# THE IMPACT OF THE THERMAL PROCESS APPLIED ON POLYCYCLIC AROMATIC HYDROCARBONS IN WILD BOAR MEAT

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#### Abstract

Thermal processes applied to meat significantly influence its chemical composition. Thus, meat is considered an essential food due to its high nutritional value, good digestibility and appreciated culinary quality. PAHs in preserved products vary significantly, due to differences in preservation processes, chemical composition of the product, fat and water content. These organic compounds are formed from aromatic nuclei combined during pyrolysis or incomplete combustion of organic matter. The HPLC method was used to analyze polycyclic aromatic hydrocarbons. Following the study conducted on wild boar meat, it was found that all PAHs had the lowest values in the case of the boiling process. The highest average content was represented by the following hydrocarbons: phenanthrene (95.43 µg/kg) followed by naphthalene (46.55 µg/kg), fluoranthene (18.16 µg/kg), pyrene (10.24 µg/kg) and chrysene (9.78 µg/kg), all identified in wild boar meat (in sunflower oil). The aim of the study was to analyze polycyclic aromatic hydrocarbons in wild boar meat under the influence of thermal processes and under the influence of the vegetable (sunflower oil) and animal (lard) sources used for cooking.

Key words: lard, oil, PAH, thermal processes, wild boar meat.

# INTRODUCTION

The food chain is the main source of contamination with polycyclic aromatic hydrocarbons (PAHs), a statement supported by research in the specialized literature (Duedahl-Olesen & Ionas, 2021).

Game meat is a main source of protein that is increasingly sought after nowadays, in many developed countries including Europe (Vidrean et al., 2024).

An inadequate culinary education leads to the formation of polycyclic aromatic hydrocarbons both in meat from domestic animals and in meat from game animals (Lee et al., 2016). PAHs are considered to be carcinogenic compounds for humanity, with a high degree of triggering of oncological diseases but also of other diseases associated with this disease (Diggs et al., 2011). According to the existing the specialized literature, beanzo[a]pyrene, is a compound that is part of category and b, being considered carcinogenic, capable of producing both

genotoxic and mutagenic effects (Ramesh et al., 2004; Mastan et al., 2023). Currently, there are various concepts regarding the thermal processes used to reduce the formation of PAHs in meat (Ciecierska et al., 2023).

The correct selection of oil or lard used, as well the temperature and humidity are essential points to have an acceptable concentration of PAHs (Diggs et al., 2011; Vidrean et al., 2024). Levels of PAHs are detectable in all internal organs according to data from the specialized literature, and especially in organs with rich adipose tissue, these being considered a constant reservoir for the gradual elimination of PAHs (Ciecierska & Obiedzinski, 2007).

PAHs have the following essential characteristics: high molecular mass, at room temperature they become solid, are insoluble in water, can be oxidized and degraded in the presence of light and thus have a lower molecular mass (Duedahl-Olesen & Ionas, 2021; Mastan et al., 2023).

Contaminants such as PAHs represent an important family of toxic substances, which are

also ubiquitous in the environment, and human contamination can also be achieved by inhaling cigarette smoke and the most important source of contamination being the dietary intake of contaminated food due to incorrect thermal processing (Kazerouni et al., 2001; Djinovic et al., 2009; Vidrean et al., 2024). These contaminants can reach the body including through contaminated air due to exhaust gases produced by cars or industry (Ramesh et al., 2004).

PAHs constitute a class of pollutants resulting from the process of incomplete combustion of organic matter (Nielsen et al., 1996; Ferhadian et al., 2010; Savin et al., 2024). Several types of meat that are subjected to thermal cooking processes, especially smoking and grilling, trigger the formation of PAHs (Hitzel et al., 2013; Olatunji et al., 2022). High heat processes through the use of the grill or through the use of coal or wood can lead to an increase in the concentration of PAHs (Barranco et al., 2004; Ifeyinwa et al., 2024).

In the case of the roasting process, the meat may present levels higher than 10-20 pg/kg, Lee et al., 2016, states that these compounds can have many serious effects on the body (genotoxicity, carcinogenicity, immunotoxicity, teratogenicity). The major role of meat processing is to improve its quality, by changing the sensory properties and helping the digestion process, including obtaining a longer shelf life of meat-based preparations (Biesalski, 2005; Wretling et al., 2010). Considering the increasing consumption of meat, it is mandatory to study the degree of meat contamination due to thermal processes and the way in which they are carried out (Halagarda & Wojciak, 2022).

