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What is This?



# Detection of polycyclic aromatic hydrocarbons in different types of processed foods

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#### Abstract

Polycyclic aromatic hydrocarbons (PCAHs), particularly those with a high molecular weight, have been classified as probable carcinogens to humans. The aim of the present study is to determine the levels of PCAHs in samples of meat, fish, chicken, fried potato, and toasted bread, which will be thermally processed using conventional and microwave ovens. Different samples will be collected and analyzed for five PCAHs including pyrene, benzo(a)anthracene, benzo(e)pyrene, benzoflouroanthene, and benzo(a)pyrene. The analytical method involves saponification with methanolic potassium hydroxide, extraction with cyclohexane, and determination by high-performance liquid chromatography. The obtained results showed that there is a variation in the detected PCAHs in different foodstuffs. Fried potato processed by conventional oven or microwave oven showed none of the selected studied PCAHs. It was found that, chicken showed higher content levels of total PCAHs than the meat and fish. Data are the highest mean concentrations of fluoranthene and benzo(a)pyrene but within low limit. The obtained results were compared with international permissible levels to avoid pollution, which may cause hazardous effects on individual and society.

#### **Keywords**

PCAH, foodstuff, processing

# Introduction

Polycyclic aromatic hydrocarbons (PCAHs) comprise the largest class of chemical compounds known to be cancer-causing agents. Some, while not carcinogenic, may act in synergistic way. PCAHs are being found in water, air, soil, and, therefore, also in food. They originate from diverse sources such as tobacco smoke, engine exhausts, petroleum distillates, and coal-derived products. PCAHs may also form directly in food as a result of some heating processes like charcoal grilling (Lawrence and Weber, 1984), roasting (Lo and Sandi, 1978), and smoke drying (Lodovici et al., 1995).

PCAHs are the compounds with two or more fused benzene rings produced by incomplete combustion of organic substances involved in natural and anthropogenic processes. It has been well established that PCAHs have carcinogenic, mutagenic, and teratogenic effects (Miller and Miller, 1993). Exposure to these compounds is a public health concern particularly in children who are one of the most susceptible groups of the population. Moreover, exposure to genotoxic carcinogenic compounds at a young age may represent a health risk, that is, causing genetic damage (mutation, sister chromatid exchanges, and other genetic disruption; Phillips, 1999) that may increase the risk of cancer later in life (Speer et al., 1990).

PCAHs are reported to disturb the antioxidant defense system and are responsible to induce oxidative stress. It is well known that PCAHs are not known to exhibit acute symptoms; metabolic activation of PCAHs by cytochrome P450 1A1-catalyzed reactions

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Said S Moselhy, Biochemistry Department, Faculty of Science, Ain Shams University, Cairo 22015, Egypt Email: moselhy6@hotmail.com generates electrophilic metabolites and other reactive oxygen species (ROS), which tends to bind covalently with DNA and also cause interference with cell homeostasis (Chen and Chen, 2001). Increased ROS generation or state of oxidative stress has been shown to be linked with a lot of pathophysiological changes like asthma, chronic obstructive pulmonary disease, cystic fibrosis, juvenile rheumatoid arthritis, cholestatic liver diseases, and diarrheal diseases (Chen and Chen, 2003) in children.

Contamination of foodstuffs by PCAHs can occur during smoking and during particularly intense thermal processing (toasting, roasting, frying, etc.). Intense thermal processes can be applied to foods in an indirect or direct way. When the thermal agent or smoke does not enter in direct contact with food, an indirect thermal processing is used. Electric oven toasting could be an example of this process. A direct contact between the thermal agent or smoke and food takes place in direct thermal processes like barbecues and gas oven toasting. Contamination of PCAHs by intense thermal processing occurs due to the generation of food nutrients by direct pyrolysis and also due to direct deposition of PCAHs from smoke produced through incomplete combustion of different thermal agents (Devanesan et al., 1999). Food, water, and air are the main exposition routes to PCAHs (Dennis et al., 1983). Food accounts for the 99% being smoked foods and those submitted to severe thermal treatments.

Bread toasting includes an intense thermal process, which can be applied by direct (flame-toasting, coal-grilling, or gas oven-toasting) or indirect (electric oven-toasting) way. Although commercial bread toasting is performed at 220–250°C in an electric oven, higher temperatures could be easily reached by consumers at home (Dennis et al., 1991).

Microwave heating refers to the use of electromagnetic waves of certain frequencies to generate heat in a material. Microwave food processing uses the two frequencies 2450 and 915 MHz. Of these two, the 2450 MHz frequency is used for home ovens and both are used in industrial heating (Duedahl-Olesen et al., 2006).

