

Effect of Heat Treatments on Polycyclic Aromatic Hydrocarbons Formation in Meat

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Abstract

A total of 80 samples of both beef and mutton (40, each), which were either raw or cooked by different cooking methods such as pan-frying, charcoal-grilling and charcoal-grilling with aluminum foils covers (n=10 for each of beef and mutton) besides 10 samples from raw meat of each type. The samples were collected from different restaurants at Zagazig City, Sharkia Governorate, Egypt. The samples were prepared for detection of 16 polycyclic aromatic hydrocarbons (PAHs) (naphthalene, acenaphthylene, acenaphthene, fluorene, phenanthrene, anthracene, fluoranthene, pyrene, benzo [a] anthracene, chrysene, benzo [b] fluoranthrene, benzo [k] fluoranthene, benzo [a] pyrene, Indeno [1,2,3c-d] pyrene, Dibenzo [a,h] anthracene and Benzo [g,h,i] perylene). The recorded results showed that the total PAHs for raw, fried, charcoal grilled and foil grilled beef samples were 0.247, 1.152, 6.833 and 1.265 µg/kg respectively. Meanwhile, PAHs residual concentrations in mutton samples were 1.09, 4.606, 26.819 and 6.279 µg/kg in raw, pan-fried, charcoal-grilled and foil-grilled mutton samples respectively. We found also when meat wrapped in aluminum foil during grilling, it leads to a decrease in the total PAHs in the meat samples.

Keywords: PAHs, Pan-frying, Charcoal-grilling, Foil-grilled, Aluminum foil

Introduction

Polycyclic aromatic hydrocarbons are wide spread as environmental pollutants, which can be generated during the preparation of food [1]. PAHs are originated from many sources of environment (natural and anthropogenic), industry of food processing (such as; heating, drying, and smoking processes), materials of package and some cooking practices (such as; grilling, roasting, and frying processes) [2]. Food is the main source of exposure to PAHs for people who do not smoke, diet may cause more than 90% of total PAHs exposure of general population in different countries [3]. For mean consumers across the European countries, dietary exposure for the sum of eight carcinogenic and genotoxic PAHs (PAH8) (chrysene, benzo [a] pyrene, benzo [b] fluoranthene, benzo [k] fluoranthene, dibenzo [a,h] anthracene, benzo [g,h,i] perylene, indeno [1,2,3-c,d] pyrene and fluorene) was estimated at 1.73 mg/kg [2]. Despite the high level of PAHs is not observed in raw food as

usual, the grilled food has been reported to contain PAHs at levels of 0 to 130 mg/kg [4,5]. The difference in PAHs levels in food could be attributed to the type and fat content, process of cooking (fried, grilled, roasted, boiled and smoked), temperature and cooking duration as well as fuel type used (electrical, gas, wood, and charcoal) [5].

Benzo [a] pyrene (BaP) is probably the most studied PAH. The International Agency for Research on Cancer (IARC) described BaP as probable human carcinogen in 1987 [6]. Thus, the BaP determination has been widely used in the analysis of the environment as an indicator for the PAH content [7]. Meat meals that are cooked by charcoal grilling and pan-frying are common at both home and restaurants in Egypt, also in other Arabian countries. Therefore, this study was performed to determine the effect of different cooking methods on the formation of PAHs in meat.

Material and Methods

Samples

A total of 80 samples of both beef and mutton (40, each), which were either raw or cooked by different cooking methods (pan-frying, charcoal-grilling and charcoal-grilling with aluminum foils covers, n=10 for each beef and mutton). Samples were collected from different restaurants at Zagazig City, Sharkia Governorate, Egypt. The samples were prepared for the detection of 16 polycyclic aromatic hydrocarbons (PAHs) (naphthalene, acenaphthylene, acenaphthene, fluorene, phenanthrene, anthracene, fluoranthene, pyrene, benzo [a] anthracene, chrysene, benzo [b] fluoranthene, benzo [k] fluoranthene, benzo [a] pyrene, Indeno [1,2,3c-d] pyrene, Dibenzo [a,h] anthracene and Benzo [g,h,i] perylene).

The beef and mutton samples were fried in small amounts of margarine. For charcoal-grilling, beef and mutton samples were grilled over a grill fueled by charcoal at different restaurants and shops till the color became yellowish brown (well done). In addition, for charcoal grilling with aluminum foils, beef and mutton samples were wrapped by aluminum foils then grilled on a grill till the color became yellowish brown (well done).

Extraction and preparation of samples

Samples were transferred in a separate labeled aluminum foil to the Pesticide Residue Department, Central Pesticide Laboratory, Agriculture Research Center, Giza, where the extraction procedures, clean up and estimation of PAH levels by gas chromatography were conducted. Analysis of the PAHs residues was done according to Villeneuve *et al.* [8]. Twenty grams from each examined sample were grinded in a mortar with anhydrous sodium sulfate (2 g). The mixture was then squeezed with 60 mL of hexane-acetone (1-1) (v/v) mixture, filtered and the tissue was extracted twice more. Organic solvent fractions were united and filtered through filter paper with 1 g anhydrous sodium sulfate. The extract was then evaporated to about 2 mL. The extract was transferred to a round-bottom flask and 100 mL of 10% aqueous methanolic potassium hydroxide were added. The mixture was refluxed for 3 h in order to saponify the

lipids. Finally, the content found in round-bottom flask was transferred to a separately funnel and cleansed with 150 mL of methanol-water (4:1) (v/v) mixture, then extracted with hexane (80 mL) to get back the non-saponified lipids.