## MATERIALS AND METHODS

The samples in the case of this study were represented by meat from the wild boar from the Transylvania region. The aim of this study was to determine the level of PAHs in wild boar meat after applying the thermal process of frying, boiling and grilling. Animal lard and sunflower vegetable oil were used in the thermal frying process. The wild boar meat was cooked as follows: grilled, at a temperature of 190°C, for 50 minutes; fried in oil and lard at a temperature of 180°C, for 40 minutes; boiled at a temperature of 120°C, for 30 minutes.

To determine the level of PAHs, chromatographic method was applied, using the Perkin-Elmer HPLC system. The degassing system used was: model 200 Manufacturer Perkin-Elmer, binary pump, automatic sample introduction system: model Flexar. With the help of the thermostat (model Spark), temperature control is ensured in the range of 10°C-90°C. The fluorescence detector (200 series; Manufacturer Perkin-Elmer), has a double monochromator - excitation emission; the wavelength range is: excitation 200-850 nm; emission 250-900 nm. The chromatographic columns used: ZORBAX Eclipse PAH: Manufacturer Agilent Technologies with column dimensions of 4.6×150 mm, diameter of 5 μm. The principle of the method consists in the use of highperformance liquid chromatography (HPLC), with fluorescence detection after the solidliquid extraction stage to determine the 15 PAHs in wild boar meat. Interferences are represented by asymmetric peaks or peaks that are wider than those corresponding to the reference compound.

Cases of interference can exist, for example, in the case of naphthalene and phenanthrene. Reagents used are of recognized analytical quality. We used blank samples to guarantee that the reagents do not contain PAHs in detectable concentrations. The working method includes three important stages: the extraction stage, the measurement stage and the stage of expressing the results. The extraction stage includes: the exact weighing of 10 grams of wild boar meat which is homogenized with the help of a blender, then we added 50 ml of 0.4 M KOH solution in ethanol and water (9:1) for saponification, which is carried out in an ultrasound bath for 30 minutes. Filtration is carried out with the help of filter paper, the obtained product was extracted twice with 15 ml of cyclohexane, then the obtained supernatant was purified on a Florisil column, followed by evaporation to dryness under nitrogen flow, and finally it is brought back with 1 ml of acetonitrile. Before the injection process, the samples will be filtered on 0.45 µm cartridges. For the measurement step, a gradient and wavelength program was used to identify the 15 PAHs. To express the results, we used a calculation formula that includes

several elements: the concentration read by HPLC, the dilution factor, the mass of the sample in grams and the % recovery.

### RESULTS AND DISCUSSIONS

Meat and meat products are widely consumed, especially smoked and grilled (Ciecierska & Obiedzinski, 2007). The data from the specialized literature are still not sufficient about the acceptable levels of PAH, therefore a more detailed investigation of the meat subjected to different thermal cooking processes different species of animals is needed, considering the high risk of affecting the human body. Figures 1-5 show the average values for polycyclic aromatic hydrocarbons analyzed from wild boar meat depending on the treatment applied and the fat source (sunflower oil and pork lard). Other studies were conducted by: Mastan et al. (2023), who evaluated the influence of heat treatment on PAH in fish: the impact of wood used for smoking and fish species on PAH content in smoked fish (Savin et al., 2024); the content of polycyclic aromatic hydrocarbons (PAH) in chicken, turkey and duck meat after exposure to traditional smoking with different types of wood and different periods of smoke exposure (Coroian et al., 2023).

According to the graph above (Figure 1), the highest average value is represented by naphthalene in the game meat sample through the thermal process of frying in sunflower oil.

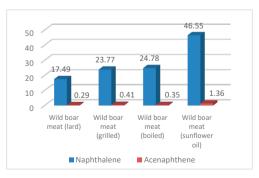


Figure 1. Naphthalene, acenaphthalene from wild boar meat (in lard), wild boar meat (grilled), wild boar meat (boiled), wild boar meat (in sunflower oil) (µg/kg)

Acenaphthene shows lower average values compared to naphthalene in all thermal processes used for game meat (Figure 1). The

lowest average values for both indicators are found in the game meat sample subjected to the frying process in lard, and the highest values are in the case of the sample subjected to the sunflower oil frying process (Figure 1).

In the study carried out by Viegas et al. (2012), the authors found that meat subjected to grilling, using charcoal for fire, observed that maintaining stable combustion leads to a decrease in PAHs. One of the most important triggering factors that lead to the production of PAHs through the grilling process, is the smoke that comes from the incomplete burning of the fat that comes into contact with the flame (Viegas et al., 2012).

