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Occurrence and exposure to polycyclic aromatic hydrocarbons in kindling-free-charcoal grilled meat products in Taiwan

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ABSTRACT

This study aimed to determine the contents of 16 PAHs in kindling-free-charcoal grilled meat and seafood products by GC–MS coupled with a QuEChERS method, and estimate the potential risk associated with consumption of those products in Taiwan. Results showed that the total PAHs contents ranged from 6.3 ± 0.9 to 238.8 ± 8.3 ng/g in poultry meat, 0.1 ± 0.0–547.5 ± 12.2 ng/g in red meat, and 6.6 ± 1.4–249.7 ± 6.4 ng/g in seafood products. Among various PAHs, the highly carcinogenic benzo[a]pyrene was detected in chicken breast grilled at 84 °C (30 min), chicken heart at 100 °C (26 min), chicken drumstick at 74 °C (20 min), duck drumstick at 85 °C (40 min), and lamb steak at 88 °C (12 min), with its level amounting to 1.3 ± 0.0, 2.4 ± 0.1, 4.0 ± 1.3, 3.1 ± 0.0, and 5.8 ± 0.5 ng/g, respectively. The generation of PAHs was associated with grilling time, temperature and fat content. Risk assessment of dietary exposure to PAHs revealed toxicity equivalent to range from ND – 6.174 ± 0.505 μg/g and margin of exposure was >10,000, which agreed with the EFSA's definition of low public health concern. The lifelong average daily PAHs intake was higher for adults than for elderly people in Taiwan, however, consumption of kindling-free-charcoal grilled meat should not be a public health concern based on cancer risk potency.

1. Introduction

Polycyclic aromatic hydrocarbons (PAHs) are a class of environmental pollutants that contaminate food through polluted soil, water and air. More than 100 PAHs have been characterized so far, some of which are shown to be mutagenic and carcinogenic. Among various food products, PAHs often occur in meat products because of pyrolysis of major components like fat when heated at a temperature >200 °C (EC, 2006; Alomirah et al., 2011). More elaborately, during heating, lipid can be degraded into hydroperoxide or cyclohexene compounds generating PAHs in the smoke, which in turn adhere to food surface (Chen and Chen, 2001). Of the various PAHs, naphthalene with 2 aromatic rings was more susceptible to formation than the other PAHs during meat grilling (Ferrarese et al., 2008). In addition, unlike the highly toxic benzo[a]pyrene containing 5 aromatic rings, naphthalene is characterized as possibly carcinogenic (group 2B) (IARC, 2002).

In Taiwan, grilled meat is becoming increasingly popular in both homes and restaurants as grilling can impart unique flavor and tenderness. However, grilled meat often pose a potential health risk because of presence of carcinogenic PAHs. Pan and Cao (2010) studied the effect of charcoal grilling time on PAH formation, and reported a rise in benzo[a]pyrene content following an increase in heating time. In another study dealing with PAHs occurrence in Peking duck prepared by the hung oven process, benzo[a]pyrene and total PAHs were present at 8.7 and 54.7 μg/kg, respectively, with skin containing the highest level (Lin et al., 2011). Similarly, a study conducted in Taiwan revealed the presence of total PAHs ranged from 6.48 to 7.26 μg/kg in grilled meat with benzo[a]pyrene accounting for 52.9–54.4% of total PAHs (Chien and Yeh, 2010). Interestingly, Farhadian et al. (2010) demonstrated a higher concentration of PAHs in charcoal-grilled meat than in flame-gas or oven-grilled meat, however, no PAHs were detected upon heating by an electric oven or toaster. The authors also concluded that both grilling method and fat content in meat should have a great impact on PAHs formation in foods.

Though the PAHs formation in charcoal-grilled beef and pork have been extensively studied, the effects of kindling-free-charcoal grilling on PAHs generation in meat products still remains unexplored. Many studies dealing with human exposure to PAHs based on dietary habit have been published (Akpambang et al., 2009; Martorell et al., 2012; Reinik et al., 2007; Veyrand et al., 2013), however, the risk assessment of dietary exposure to PAHs in grilled meat products in Taiwan remains unknown. Therefore, the objectives of this study were to determine PAHs contents in

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kindling-free-charcoal grilled poultry meat, red meat and seafood products by employing a GC–MS technique coupled with QuEChERS method. In addition, the toxicity equivalent (TEQ) and margin of exposure (MOE) based on PAHs in grilled meat products as well as the lifelong average daily dietary intake (LADI) of PAHs in Taiwan were investigated.

2. Materials and methods

2.1. Chemicals

A total of 16 PAHs standards, including naphthalene, acenaphthylene, acenaphthene, fluorene, phenanthrene, anthracene, fluoranthene, pyrene, benzo[a]anthracene, chrysene, benzo[b]fluoranthene, benzo[k]fluoranthene, benzo[a]pyrene, indeno[1,2,3-c,d]pyrene, dibenzo[a,h]anthracene and benzo[g,h,i]perylene, were purchased from Supelco (Bellefonte, PA, USA). The QuEChERS kits used for extraction and purification of PAHs from meat samples were procured from Agilent Technologies (Palo Alto, CA, USA). Deionized water was obtained through a Barnstead Easypure II water purification system (Thermo Scientific Co., Waltham, MA, USA). Soy sauce and crystal sugar were bought from Gin-Lan Food Co. (Taoyuan, Taiwan) and Taiwan Sugar Co. (Tainan, Taiwan), respectively, while solvents acetonitrile and acetone were from Merck (Darmstadt, Germany).