Clean up of samples

Clean up was achieved with a silica/alumina column. Aromatic hydrocarbons were eluted with 30 ml of a mixture of hexane and dichloromethane (90:10) (v/v). The volume of the eluted fraction was reduced to 1 mL and analyzed by a gas liquid chromatography equipped with a flame ionization detector GC/FID.

Preparation of blank solution

The same volume of solvents and anhydrous sodium sulfate, that were used in the PAHs extraction of the examined samples were exposed to the same routine for the detection of any possible traces of the studied PAHs in the solvents or distilled water.

Gas chromatographic analysis

The procedure was carried out according to Moret and Conte [9]. The polycyclic aromatic hydrocarbon fraction was injected into a gas liquid chromatography equipped with a flame ionization detector GC/FID. The gas chromatograph used was Hewlett Packard GC Model 6890 equipped with a flame ionization detector (GC/FID). GC analysis was conducted on HP-608 (Agilent, Folsom, CA) fused silica capillary column (30-meter length x 0.53 millimeter internal diameter x 0.5 micrometer film thickness).

Gas chromatography operating conditions

Injector and detector temperatures were maintained at 280°C and 300°C, respectively. Initial oven temperature, 100°C for 2 min hold to 280°C at the rate of 6°C/min and was maintained at 280°C for 15 min.

Determination of the recovery percentage

The recoveries were done by adding the standard of PAHs mixture at 3 concentrations (1, 5 and 10 micrograms). The average percentages of recovery of PAHs for the examined samples at 3 levels were determined and estimated for all the tested PAHs in each sample (Figure 1).

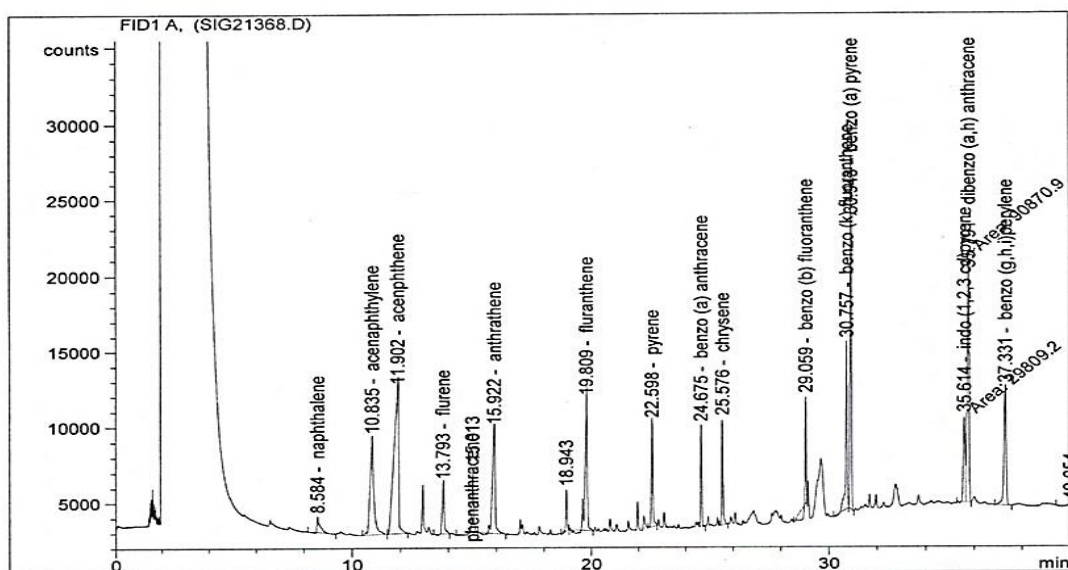


Figure 1: Exemplar for chromatogram of detected PAHs with their retention time.

Results and Discussion

The current study examined the presence of 16 PAHs compounds; namely; naphthalene, acenaphthylene, acenaphthene, fluorene, phenanthrene, anthracene, fluoranthene, pyrene, benzo [a] anthracene, chrysene, benzo [b] fluoranthene, benzo [k] fluoranthene, benzo [a] pyrene, Indeno [1,2,3c-d] pyrene, Dibenzo [a,h] anthracene and Benzo [g,h,i] perylene in raw, pan-fried, charcoal-grilled and foil-grilled beef and mutton samples.

PAHs in beef

From Table (1), it is clear that the percentages of phenanthrene and anthracene in the examined samples were 10% and 10% in raw samples, 30% and 10% in pan-fried samples, 50% and 10% in charcoal-grilled samples, 10% and 10% in foil-grilled samples, respectively. Regarding to their residual concentrations, they were detected with mean values of 0.144 ± 0.144 and 0.103 ± 0.103 $\mu\text{g}/\text{kg}$ in raw samples, 0.787 ± 0.420 and 0.248 ± 0.248 $\mu\text{g}/\text{kg}$ in pan-fried samples, 1.501 ± 0.508 and 0.61 ± 0.61 $\mu\text{g}/\text{kg}$ in charcoal-grilled samples and 0.571 ± 0.572 and 0.174 ± 0.174 $\mu\text{g}/\text{kg}$ in foil-grilled samples, respectively.

