determined in chicken, which was cooked using different Arabian cooking methods (Table 3). The cooking methods were charcoal mandi, gas flame oven mandi, electric oven mandi, madhbi and shawayah. In the experiment, to confirm the absence of PAH compounds in uncooked chickens, four raw chicken meat samples were selected and analyzed. For descriptive analysis, a zero value was set for any compound below LOQ.

The research revealed that concentrations of PAH compounds in raw chicken samples were below LOQ, which proves absence of the targeted compounds. Fluorene and phenanthrene were found in both chicken parts (breast and thigh), which were cooked by the electric oven mandi method. The mean concentrations of phenanthrene in thigh and breast samples were 18 μg/kg and 17.23 μg/kg, respectively, with a detection rate of 100%. Moreover, fluorene was detected in breast and thigh samples with mean concentrations 5.22 μg/kg and 5.25 μg/kg, respectively.

Shawaya breast chicken meat contained phenanthrene (1.22 µg/kg) and anthracene (0.52 μg/kg). In Shawaya thigh meat, the same compounds were found but benz(a)anthracene was also detected in the thighs with a mean concentration of 0.72 μg/kg, with a detection rate of only 20%. Interestingly, in chicken cooked by gas flame oven, all targeted PAHs were below the LOQ.

The data also revealed that madhbi chicken samples were the most heavily loaded with the PAHs. In both chicken parts, phenanthrene had the highest concentration at 49.12 μg/kg in the breast and 41.81 μg/kg in the thighs. Fluorene, anthracene, fluoranthene and pyrene were also found in madhbi chicken samples, with a 100 % detection rate. The lowest concentration measured of chrysene was in madhbi breast chicken (1.05 μg/kg).

Phenanthrene, anthracene, pyrene and chrysene were found in charcoal mandi breast chicken, with mean concentrations of 8.54 µg/kg, 3.09 μg/kg, 0.77 μg/kg and 0.73 μg/kg, respectively. Also, the same compounds were detected in the thighs but with slight differences in the mean concentrations.

4 Discussion

4.1 The influence of the method of cooking on the formation of PAHs

The data obtained were evaluated by analysis of variance (ANOVA), and the difference was considered at $p \leq 0.05$. The impact of the cooking method on the formation of PAHs was investigated using different methods of cooking. The results showed that the method of cooking had a significant effect on the formation of PAHs (p-value ≤ 0.05). These findings agree with previous research conducted by Büyükkurt et al. (2020), which confirmed that food processing, food composition, type of heat source and contact with the heat source have a great effect on the concentration of PAHs formed in beef meat. Moreover, the conclusion of reviewing 7 studies showed that charcoal formed a significantly higher concentration of PAHs in cooked meat compared with gas (Ghorbani et al., 2020). In our study, madbhi chicken had the highest concentration of PAHs compared with other cooking methods. This may be attributed to the cooking style as madbhi chicken is usually cooked directly over hot stones, which leads to the increased formation of PAHs. Several research projects have been carried out on the formation of PAHs in foodstuff. Based on Alomirah et al. (2011), phenanthrene had the highest mean concentration (54.9 μg/kg) measured in various types of meat. Malarut and Vangnai (2018) detected 16 PAHs, ranging from 24.42 μg/kg to 34.07 μg/kg in sausages, which were smoked using different types of woodchips. Moreover, benzo(a)pyrene was evaluated in donor kebabs, which were cooked by different methods. The higher concentration of benzo(a)pyrene was found in doner kebabs cooked in a charcoal fire (24.2 μg/kg), compared to doner kebabs cooked in a gas fire (Terzi et al., 2008).

4.2 PAH formation in different chicken parts

PAH formation was examined in chicken thighs and breasts to determine the influence of chicken