Heating with microwave involves primarily two mechanisms: dielectric and ionic. Water in the food is often the primary component responsible for dielectric heating. Due to their dipolar nature, water molecules try to follow the electric field associated with electromagnetic radiation, as it oscillates at the very high frequency of the microwave. Such oscillation of the water molecules produces heat. The second major mechanism of heating with microwaves is through the oscillatory migration of ions in the food that generates heat under the influence of the oscillating electric field. Microwave heating for pasteurization and sterilization is preferred to conventional heating for the primary reason that they are rapid and therefore require less time to come up with the desired process temperature. This is particularly true for solids and semisolid foods that depend on the slow thermal diffusion process in conventional heating. They can approach the benefits of high temperature short time processing whereby bacterial destruction is achieved, but thermal degradation of the desired components is reduced (Fazio and Howard, 1983).

Microwave ovens heat foods through a process of creating molecular frictions, but this same molecular friction may quickly destroys the delicate molecules of vitamins and phytonutrients (plant medicines) naturally found in food. As a result of intense thermal processes, partial carbonizations could take place (García-Falcon et al., 1996).

The aim of the present study is to determine the levels of selected PCAHs including pyrene, benzo(a)anthracene, benzo(e)pyrene, benzoflouroanthene, and benzo(a) pyrene in some commercial foods (fried potato, fish, chicken, meat, and toasted bread) processed by different processing methods by conventional oven or microwave oven.

# Materials and methods

#### Samples

Samples of fried potato, fish, chicken, meat, and toasted bread (500 gm for each) were processed by oven or microwave. All samples for each type were combined, and the mixed samples were homogenized with no addition of fluids. The homogenates were kept frozen at  $-20^{\circ}$ C until analysis.

#### Reagents

*Polycyclic aromatic hydrocarbons.* Ten PCAHs selected for analysis, including pyrene, benzo(a)an-thracene, benzo(e)pyrene, benzoflouroanthene, and benzo(a)pyrene, were purchased from Sigma.

#### Methods

*Extraction.* A homogenized portion of 100 g of each sample was boiled under reflux with 400 ml of a 2-M solution of potassium hydroxide in methanol for 2 h.

Sample PCAHs	Fried potato	Fish	Chicken	Meat	Toasted bread
Pyrene	ND	ND	0.09	ND	ND
Benzo(a)anthracene	ND	0.003	0.06	ND	ND
Benzo(e)pyrene	ND	0.09	0.03	0.08	ND
Benzo(a)pyrene	ND	0.01	0.02	0.05	0.04
Benzoflouranthene	ND	0.01	ND	0.08	ND

**Table 1.** The levels of PCAHs in different foods processed by conventional oven  $(\mu g/100g; \text{ mean } \pm \text{ SD})^a$ 

PCAH: polycyclic aromatic hydrocarbon; ND: not detected.

<sup>a</sup>Values are average of three assays.

The saponified material was transferred into a 500-ml separating funnel. The flask was rinsed with 400 ml methanol/distilled water (9:1, v/v). The mixture was extracted twice for 2 min with 600 ml cyclohexane. The organic layer was first washed with 400 ml methanol:water (1:1, v/v) and finally with 100 ml distilled water. Then the organic layer was transferred in to a round-bottom flask. The volume of the samples was reduced to 50 ml at 40°C using a vacuum rotary evaporator. The cyclohexane was extracted with three aliquots of N,N-dimethylformamide-water (9:1, v/v), then the dimethylformamide extract was diluted with 100 ml of a 1% sodium sulfate solution. The combined solution is dried and concentrated on a rotary evaporator to 10 ml at 40°C (García-Falcon et al., 2005a).

# Analysis by HPLC and GC/MS

Analysis was carried out using Waters (USA) highperformance liquid chromatography (HPLC) delivery system with fluorescence detector (excitation wavelength 290 nm and emission wavelength 430 nm). A C18 column, a mobile phase composed of acetonitrile: water (75:25, v/v), at a flow rate of 1 ml/min, was used to separate the PCAHs (García-Falcon et al., 2005b).

### Peak identity

The identification of PCAHs was conducted on a Shimadzu (USA) gas chromatography/mass spectrometer (GC/MS), which was used with helium carrier at 0.7 ml/min. The GC oven temperature program was as follow: 120°C for 3 min, 5°C/min to 280°C held for 26 min. Total run time was 40 min. Peak spectra were compared with the mass spectra of PCAHs standards and library supplied with the instrument. The MS detector shows a typical HPLC chromatogram of a PCAHs standard solution and an extract of sample (García-Falcon and Simal-Gándara, 2005).

#### Statistical analysis

To verify significant differences in relation to species, size, and place of capture, the results were submitted to analysis of variance at 5% level of confidence. The results for the principal fatty acids, total lipid, and cholesterol were also submitted to principal component analysis, using SPSS Version 10.

#### Results

Results obtained (in Tables 1 and 2) show the concentrations of PCAHs found in different foodstuffs processed by conventional oven or microwave oven. There is a variation in the level of PCAHs in different foodstuffs. Fried potato processed by conventional oven or microwave oven showed none of the selected studied PCAHs. Chicken showed higher content level of total PCAHs than meat and fish. The data collected showed the highest mean concentrations of fluoranthene and benzo(a)pyrene but within low limit.