Nearly similar results were obtained by Martorell *et al.* [10] who recorded the mean values of 0.16 and 0.16 $\mu\text{g}/\text{kg}$ for both phenanthrene and anthracene in veal respectively. Unlikely, Mishref [11] did not detect phenanthrene in kebab samples but

anthracene was recorded with a mean value of 18.2 ± 25.04 $\mu\text{g}/\text{kg}$ which is higher than the current results in charcoal grilled samples. Higher results were also obtained by Sinha *et al.* [12] and Falcó *et al* [13] who detected phenanthrene with mean values of 5.3 and 16.7 $\mu\text{g}/\text{kg}$, respectively.

Fluoranthene, pyrene, benzo [a] anthracene and benzo [b] fluoranthene compounds were detected only in charcoal grilled samples with the percentages of 10%, 10%, 30% and 10% with mean values of 0.451 ± 0.451 , 0.543 ± 0.543 , 0.545 ± 0.280 and 0.189 ± 0.189 $\mu\text{g}/\text{kg}$, respectively.

Higher results were obtained by Mishref [11] who detected fluoranthene and benzo [a] anthracene with mean values of 57 and 16.8 $\mu\text{g}/\text{kg}$, respectively, in kebab samples. Moreover, Hassan [14] detected fluoranthene and pyrene in charcoal grilled meat with mean values of 9.22 and 704.24 $\mu\text{g}/\text{kg}$, respectively. Additionally, Akpambaget al [15] recorded mean values of both fluoranthene and pyrene as 9.7 and 9.7 $\mu\text{g}/\text{kg}$, respectively, which were higher than the recorded results.

Regarding to benzo [a] pyrene, the most dangerous carcinogenic PAHs compound, it was detected in pan-fried, charcoal-grilled and foil-grilled meat samples with the percentages of 20%, each, with mean values of 0.117 ± 0.085 , 0.457 ± 0.306 and 0.092 ± 0.062 $\mu\text{g}/\text{kg}$, respectively, but was not detected in raw samples.

Table 1: Percentages of PAHs in beef and mutton meat samples exposed to different heat treatments

| No. | PAHs | Beef samples% | | | | Mutton samples% | | | |
|-----|------------------------|---------------|--------|----------|------------|-----------------|--------|----------|------------|
| | | Raw | Frying | grilling | Foil-grill | Raw | Frying | grilling | Foil-grill |
| 1 | Naphthalene | 0% | 0% | 0% | 0% | 0% | 0% | 40% | 20% |
| 2 | Acenaphthylene | 0% | 0% | 0% | 0% | 0% | 10% | 20% | 10% |
| 3 | Acenaphthene | 0% | 0% | 0% | 0% | 0% | 0% | 0% | 0% |
| 4 | Fluorine | 0% | 0% | 0% | 0% | 0% | 20% | 40% | 20% |
| 5 | Phenanthrene | 10% | 30% | 50% | 10% | 0% | 30% | 60% | 30% |
| 6 | Anthracene | 10% | 10% | 10% | 10% | 0% | 0% | 10% | 0% |
| 7 | Fluoranthene | 0% | 0% | 10% | 0% | 0% | 0% | 70% | 50% |
| 8 | Pyrene | 0% | 0% | 10% | 0% | 40% | 50% | 100% | 60% |
| 9 | Benzo(a)anthracene | 0% | 0% | 30% | 0% | 0% | 0% | 30% | 0% |
| 10 | Chrysene | 0% | 0% | 0% | 0% | 0% | 10% | 40% | 20% |
| 11 | Benzo(b)fluoranthene | 0% | 0% | 10% | 0% | 0% | 0% | 0% | 0% |
| 12 | Benzo(k)fluoranthene | 0% | 0% | 60% | 30% | 0% | 0% | 0% | 0% |
| 13 | Benzo(a)pyrene | 0% | 20% | 20% | 20% | 0% | 10% | 20% | 0% |
| 14 | Indeno(1,2,3c-d)pyrene | 0% | 0% | 0% | 0% | 0% | 0% | 10% | 0% |
| 15 | Dibenzo(a,h)anthracene | 0% | 0% | 0% | 0% | 30% | 50% | 80% | 50% |
| 16 | Benzo(g,h,i)perylene | 0% | 0% | 20% | 10% | 0% | 10% | 10% | 0% |

Higher results were obtained by Terzi *et al.* [16], Akpambagetal [15] and Mishref [11] who detected benzo [a] pyrene with mean values of 24.2, 2.8 and 9.2 $\mu\text{g}/\text{kg}$, respectively, in charcoal-grilled meat. However, lower results were recorded by Chung *et al.* [17], who found mean concentration of benzo [a] pyrene as 0.15 $\mu\text{g}/\text{kg}$.