Table 3: Mean concentration (μg/kg) of 13 PAHs in cooked chicken samples

PAH compoumds	Charcoal Mandi		Madhbi		Shawaya		Electric oven Mandi		Gas flame oven Mandi	
	Breast ± U n=5	Thigh ± U n=5	Breast ± U n=5	Thigh ± U n=5	Breast ± U n=5	Thigh ± U n=5	Breast ± U n=5	Thigh ± U n=5	Breast ± U n=5	Thigh ± U n=5
Fluorene	<LOQ	<LOQ	6.39 ± 0.72	6.32 ± 0.71	<LOQ	<LOQ	5.22 ± 0.59	5.25 ± 0.59	<LOQ	<LOQ
Phenanthrene	8.54 ± 0.99	8.10 ± 0.94	49.12 ± 5.67	41.81 ± 4.83	1.22 ± 0.14	1.28 ± 0.15	17.23 ± 1.99	18.00 ± 2.08	<LOQ	<LOQ
Anthracene	3.09 ± 0.37	1.03 ± 0.12	6.67 ± 0.79	6.06 ± 0.72	0.52 ± 0.06	1.24 ± 0.15	<LOQ	<LOQ	<LOQ	<LOQ
Fluoranthene	<LOQ	<LOQ	12.01 ± 1.48	10.29 ± 1.27	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ
Pyrene	0.77 ± 0.09	0.54 ± 0.07	9.26 ± 1.11	7.99 ± 0.96	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ
Benz(a)anthracene	<LOQ	<LOQ	3.22 ± 0.36	2.01 ± 0.22	<LOQ	0.72 ± 0.08	<LOQ	<LOQ	<LOQ	<LOQ
Chrysene	0.73 ± 0.10	0.31 ± 0.04	1.05 ± 0.15	1.08 ± 0.15	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ
Benzo(b)fluoranthene	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ
Benzo(k)fluoranthene	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ
Benzo(a)pyrene	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ
Dibenz(a,h)anthracene	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ
Benz(g,h,i)perylene	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ
Indeno(1,2,3, -c, d)pyrene	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ
Total 13 PAHs	13.12	9.98	87.72	75.57	0.87	3.23	22.45	23.25		

n stands for the number of the samples

Table 4: Estimation of MOE in adult consumers due to madhbi chicken ingestion (result below LOQ substituted by LOQ value)

			Chronic daily intake μg/kg bw /day		MOE	
Cooking method	**Sample portion**	**TEQBaP μg/kg**	**Mean**	**P95**	**Mean**	**P95**
Madhbi	Thigh	2.359	2.5× 10-4	3.9× 10-4	1,888,375	1,238,281
	Breast	2.480	3.4× 10-4	5.3× 10-4	1,131,575	708,811

parts on PAH formation. The results confirmed that there were no significant differences between PAH levels in the breast and thigh (> 0.05) of cooked chicken. According to Lee et al. (2020), the fat content is one of the essential factors contributing to PAH formation in meat. Under sufficient heat, fat is pyrolyzed to form PAHs, which explains the increased PAH levels detected in cooked fatty foods (Oz, 2021). Fat content in chicken breast and thigh are similar (2.22% and 2.99%, respectively), explaining the unobserved differences in PAH concentrations between breast and thigh (Edris et al., 2012). When comparing chicken with red meat, which has higher fat content, Alomirah et al. (2011) found significant variations in the concentration of PAHs between a chicken burger (13.2 μg/kg) and a red meat burger (110 μg/kg). Hence, lipid content in the samples has an evident impact on the formation of PAHs. Moisture content also plays a significant role in the formation of PAHs since it provides oxygen during heating, which prevents incomplete combustion (Lee et al., 2020).

4.3 Risk exposure of PAHs

Based on the dietary exposure assessment results, the MOE was estimated to characterize the risk of exposure to PAH4. The risk exposure of PAH4 was only measured in madhbi chicken as based on our findings, this kind of cooking method produced higher concentrations of PAHs compared to other cooking styles. The average concentration was used, and a worst-case scenario approach was also used by assigning the LOQ to all samples determined to be below the LOQ as suggested by Food and Agriculture Organization of the United Nations

Table 5: Estimation of MOE values from various studies

		Chronic daily intake ng/kg bw /day		MOE		
Country	**Foodstuffs**	**Mean**	**P95**	**Mean**	**P95**	**Reference**
Saudi	Chicken (Thigh)	0.25	0.39	1,888,375	1,238,281	This study
	Chicken (Breast)	0.34	0.53	1,131,575	708,811	
Korea	Fish and Shellfish	0.012	0.20	8,333,333	485,437	(Kim et al., 2014)
	Meat	0.290	3.90	344,828	25,634	
	Smoked products	0.038	0.37	2,631,579	265,957	
Turkey	Meat doner	1.24		274,193		(Sahin et al., 2020)
	Chicken doner	2.06		165,048		
	Grilled chicken	1.79		189,944		
France	Meat	0.040	0.066			(Veyrand et al., 2013)
	Poultry and game	0.029	0.089			
	Foodstuffs	1.4	2.99	230	113	
Denmark	grilled meat (Restaurant maximum)	48		7,08		(Duedahl-Olesen et al., 2015)
	grilled meat (Restaurant)	2.2		152		
	Home-grilled	10		33,8		
	barbecued meat	40		8,45		

and World Health Organization (2009) (Table 4). The result of the toxicity equivalency quotient of benzo(a)pyrene (TEQ_{BaP}) showed no significant difference between the TEQ_{BaP} of madhbi breast chicken (2.48 μg/kg) and madhbi thigh chicken (2.36 μg/kg) (Table 4). The 95% percentile of the CDI of PAH4 for adults in Saudi Arabia was estimated to be 5.3×10^{-4} μg/kg bw /day and 3.9×10^{-4} μg/kg bw /day for those eating madhbi breast chicken and madhbi thigh chicken, respectively.