2.2. Meat and seafood samples

A total of 15 meat and seafood commodities including chicken heart, chicken drumstick, chicken gizzard, chicken breast, duck drumstick, beef steak, pork knuckle, ham, lamb steak, pork chop, oyster, octopus, salmon, shrimp and squid were purchased from a butcher shop in New Taipei City, Taiwan. Then, all the samples were washed with tap water and weighed before grilling, followed by dividing each into two portions, placing into two separate bags and storing at −20 °C freezer. Of all the samples, only oyster was bought from a local seafood market and sent to the pilot plant immediately prior to grilling, as freezing might cause texture change after thawing. The fat contents in all the samples were determined by using a Soxhlet ethyl ether extraction method (AOAC, 2000).

2.3. Kindling-free charcoal grilling

Three types of charcoals, including kindling-free-charcoal, traditional wood charcoal and smoke-free charcoal, are widely used for meat grilling in Taiwan, especially in restaurants. The kindling-free-charcoal is a wood-basis coal made of wood, binder and flash powder containing potassium nitrite and magnesium powder. This charcoal is chosen as it is prone to get burned without much smoke production. Also, it possesses fast ignition and less incomplete combustion characteristics during grilling. Initially, 10 chucks of kindling-free-charcoal were placed onto the bottom of a home-use griller (Classic Barrel Grill Model 20040410, Masterbuilt Co., GA, USA), after which the griller was ignited and covered with a lid. After the griller temperature reached 200 °C, meat samples were placed over the grill stand at a distance of about 15 cm from the charcoal. The grilling condition was carefully controlled according to USDA recommendation by maintaining an appropriate grilling time and internal temperature for each meat commodity to attain optimal tenderness as shown in Table 1. Additionally, a longer grilling time and higher internal temperature was used for studying its effect on PAHs generation (Table 1). The internal temperature was determined using a probe-type thermometer (model 307, Digital Thermometer, Deange Industry Co., Taiwan), with the diameter being 2.58 mm. Then the thermometer was inserted into sample during measurement. All the meat samples were turned over occasionally (every 30–60 s) during grilling to prevent overcooking and meat surface from burning. After cooking, all the samples were cooled and then stored at −20 °C for PAHs analyses.

2.4. Extraction and purification of PAHs

Initially grilled and stored meat and seafood samples were removed from freezer, thawed at 4 °C, and then ground into pieces by a mechanical blender (Model 890-68, Oster Co., Wisconsin, USA) prior to extraction and purification by QuEChERS column. The QuEChERS purification technique in combination with GC–MS identification and quantitation offers a fast, selective, efficient and precise method for determination of PAHs in high-fat smoked products (Kao et al., 2012; Chen et al., 2013). In brief, a 5-g ground meat or seafood sample was poured into a tube and mixed with 10 mL of deionized water for homogenization for 1 min, after which 10 mL of acetonitrile was added and the mixture shaken vigorously for 1 min. Then, the mixture was poured into a QuEChERS column containing 6 g of $MgSO_4$ and 1.5 g of sodium acetate, followed by shaking the column for 1 min and centrifuging at 4000 rpm for 5 min. Next, 6 mL aliquot was collected from the supernatant and transferred into a QuEChERS clean-up column containing 400 mg of primary secondary amine (PSA), 1200 mg of $MgSO_4$ and 400 mg of endcapped octadecylsilane silica gel particles for purification. The eluates were then centrifuged at 4000 rpm for 5 min and 1-μl aliquot from the supernatant was injected into GC–MS for PAHs analysis.

2.5. GC–MS analysis

Identification and quantitation of PAHs in meat and seafood samples were performed by employing a GC–MS method (Kao et al., 2012; Chen et al., 2013). An Agilent 30-m HP-5MS column (0.25 mm I.D., 0.25 μm film thickness) was used to separate 16 PAHs standards and various PAHs in meat and seafood samples. However, owing to the matrix complexity in meat samples, a 5-m guard column was installed before analytical column to extend the column life. Samples were injected in a splitless mode with helium as carrier gas at a flow rate of 1.0 mL/min and the GC operation condition for PAHs separation is shown below: injector temperature 290 °C, oven temperature at 70 °C initially, increased to 195 °C at 15 °C/min and maintained for 2.5 min, further raised to 240 °C at 15 °C/min, holding for 17 min, increased again to 270 °C at 5 °C/min, and finally to 310 °C at 15 °C/min with a holding time for 10 min. Following this approach, a total of 16 PAHs standards were adequately resolved within 40 min. For MS condition, the interface temperature was set at 270 °C, electron multiplier voltage at 70 eV, and detection by selected ion monitoring (SIM) mode based on elution order or retention time as well as specific

Table 1
Kindling-free-charcoal grilling conditions.

Meat product	I[a]		II	
	Internal Temp. (°C)	Grilling Time (min)	Internal Temp. (°C)	Grilling Time (min)
Poultry				
Chicken heart	85	13	100	26
Chicken drumstick	74	20	88	40
Chicken gizzard	83	8	88	16
Chicken breast	75	15	84	30
Duck drumstick	78	20	85	40
Red meat				
Beef steak	68	16	87	32
Pork knuckle	81	23	94	46
Ham	76	6	89	12
Lamb steak	77	6	88	12
Pork chop	80	8	99	16
Seafoods				
Oyster	78	10	83	20
Octopus	72	40	85	80
Salmon	72	9	82	18
Shrimp	74	5	100	10
Squid	80	10	90	20

[a] The grilling condition was selected based on USDA recommendation.