The current findings concerning benzo [a] pyrene didn't exceed the MPL recommended by FAO/WHO (10 part per billion 'ppb'). Concerning benzo [k] fluoranthene and benzo [g,h,i] perylene, they could be detected in charcoal and foil grilled samples only, with the percentages of 60% and 20% in charcoal-grilled samples and 30% and 10% in foil-grilled samples, respectively. The mean values of benzo [k] fluoranthene in these samples were 1.933 ± 0.548 and 0.172 ± 0.089 $\mu\text{g}/\text{kg}$, while they were 0.604 ± 0.442 and 0.256 ± 0.256 $\mu\text{g}/\text{kg}$ for Benzo [g,h,i] perylene. Lower results were obtained by Martorell *et al.* [10], who detected benzo [k] fluoranthene and benzo [g,h,i] perylene with mean values of 0.06 and 0.07 ppb respectively, also, Olatunde *et al.* [18] detected benzo [k] fluoranthene and benzo [g,h,i] perylene with mean values of 0.1 and 0.3 ppb, respectively. Both of Mishref [11] and Hassan [14] did not detect any of these two compounds in such samples.

In the current study, it is clear that none of naphthalene, acenaphthylene, acenaphthene, fluorene, chrysene, indeno [1,2,3c-d] pyrene and dibenzo [a,h] anthracene were detected in raw, pan-fried, charcoal-grilled and foil-grilled meat samples.

The total PAHs in raw, pan-fried, charcoal-grilled and foil-grilled meat samples were 0.247, 1.152, 6.833 and 1.265 $\mu\text{g}/\text{kg}$ respectively (Table 2). Nearly similar results were obtained by Janoszka *et al.* [19] who recorded 2.77 $\mu\text{g}/\text{kg}$ total PAHs in pan-fried beef collar, while higher results were obtained by Sinha *et al.* [12] who recorded total PAHs in pan-fried meat as 10.7 $\mu\text{g}/\text{kg}$.

Chung *et al.* [17], who reported that the total PAHs of 0.80 ppb in charcoal-grilled samples which was lower than that obtained in the current study. Higher results were obtained by Falcó *et al.* [13], Farhadian *et al.* [5], Jahurul *et al.* [20], Mishref [11] and Hassan [14], who reported that total PAHs as 13.4, 132, 66.28, 119.8 and 1170.94 $\mu\text{g}/\text{kg}$, respectively.

The differences between the obtained results in this study and the others may be attributed to the fat percentage in the meat used in this study, type of charcoal, thickness of meat and well-doneness of meat.

Table 2: Residual concentrations of PAHs in the examined raw and heat-treated beef samples (n=10)

| PAHs | Raw | | | Fried | | | Charcoal-grilled | | | Foil-grilled | | |
|------------------------|-----|------|--------------|-------|------|--------------|------------------|------|--------------|--------------|------|--------------|
| | Min | max | Mean ± SE | min | Max | Mean ±SE | min | max | Mean ±SE | min | max | Mean SE |
| Naphthalene | ND | ND | ND | ND | ND | ND | ND | ND | ND | ND | ND | ND |
| Acenaphthylene | ND | ND | ND | ND | ND | ND | ND | ND | ND | ND | ND | ND |
| Acenaphthene | ND | ND | ND | ND | ND | ND | ND | ND | ND | ND | ND | ND |
| Fluorine | ND | ND | ND | ND | ND | ND | ND | ND | ND | ND | ND | ND |
| Phenanthrene | 0 | 1.44 | 0.144±0.144 | 0 | 3.47 | 0.787±0.420 | 0 | 3.33 | 1.501±0.508 | 0 | 5.71 | 0.571±0.572 |
| Anthracene | 0 | 1.03 | 0.103±0.103 | 0 | 2.48 | 0.248±0.248 | 0 | 6.1 | 0.61±0.61 | 0 | 1.74 | 0.174±0.174 |
| Fluoranthene | ND | ND | ND | ND | ND | ND | 0 | 4.51 | 0.451±0.451 | ND | ND | ND |
| Pyrene | ND | ND | ND | ND | ND | ND | 0 | 5.43 | 0.543±0.543 | ND | ND | ND |
| Benzo(a)anthracene | ND | ND | ND | ND | ND | ND | 0 | 2.06 | 0.545±0.280 | ND | ND | ND |
| Chrysene | ND | ND | ND | ND | ND | ND | ND | ND | ND | ND | ND | ND |
| Benzo(b)fluoranthene | ND | ND | ND | ND | ND | ND | 0 | 1.89 | 0.189±0.189 | ND | ND | ND |
| Benzo(k)fluoranthene | ND | ND | ND | ND | ND | ND | 0 | 4 | 1.933±0.548 | 0 | 0.66 | 0.172±0.089 |
| Benzo(a)pyrene | ND | ND | ND | 0 | 0.81 | 0.117±0.085 | 0 | 2.48 | 0.457±0.306 | 0 | 0.52 | 0.092±0.062 |
| Indeno(1,2,3c-d)pyrene | ND | ND | ND | ND | ND | ND | ND | ND | ND | ND | ND | ND |
| Dibenzo(o,h)anthracene | ND | ND | ND | ND | ND | ND | ND | ND | ND | ND | ND | ND |
| Benzo(g,h,i)perylene | ND | ND | ND | ND | ND | ND | 0 | 4.24 | 0.604±0.442 | 0 | 2.56 | 0.256±0.256 |
| Total PAHs | | | 0.247 | | | 1.152 | | | 6.833 | | | 1.265 |