The 95% percentile of MOE in adults due to the ingestion of madhbi chicken breasts and thighs was calculated as 708,811 and 1,238,281, respectively, which indicates no health concern. MOE values estimated in this study agreed with most published data from different countries (Table 5). Sahin et al. (2020) investigated the types and quantities of PAH compounds in doner kebabs (red meat and chicken), meatballs, grilled chicken and fish. In different heat-treated samples, the calculated MOE for PAH4 ranged between 165,048 and 274,193. In addition, Kim et al. (2014) found that the MOE for PAH4 was 485,437 for the consumption of fish and shellfish, 25,634 for the consumption of meat, and 265,957 for the consumption of smoked products in Korea. Veyrand et al. (2013) analyzed 725 foodstuffs, containing meat, consumed by the French population and found that the chronic daily intake of the 4 PAHs was low. In contrast, in Denmark, a worst-case scenario, assuming daily consumption of barbecued meat, estimated the MOE for PAH4 to be 8450 (Duedahl-Olesen et al., 2015).

5 Conclusions

PAH contamination of chicken thighs and breasts, cooked by traditional Arabian methods, was investigated using HPLC. The madhbi method was found to generate the highest levels of PAHs in comparison with other methods of cooking. Phenanthrene was the compound detected at the highest concentration in all cooking styles. There was no significant difference in the concentration of PAHs between the parts of chicken cooked with the same method. However, the method of cooking had a significant effect on PAH formation. Therefore, it can be stated with high certainty that PAHs in chicken could be decreased by choosing the appropriate cooking method. However, the calculated MOE for adults in Saudi Arabia was found to be more than the 10,000 critical limits reported by the EFSA, indicating that the results are within a safe range.

Acknowledgements

This research was financially supported by the Saudi Food and Drug Authority (SFDA). The authors are grateful to the Research and Laboratories Sector in the SFDA for their keen interest in this study and its instrumental support.

References

Almutairi, M., Alsaleem, T., Jeperel, H., Alsamti, M., & Alowaifeer, A. M. (2021). Determination of inorganic arsenic, heavy metals, pesticides and mycotoxins in Indian rice (Oryza sativa) and a probabilistic dietary risk assessment for the population of Saudi Arabia. *Regulatory toxicology and pharmacology*, *125*, Article 104986. https://doi.org/10.1016/j.yrtph.2021.104986

Alomirah, H., Al-Zenki, S., Al-Hooti, S., Zaghloul, S., Sawaya, W., Ahmed, N., & Kannan, K. (2011). Concentrations and dietary exposure to polycyclic aromatic hydrocarbons (PAHs) from grilled and smoked foods. *Food Control*, *22*(12), 2028–2035. https://doi.org/10.1016/j.foodcont.2011.05.024

Babić, J., Vidaković, S., Škaljac, S., Kartalović, B., Ljubojević, D., Ćirković, M., & Teodorović, V. (2017). Factors affecting elimination of polycyclic aromatic hydrocarbons from traditional smoked common carp meat. *IOP Conference Series: Earth and Environmental Science*, *85*, Article 012086. https://doi.org/10.1088/1755-1315/85/1/012086

Büyükkurt, Ö. K., Dinçer, E. A., Çam, İ. B., Candal, C., & Erbaş, M. (2020). The influence of cooking methods and some marinades on polycyclic aromatic hydrocarbon formation in beef meat. *Polycyclic Aromatic Compounds*, *40*(2), 195–205. https://doi.org/10.1080/10406638.2017.1392328

Chen, B. H., & Lin, Y. S. (1997). Formation of polycyclic aromatic hydrocarbons during processing of duck meat. *Journal of Agricultural and Food Chemistry*, *45*(4), 1394–1403. https://doi.org/10.1021/jf9606363

Codex Alimentarius Commission. (2009). *Code of practice for the reduction of contamination of food with polycyclic aromatic hydrocarbons (PAH) from smoking and direct drying processes* (Code of practice No. CXC 68-2009). CAC. https://www.fao.org/fao-who-codexalimentarius/sh-proxy/en/?lnk=1&url=https%253A%252F%252Fworkspace.fao.org%252Fsites%252Fcodex%252FStandards%252FCXC%2B68-2009%252FCXP_068e.pdf