# Discussion

In the present study, five selected PCAHs were detected in different foodstuffs processed by different methods. It is well known that commercial bread toasting is performed between 200 and 260°C but higher temperatures could be reached easily. PCAH generation has been evaluated under temperature conditions between 300 and 700°C. At this point, a new matrix appears because of the activated carbon generation in toasted bread samples.

The samples analyzed were processed together with a blank to test for the background PCAH levels in the foodstuffs. Bread samples were processed by means of conventional oven and microwave oven. Results obtained by oven toasting showed that PCAH pollution is mainly due to the pyrolysis of macronutrients (carbohydrates, lipids, and proteins). Commercial bread toasting is performed at 200–260°C.

Sample PCAHs	Fried potato	Fish	Chicken	Meat	Toasted bread
Pyrene	ND	0.009	0.01	ND	ND
Benzo(a)anthracene	ND	0.06	0.03	ND	ND
Benzo(e)pyrene	ND	0.09	0.001	0.11	ND
Benzo(a)pyrene	ND	ND	ND	0.05	ND
Benzoflouranthene	ND	ND	ND	0.08	ND

**Table 2.** The levels of PCAHs in different foods processed by microwave oven  $(\mu g/100g; \text{ mean } \pm \text{ SD})^a$ 

PCAH: polycyclic aromatic hydrocarbon; ND: not detected.

<sup>a</sup>Values are average of three assays.

It would be pointed out that at the beginning of chromatogram, benzo(a)pyrene was the only PCAHs detected in oven. Evidence exists of lipid pyrolysis, which produces higher PCAH levels. With regard to toasting oven, the grilling of bread in the flames of a log fire produces the higher PCAH levels, followed by charcoal grilling and grilling in the flames of a gas source (Kazerouni et al., 2001). When toasting is carried out directly, PCAH pollution is generated by the pyrolysis of macronutrients and also by PCAH deposition from smoke of combustion.

Because the pyrolysis of macronutrients does not contribute to increased PCAH levels notably, PCAH deposition from smoke of combustion would be the most important source of PCAHs in the bread.

The levels of PCAH in smoke depend on heat source (coal, wood, gas, etc.), temperature, flame intensity in flame combustion, particulate material generated during combustion, etc. (Larsson et al., 1983). Of the selected heat procedures in this study.

Several authors determined the effects of various processing methods, such as steaming, roasting, smoking, charcoal grilling, etc. on foods (Lintas et al., 1979). All mentioned authors attribute to the highest PCAH generation during grilling or barbecue through pyrolysis of meat products and either deposition and penetration of smoke components into foods. Moreover, they found a link between fat foods and PCAH levels. The melted fat from the heated meat drips onto the hot coals and is pyrolyzed, giving rise to PCAHs generation, which are then deposited on the meat surface as the smoke rises.

Larsson et al. checked different methods of cooking meat products and the results revealed that the grilling of frankfurters in the flames of a log fire resulted in extremely high PCAH levels, up to 212 lg/kg benzo(a)pyrene, depending on the fat content of the food. Frying or electrical broiling does not lead to the production of PCAHs. Chen and Lin detected that charcoal grilling of duck samples with skin contained the highest amount of total PCAHs, smoking contained the highest amount, followed by charcoal grilling and roasting.

The highest levels of benozo(a)pyrene were found in very well done chicken steaks and meat and in microwave. Wu et al. detected fluorene, phenanthrene, anthracene, benzo[a]anthracene, chrysene, and benzo[k]fluoranthene in a range between 14 and 54  $\mu$ g/ kg in rougan, a traditional barbecued food. Mottier et al. analyzed PCAHs in barbecued meat sausages, determining the concentration levels below the quantification limit in all the products.

Samples of commercial-toasted bread were obtained from stores in Jeddah in order to determine PCAHs contents. Two pollution sources could be attributed to PCAH generation in commercially toasted bread, PCAH contamination in raw materials (at source), or during thermal processing.

naphthalene, The detected acenaphthalene, phenanthrene, and dibenzo[a,h]anthracene in all bread samples detected within the normal range of 0.32-9.4 µg/kg toasted bread. PCAHs in commercial toasted bread. Only fluorene, phenanthrene, anthracene, fluoranthene, and chrysene were analyzed in a range of concentrations between 7.4 and 18 lg/kg for anthracene and fluoranthene, respectively (Dennis et al., 1991). Some authors have verified higher PCAH levels in products that contain granary flour (Dou Abdul et al., 1997). Total PCAHs at 5.6 lg/kg were obtained in products with granary flour and 1.5  $\mu$ g/kg with white flour. These authors also proved that fats and oils contained the highest PCAHs levels due to their lipophilic character.

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