Min= minimum, Max= maximum, SE= standard error, ND= not detected

Table 3: Residual concentrations of PAHs in examined raw and heat-treated mutton samples (n=10)

| PAHs | Raw | | | Fried | | | Charcoal-grilled | | | Foil grilled | | |
|------------------------|-----|------|--------------|-------|------|--------------|------------------|-------|---------------|--------------|------|--------------|
| | Min | max | Mean ± SE | Min | max | Mean ±SE | min | max | Mean ±SE | Min | max | Mean SE |
| Naphthalene | ND | ND | ND | ND | ND | ND | 0 | 6.62 | 1.783±0.823 | 0 | 3.58 | 0.69±0.458 |
| Acenaphthylene | ND | ND | ND | 0 | 0.53 | 0.053±0.053 | 0 | 3.3 | 0.596±0.400 | 0 | 0.71 | 0.071±0.071 |
| Acenaphthene | ND | ND | ND | ND | ND | ND | ND | ND | ND | ND | ND | ND |
| Fluorine | ND | ND | ND | 0 | 1.27 | 0.23±0.154 | 0 | 8.75 | 1.848±0.952 | 0 | 3.31 | 0.526±0.365 |
| Phenanthrene | ND | ND | ND | 0 | 1.44 | 0.289±0.160 | 0 | 1.46 | 0.662±0.189 | 0 | 0.81 | 0.200±0.105 |
| Anthracene | ND | ND | ND | ND | ND | ND | 0 | 0.49 | 0.049±0.049 | ND | ND | ND |
| Fluoranthene | ND | ND | ND | ND | ND | ND | 0 | 1.52 | 0.654±0.182 | 0 | 1.81 | 0.459±0.191 |
| Pyrene | 0 | 3.67 | 0.917±0.417 | 0 | 8.3 | 3.177±1.121 | 1.02 | 23.42 | 13.979±1.884 | 0 | 6.91 | 3.426±0.968 |
| Benzo(a)anthracene | ND | ND | ND | ND | ND | ND | 0 | 16.57 | 2.517±1.744 | ND | ND | ND |
| Chrysene | ND | ND | ND | 0 | 4.38 | 0.438±0.438 | 0 | 14.6 | 3.491±1.729 | 0 | 3 | 0.565±0.378 |
| Benzo(b)fluoranthene | ND | ND | ND | ND | ND | ND | ND | ND | ND | ND | ND | ND |
| Benzo(k)fluoranthene | ND | ND | ND | ND | ND | ND | ND | ND | ND | ND | ND | ND |
| Benzo(a)pyrene | ND | ND | ND | 0 | 0.19 | 0.019±0.019 | 0 | 0.19 | 0.030±0.021 | ND | ND | ND |
| Indeno(1,2,3c-d)pyrene | ND | ND | ND | ND | ND | ND | 0 | 5.33 | 0.533±0.533 | ND | ND | ND |
| Dibenzo(a,h)anthracene | 0 | 0.9 | 0.173±0.098 | 0 | 1.05 | 0.4±0.148 | 0 | 0.98 | 0.503±0.099 | 0 | 0.71 | 0.246±0.863 |
| Benzo(g,h,i)perylene | ND | ND | ND | ND | ND | ND | 0 | 1.74 | 0.174±0.174 | 0 | 0.96 | 0.096±0.096 |
| Total PAHs | | | 1.09 | | | 4.606 | | | 26.819 | | | 6.279 |

Min= minimum, Max= maximum, SE= standard error, ND= not detected

PAHs in mutton

The results in Table (3) indicated that the percentages of pyrene and dibenzo [a,h] anthracene in the examined samples were 40% and 30% in raw samples, 50% and 50% in pan-fried samples, 100% and 80% in charcoal-grilled samples and 60% and 50% in foil-grilled samples, respectively. Regarding to their residual concentrations, they were detected with mean values of 0.19 ± 0.417 and 0.173 ± 0.098 $\mu\text{g}/\text{kg}$ in raw samples, 3.177 ± 1.121 and 0.4 ± 0.148 $\mu\text{g}/\text{kg}$ in pan-fried samples, 13.979 ± 1.884 and 0.503 ± 0.099 $\mu\text{g}/\text{kg}$ in charcoal-grilled samples and 3.426 ± 0.968 and 0.246 ± 0.863 $\mu\text{g}/\text{kg}$ in foil-grilled samples, respectively.

The percentages of acenaphthylene, flourene, phenanthrene and chrysene in the current study were 10%, 20%, 30% and 10% in pan-fried samples, 20%, 40%, 60% and 40% in charcoal-grilled samples and 10%, 20%, 30% and 20% in foil-grilled samples, respectively. Their mean residual concentrations were 0.053 ± 0.053 , 0.23 ± 0.154 , 0.289 ± 0.159 and 0.438 ± 0.438 $\mu\text{g}/\text{kg}$ in pan-fried samples, 0.596 ± 0.4 , 1.848 ± 0.952 , 0.662 ± 0.189 and 3.491 ± 1.729 $\mu\text{g}/\text{kg}$ in charcoal-grilled samples and 0.071 ± 0.071 , 0.526 ± 0.365 , 0.2 ± 0.105 and 0.565 ± 0.378 $\mu\text{g}/\text{kg}$ in foil-grilled samples, respectively.

Concerning benzo [a] pyrene, it was detected in fried and charcoal-grilled samples with mean values of 0.019 ± 0.019 and 0.030 ± 0.021 $\mu\text{g}/\text{kg}$, respectively. Higher results were obtained by Ayguns and Kabadayi, [21] and Kao et al. [22], who recorded benzo [a] pyrene levels as 43.8 ± 1.8 and 5.8 ± 0.5 $\mu\text{g}/\text{kg}$ in grilled lamb meat and lamb steak, respectively.