Duedahl-Olesen, L., Aaslyng, M., Meinert, L., Christensen, T., Jensen, A. H., & Binderup, M. L. (2015). Polycyclic aromatic hydrocarbons (PAH) in Danish barbecued meat. *Food Control*, *57*, 169–176. https://doi.org/10.1016/j.foodcont.2015.04.012

Edris, A. M., Hemmat, M. I., Shaltout, F. A., Elshater, M. A., & Eman, F. M. I. (2012). Chemical analysis of chicken meat with relation to its quality. *Benha Veterinary Medical Journal*, *23*(1), 87–93.

European Food Safety Authority. (2005). Opinion of the scientific committee on a request from EFSA related to a harmonised approach for risk assessment of substances which are both genotoxic and carcinogenic. *EFSA Journal*, *3*(10), Article 282. https://doi.org/10.2903/j.efsa.2005.282

European Food Safety Authority. (2008). Polycyclic aromatic hydrocarbons in food: Scientific opinion of the panel on contaminants in the food chain. *EFSA Journal*, *6*(8), Article 724. https://doi.org/10.2903/j.efsa.2008.724

Falcó, G., L. Domingo, J., M. Llobet, J., Teixidó, A., Casas, C., & Müller, L. (2003). Polycyclic aromatic hydrocarbons in foods: Human exposure through the diet in Catalonia, Spain. *Journal of Food Protection*, *66*(12), 2325–2331. https://doi.org/10.4315/0362-028X-66.12.2325

Farhadian, A., Jinap, S., Hanifah, H. N., & Zaidul, I. S. (2011). Effects of meat preheating and wrapping on the levels of polycyclic aromatic hydrocarbons in charcoal-grilled meat. *Food Chemistry*, *124*(1), 141–146. https://doi.org/10.1016/j.foodchem.2010.05.116

Food and Agriculture Organization of the United Nations & World Health Organization. (2006). *Evaluation of certain food contaminants: Sixty-fourth report of the Joint FAO/WHO Expert Committee on Food Additives* (Technical No. 930) (OCLC: 133166834). World Health Organization. Geneva. Retrieved January 17, 2023, from https://apps.who.int/iris/handle/10665/43258

Food and Agriculture Organization of the United Nations & World Health Organization. (2009). *A risk-based decision tree approach for the safety evaluation of residues of veterinary drugs* (tech. rep.). JECFA. https://www.who.int/publications/m/item/a-risk-based-decision-tree-approach-for-the-safety-evaluation-of-residues-of-veterinary-drugs

Ghorbani, M., Saleh, H. N., Barjasteh-Askari, F., Nasseri, S., & Davoudi, M. (2020). The effect of gas versus charcoal open flames on the induction of polycyclic aromatic hydrocarbons in cooked meat: A systematic review and meta-analysis. *Journal of Environmental Health Science and Engineering*, *18*(1), 345–354. https://doi.org/10.1007/s40201-020-00457-0

Gratz, S., Mohrhaus, A., Gamble, B., Gracie, J., Jackson, D., Roetting, J., Ciolino, L., McCauley, H., Schneider, G., Crockett, D., Krol, W., Arsenault, T., White, J., Flottmeyer, M., Johnson, Y., Heitkemper, D., & Fricke, F. (2010). Screen for the presence of polycyclic aromatic hydrocarbons in select seafoods using LC-fluorescence. *Laboratory Information Bulletin*, (4475), 1–39.

Hokkanen, M., Luhtasela, U., Kostamo, P., Ritvanen, T., Peltonen, K., & Jestoi, M. (2018). Critical effects of smoking parameters on the levels of polycyclic aromatic hydrocarbons in traditionally smoked fish and meat products in Finland. *Journal of Chemistry*, *2018*([Special Issue]), Article 2160958. https://doi.org/10.1155/2018/2160958

Kim, M.-J., Hwang, J.-H., & Shin, H.-S. (2014). Evaluation of polycyclic aromatic hydrocarbon contents and risk assessment for fish and meat products in Korea. *Food Science and Biotechnology*, *23*(3), 991–998. https://doi.org/10.1007/s10068-014-0134-0

Lee, J.-S., Han, J.-W., Jung, M., Lee, K.-W., & Chung, M.-S. (2020). Effects of thawing and frying methods on the formation of acrylamide and polycyclic aromatic hydrocarbons in chicken meat. *Foods*, *9*(5), Article 573. https://doi.org/10.3390/foods9050573