Naphthalene, fluoranthene and benzo [g,h,i] perylene compounds were detected in charcoal and foil-grilled samples only, with the percentages of 40%, 70% and 10% in charcoal-grilled samples and 20%, 50% and 10% in foil-grilled samples, respectively. Their mean values were 1.783 ± 0.823 and 0.69 ± 0.458 $\mu\text{g}/\text{kg}$ for naphthalene, 0.654 ± 0.182 and 0.459 ± 0.191 $\mu\text{g}/\text{kg}$ for fluoranthene, 0.174 ± 0.174 and 0.096 ± 0.096 for benzo [g,h,i] perylene.

Anthracene, benzo [a] anthracene and indeno [1,2,3c-d] pyrene compounds were detected only in charcoal-grilled samples in our study with the percentages of 10%, 30% and 10%, respectively. Their mean values were 0.049 ± 0.049 , 2.517 ± 1.744 and 0.533 ± 0.533 $\mu\text{g}/\text{kg}$, respectively. In the current study, it is clear that non of acenaphthene, benzo [b] fluoranthene and benzo [k] fluoranthene could be detected in raw, fried, charcoal-grilled and foil-grilled mutton samples. It is clear from Table (3) that the total PAHs for raw, pan-fried, charcoal-grilled and foil-grilled mutton samples were 1.09, 4.606, 26.819 and 6.279 $\mu\text{g}/\text{kg}$, respectively.

For both beef and mutton

It is obvious that quantitative PAHs profile was different in pan-fried, charcoal-grilled and foil-grilled meat samples, which may be attributed to cooking temperature, cooking method and cooking time [5]. Generally, the formation mechanism of PAHs in grilled or smoked diets is unknown, it is generally considered that at least 3 possible mechanisms exist: The 1st mechanism is the organic matter pyrolysis as fat, carbohydrates and protein at temperatures higher than 200oC, and the formation of PAH mainly arise at a temperature ranges from 500 to 900oC [23]. The largest levels of PAHs may be arising from fat pyrolysis [24].

The second mechanism is due to the direct contact of the dripping arise from lipids at an extreme heat directly over a flame. This situation could produce volatile PAHs which then sticked to the food surface and the smokes rise [25]. The third mechanism is the incomplete combustion of charcoal which can generate PAHs that are brought onto the surface of the food [26].

It is obvious that the total PAHs increased after thermal treatments and this result came in harmony with WHO [4], which reported that high levels of PAHs are not usually observed in raw food. In addition, Phillips [27] mentioned that cooking processes can generate PAHs in food. It was observed that total PAHs of both beef and mutton samples is lower in fried samples than that of charcoal and foil grilled samples. On the contrary, Perelló [28] stated that the highest PAHs concentrations found after frying.

Meanwhile, Larson et al. [29] mentioned that frying didn't lead to any appreciable increase of the original trace level, and this was in agreement with this study.

Interestingly, it was found that total PAHs decreased in foil-grilled samples than charcoal-grilled one, this mean that when meat is wrapped with aluminum foil during grilling, it leads to a decrease in the total PAHs in the meat samples. These results are in agreement with Farhadian et al [30], who used aluminum foil and banana leaves to reduce total PAHs in meat samples during grilling and reported that this method is working well.

The health risk associated to the high concentration of benzo(a)pyrene, benzo(a)anthracene and chrysene in the present study was reported by Nisbet and LaGoy [31] and IARC [32], who recorded that the PAHs were proven to be animal carcinogens and in human they are suspected to be carcinogen. Therefore, many considerations should be taken before consumption of meat grilled over charcoal because large amounts of PAHs could be eaten in a single diet.

The total PAHs in beef is lower than that of mutton. This may be due to the mutton used in this study was higher in fat than beef. The percentage of fat plays an important role in PAHs formation, because barbecuing leads to melting of fat that dropped during the grilling period which provides the formation of PAHs [33].

Conclusion

In conclusion, the total PAHs increased after thermal treatments than that in raw samples. Additionally, in both beef and mutton samples, total PAHs is lower in fried samples than that of charcoal and foil-grilled samples. Total PAHs decreased in foil-grilled samples than charcoal grilled one, this means that, when meat wrapped in aluminum foil during grilling, it leads to a decrease in the total PAHs in the meat samples. Benzo [a] pyrene didn't exceed the MPL recommended by FAO/WHO (10 ppb). So, it seems that it is preferable to the consumers to wrap the charcoal grilled meat in an aluminum foil prior to grilling.

Conflict of interest

None of the authors have any conflict of interest to declare.

References

- [1] Jägerstad, M. and Skog, K. (2005): Review genotoxicity of heat-processed foods. *Mutat Res*, 574(1-2): 156-172.
- [2] EFSA (European Food Safety Authority) (2008): Scientific opinion of the panel on contaminants in the food chain on a request from the European Commission on polycyclic aromatic hydrocarbons in food. *EFSA J*, 724:1-114.
- [3] Llobet, J. M.; Falcó, G.; Bocio, A. and Domingo, J. L. (2006): Exposure to polycyclic aromatic hydrocarbons of edible marine species in Catalonia, Spain. *J Food Protect*, 69(10): 2493-2499.
- [4] World Health Organization (WHO) (1998): Selected non-heterocyclic polycyclic aromatic hydrocarbons. *Environ. Health Crit. No. 202*. Geneva, Switzerland.
- [5] Farhadian, A.; Jinap, S.; Abas, F. and Sakar, Z.I. (2010): Determination of polycyclic aromatic hydrocarbons in grilled meat. *Food Control*, 21(5): 606-610.
- [6] International Agency for Research on Cancer (IARC) (1987): Overall evaluation of carcinogenicity: An updating of IARC Monographs, vol. 1-42, IARC Monographs on the Evaluation of Carcinogenic Risks to Humans, Suppl. 7, IARC, Lyon.
- [7] European Commission (2006): Commission regulation (EC) no. 1881/2006 of 19 December 2006 setting maximum levels for certain contaminants in foodstuffs. *Official Journal of the European Union*, 364/5.
- [8] Villeneuve, J. P.; Carvalho, F. P.; Fowler, S. W. and Cattini, C. (1999): Levels and trends of PCBs, chlorinated pesticides and petroleum hydrocarbons in mussels from the NW Mediterranean coast: comparison of concentrations in 1973 1974 and 1988 1989. *Sci Total Environ*, 237: 57-65.

- [9] Moret, S. and Conte, L.S. (2000): Polycyclic aromatic hydrocarbons in edible fats and oils: occurrence and analytical methods. *J Chromatogr A*, 882(1):245-253.
- [10] Martorell, I.; Perelló, G.; Martí-Cid, R.; Castell, V.; Llobet, J. M. and Domingo, J. L. (2010): Polycyclic aromatic hydrocarbons (PAH) in foods and estimated PAH intake by the population of Catalonia, Spain, temporal trend. *Environ Int*, 36(5):424-432.
- [11] Mishref, M. A. (2014): Assessment of some polycyclic aromatic hydrocarbons residues in prefabricated meat for consumption and its relevance to food safety. M. V. Sc. Thesis, Fac. Vet. Med., Zagazig Univ., Egypt.
- [12] Sinha, R.; Rothman, N.; Brown, E.; Mark, S.; Hoover, R.; Caporaso, N.; Levander, O.; Knize, M.; Lang, N. and Kadlubar, F. (1994): Pan-Fried Meat Containing High Levels of Heterocyclic Aromatic Amines but Low Levels of Polycyclic Aromatic Hydrocarbons Induces Cytochrome P4501A2 Activity in Humans. *Cancer Res*, 54(23): 6154-6159.
- [13] Falco, G.; Domingo, J. L.; Llobet, J. M.; Teixido, A.; Casas, C. and Müller, L. (2003): Polycyclic Aromatic Hydrocarbons in Foods: Human Exposure through the Diet in Catalonia, Spain. *J Food Protect*, 66(12): 2325-2331.
- [14] Hassan, S. M. (2016): Detection of Some Chemical Residues In Grilled Meats With Special Reference To Their Public Health Importance. M. V. Sc. Thesis, Fac. Vet. Med., Zagazig Univ., Egypt.
- [15] Akpambanga, V.O.E.; Purcarob, G.; Lajidea, L.; Amooa, I.A.; Conteb, L.S. and Moret, S. (2009): Determination of polycyclic aromatic hydrocarbons (PAHs) in commonly consumed Nigerian smoked/grilled fish and meat. *Food Addit Contam*, 26(7): 1096-1103.
- [16] Terzi, G.; Çelik, T.H. and Nisbet, C. (2008): Determination of benzo[a]pyrene in Turkish döner kebab samples cooked with charcoal or gas fire. *Irish J Agri Food Res*, 47: 187-193.
- [17] Chung, S.Y.; Ramesh, R. Y.; Kim, J.S.; Kwon, K.; Kim, M.C. and David, B. (2011): Effects of grilling and roasting on the levels of polycyclic aromatic hydrocarbons in beef and pork. *Food Chem*, 129(4): 1420-1426.
- [18] Olatunde, O.S.; Fatoki, O.S.; Opeolu, B.O. And Ximba, B.J. (2014): Determination of polycyclic aromatic hydrocarbons [PAHs] in processed meat products using gas chromatography – Flame ionization detector. *Food Chem*, 156: 296-300.
- [19] Janoszka, B.; Warzecha, L.; Błaszczuk, U. and Bodzek, D. (2004): Organic compounds formed in thermally treated high-protein food, Part I: Polycyclic aromatic hydrocarbons. *Acta Chromatogr*, 14: 115-128.
- [20] Jahurul, M.H.A.; Jinap, S.; Zaidul I.S. M.; Sahena, F.; Farhadian, A. and Hajeb, P.(2013): Determination of fluoranthenebenzo [b] fluoranthene and benzo [a] pyrene in meat and fish products and their intake by Malaysian. *Food biosciences*,1:73-80.
- [21] Aygün, S. and Kabadayi, F. (2005): Determination of benzo [a] pyrene in charcoal grilled meat samples by HPLC with fluorescence detection. *Int j food Sci Nutr*, 56(8): 581-585.
- [22] Kao, T.H.; Chen, S.; Huang, C.W.; Chen, C.J. and Chen, B.H. (2014): Occurrence and exposure to polycyclic aromatic hydrocarbons in kindling-free-charcoal grilled meat products in Taiwan. *Food Chem Toxicol*, 71: 149-158.
- [23] Knize, M. G.; Salmon, C. P.; Pais, P. and Felton, J. S. (1999): Food heating and the formation of heterocyclic aromatic amine and PAH mutagens/carcinogens. In L. S. Jackson, M. G. Knize, & J. N. Morgan (Eds.), *Impact of processing on food safety* (pp. 179-193). New York: Kluwer Academic.
- [24] Bartle, K.D. (1991): Analysis and occurrence of PAHs in food. In C. S. Creaser, & R. Purchase (Eds.), *Food contam: Sources and surveillance* (pp.