Lee, J.-G., Kim, S.-Y., Moon, J.-S., Kim, S.-H., Kang, D.-H., & Yoon, H.-J. (2016). Effects of grilling procedures on levels of polycyclic aromatic hydrocarbons in grilled meats. *Food Chemistry*, *199*, 632–638. https://doi.org/10.1016/j.foodchem.2015.12.017

Malarut, J.-a., & Vangnai, K. (2018). Influence of wood types on quality and carcinogenic polycyclic aromatic hydrocarbons (PAHs) of smoked sausages. *Food Control*, *85*, 98–106. https://doi.org/10.1016/j.foodcont.2017.09.020

Nisbet, I. C. T., & LaGoy, P. K. (1992). Toxic equivalency factors (TEFs) for polycyclic aromatic hydrocarbons (PAHs). *Regulatory Toxicology and Pharmacology*, *16*(3), 290–300. https://doi.org/10.1016/0273-2300(92)90009-X

Onwukeme, V. I., Obijiofor, O. C., Asomugha, R. N., & Okafor, F. A. (2015). Impact of cooking methods on the levels of polycyclic aromatic hydrocarbons (PAHs) in chicken meat. *IOSR Journal of Environmental Science, Toxicology and Food Technology*, *9*(4), 21–27. https://doi.org/10.9790/2402-09412127

Oz, E. (2021). The impact of fat content and charcoal types on quality and the development of carcinogenic polycyclic aromatic hydrocarbons and heterocyclic aromatic amines formation of barbecued fish. *International Journal of Food Science & Technology*, *56*(2), 954–964. https://doi.org/10.1111/ijfs.14748

Rose, M., Holland, J., Dowding, A., Petch, S., White, S., Fernandes, A., & Mortimer, D. (2015). Investigation into the formation of PAHs in foods prepared in the home to determine the effects of frying, grilling, barbecuing, toasting and roasting. *Food and Chemical Toxicology*, *78*, 1–9. https://doi.org/10.1016/j.fct.2014.12.018

Sahin, S., Ulusoy, H. I., Alemdar, S., Erdogan, S., & Agaoglu, S. (2020). The presence of polycyclic aromatic hydrocarbons (PAHs) in grilled beef, chicken and fish by considering dietary exposure and risk assessment. *Food Science of Animal Resources*, *40*(5), 675–688. https://doi.org/10.5851/kosfa.2020.e43

Szopińska, M., Szumińska, D., Bialik, R. J., Dymerski, T., Rosenberg, E., & Polkowska, Ż. (2019). Determination of polycyclic aromatic hydrocarbons (PAHs) and other organic pollutants in freshwaters on the western shore of Admiralty Bay (King George Island, Maritime Antarctica). *Environmental Science and Pollution Research*, *26*(18), 18143–18161. https://doi.org/10.1007/s11356-019-05045-w

Tang, L., Tang, X.-Y., Zhu, Y.-G., Zheng, M.-H., & Miao, Q.-L. (2005). Contamination of polycyclic aromatic hydrocarbons (PAHs) in urban soils in Beijing, China. *Environment International*, *31*(6), 822–828. https://doi.org/10.1016/j.envint.2005.05.031

Terzi, G., Celik, T. H., & Nisbet, C. (2008). Determination of benzo[alpha]pyrene in Turkish döner kebab samples cooked with charcoal or gas fire [Accepted: 2014-07-24T13:21:25Z Publisher: Teagasc, Oak Park, Carlow, Ireland]. *Irish Journal of Agricultural and Food Research*, *47*, 187–193. Retrieved January 17, 2023, from http://hdl.handle.net/11019/634

United States Environmental Protection Agency. (2001). Risk assessment guidance for superfund: Volume III: part A, process for conducting probabilistic risk assessment (No. EPA 540-R-02-002; p. 385). EPA. https://www.epa.gov/risk/risk-assessment-guidance-superfund-rags-volume-iii-part

Veyrand, B., Sirot, V., Durand, S., Pollono, C., Marchand, P., Dervilly-Pinel, G., Tard, A., Leblanc, J.-C., & Le Bizec, B. (2013). Human dietary exposure to polycyclic aromatic hydrocarbons: Results of the second French total diet study. *Environment International*, *54*, 11–17. https://doi.org/10.1016/j.envint.2012.12.011

Zelinkova, Z., & Wenzl, T. (2015). The occurrence of 16 EPA PAHs in food: A review [Publisher: Taylor & Francis]. *Polycyclic Aromatic Compounds*, *35*(2-4), 248–284. https://doi.org/10.1080/10406638.2014.918550