- 41-60). Cambridge: Royal Society of Chemistry.
- [25] European Commission Scientific Committee on Foods (2002): Opinion of the scientific committee on food on the risks to human health of polycyclic aromatic hydrocarbons in food expressed on fourth December 2002. Brussels, Eur. Comm. health and consumer Protection.
- [26] Dyremark, A.; Westerholm, R.; Övervik, E. and Gustavsson, J. (1995): PAH emissions from charcoal grilling. Atmos Environ, 13: 1553-1558.
- [27] Phillips, D. H. (1999): Polycyclic aromatic hydrocarbons in the diet. Mutat Res, 443:139-147.
- [28] Perelló, G.; Martí-Cid, R.; Castell, V.; Llobet, J.M. and Domingo, J.L. (2009): Concentrations of polybrominated diphenyl ethers, hexachlorobenzene and polycyclic aromatic hydrocarbons in various foodstuffs before and after cooking. Food Chem Toxicol, 47(4): 709-715.
- [29] Larsson, B.K.; Sahlberg, G.P.; Eriksson, A.T. and Busk, L.A. (1983): Polycyclic aromatic hydrocarbons in grilled food. J Agr Food Chem, 31(4): 867-873.
- [30] Farhadian, A.; Jinap, S.; Hanifah, H.N. and Zaidul, I.S. (2011): Effects of meat preheating and wrapping on the levels of polycyclic aromatic hydrocarbons in charcoal-grilled meat. Food Chem, 124(1): 141-146.
- [31] Nisbet, I.C.T. and LaGoy, P.K. (1992): Toxic equivalency factor (TEFs) for polycyclic aromatic hydrocarbons (PAHs). Regul Toxicol Pharm, 16(3): 290-300.
- [32] International Agency Research for Cancer (IARC) (2010): Monographs on the evaluation of carcinogenic risk to humans. Some non-heterocyclic polycyclic aromatic hydrocarbons and some related exposures, Vol. 92. Lyon, France: IARC, 36-37.
- [33] Viegas, O.; Novo, P.; Pinto, E.; Pinho, O. and Ferreira, O. (2012): Effect of charcoal types and grilling conditions on formation of heterocyclic aromatic amines (HAs) and polycyclic aromatic hydrocarbons (PAHs) in grilled muscle foods. Food Chem Toxicol, 50(6): 2128-2134.

الملخص العربي

تأثير طرق الطهي المختلفة على تكوين المواد الهيدروكربونية الأروماتية متعددة الحلقات في اللحوم السعيد أبو زيد الدالي^١, عبد السلام الدايموني حافظ^١, وجيه صبحي درويش^١, رانيا محمد عبد الحميد^٢, دعاء فوزي الملط^١

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تم تجميع ٨٠ عينة (٤٠ عينة من كل من اللحوم البقري ولحوم الضأن) مقسمة لكل منهما كالتالي: عينات اللحوم البقري (١٠ لحوم نيئة، ١٠ بعد التحمير، ١٠ بعد الشواء على الفحم، ١٠ بعد الشواء على الفحم وملفوفة في ورق الألمونيوم)، و عينات لحوم الضأن (١٠ لحوم نيئة، ١٠ بعد التحمير، ١٠ بعد الشواء على الفحم، ١٠ بعد الشواء على الفحم وملفوفة في ورق الألمونيوم) من مطاعم مختلفة في مدينة الزقازيق، محافظة الشرقية، وتم قياس ١٦ من أهم بقايا الهيدروكربونات الأروماتية متعددة الحلقات في هذه العينات. دلت النتائج على وجود إجمالي الهيدروكربونات الأروماتية متعددة الحلقات بمتوسطات قدرها ٢٤٧، ١٥٢، ١، ١٥٢، ١، ٨٣٣ و ٢٦٥ ميكروجرام لكل كجم من اللحوم النيئة، والمحمرة، والمشوية على الفحم والمشوية على الفحم والملفوفة في ورق الألمونيوم على التوالي في عينات اللحم البقري. أما بالنسبة لعينات لحوم الضأن دلت النتائج على تواجد إجمالي الهيدروكربونات الأروماتية متعددة الحلقات بمتوسطات قدرها ٢٤٧، ١٥٢، ١، ٨٣٣ و ٢٦٥ ميكروجرام لكل كجم من اللحوم النيئة، والمحمرة، والمشوية على الفحم والمشوية على الفحم وملفوفة في ورق الألمونيوم على التوالي. أوضحت النتائج ان لف اللحوم بورق الألمونيوم أدت إلى تقليل هذه المواد الخطيرة.