According to the study carried out by Rose et al. (2014) the authors of the study draw attention to the consumption of food on the grill, because enormous amounts of PAHs can be formed, which are consumed in a single meal. In the case of salmon grilled with coconut shell charcoal, the authors of the study concluded that the levels of PAHs showed a significant decrease (Rose et al., 2014). Grilling with charcoal and wood chips leads to the formation of benzo(a)pyrene in most food samples subjected to the thermal grilling process (Rose et al., 2014). Food that is cooked closer to the heat source leads to an increase in the level of PAHs, and the cooking time can lead to a moderate increase in the level of PAHs (Rose et al., 2014; Onopiuk et al., 2021). The lowest average value of fluorene can be observed in the case of the sample subjected to the boiling process, with a value of 7.96 µg/kg, compared to phenanthrene, where the lowest average value is in the case of the sample subjected to the process of frying in lard, with an average value of 16.71 µg/kg (Figure 2).

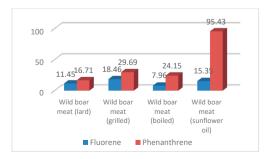


Figure 2. Fluorene, phenanthrene from wild boar meat (in lard), wild boar meat (grilled), wild boar meat (boiled), wild boar meat (in sunflower oil) (μg/kg)

The following factors can contribute to the increase or decrease of PAH depending on the management mode: the heat source, the type of grill used horizontally/vertically, the species from which the meat comes, as well as other parameters, for example the degree of maturation of the meat, marinating or even the addition of spices (Duedahl-Olesen & Ionas, 2021).

The results of the study carried out by Khan et al. (2008), confirm the fact that PAHs are both pyrogenic and petrogenic.

Ifeyinwa et al. (2024), detected the following values regarding the concentrations of PAHs: total PAHs (mg/kg) in the samples of game meat subjected to the smoking process obtained the following values  $2.763 \pm 0.185$  mg/kg, in the mackerel meat subjected to the smoking process they obtained  $1.618 \pm 0.102$  mg/kg and in the catfish meat subjected to the process of smoking obtained  $1.718 \pm 0.129$  mg/kg, while the values in chicken meat subjected to the smoking process were  $0.771 \pm 0.049$  mg/kg.

Maximum average of  $222.7 \pm 13.2 \,\mu\text{g/kg}$ , of the 16 PAHs studied, was detected in the fish samples subjected to the smoking process, and a minimum average of  $112.9 \pm 7.2 \,\mu\text{g/kg}$ , in the case of chicken kebab, for the same studied PAH (Ifeyinwa et al., 2024).

Anthracene and fluoranthene show the highest values according to Figure 3, in the case of the sample subjected to the frying process in sunflower oil. The average values for these two indicators are low for the rest of the processes, namely: fried in lard, grilled and boiled (Figure 3).

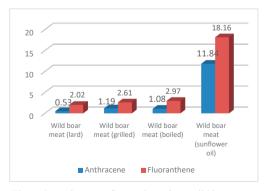


Figure 3. Anthracene, fluoranthene from wild boar meat (in lard), wild boar meat (grilled), wild boar meat (boiled), wild boar meat (in sunflower oil) (µg/kg)

The results of a study carried out in the city of Togo on samples of smoked fish, indicate concentrations above the permissible limit for benzo[a]pyrene (BaP), benz[a]anthracene, (BaA), benzo[b]fluoranthene (BbF) and chrysene (CHR); The daily dose can range from 23.22 to 130.09 ng/kg/day for the four PAHs and from 1.4 to 31.64 ng/kg/day for BaP. (Bouka et al., 2020).

An experiment carried out in Estonia, to determine the level of PAHs, from meat samples from the trade respectively grilled meat samples, after the study the results indicated values that exceeded in 3.4% of cases the limit allowed for the benzo[a]pyrene indicator (Reinik et al., 2007).

A study carried out on samples of smoked sausages, pork bacon, ham, grilled pork and poultry from Cyprus, the authors observed the highest concentrations of PAHs in the samples that were kept in the thermal process for a longer time and that have a higher fat content, also in this study it was specified that 12% of the products subjected to the smoking process and 15% of the products subjected to the thermal process at grill have exceeded the limits allowed by the legislation of the European Union (Kafouris et al., 2020).

The highest average value in the case of the benzo[a]anthracene indicator is in the meat sample subjected to the frying process in sunflower oil, with a value of  $3.29 \mu g/kg$ , and the lowest average values are in the case of the boiling and frying lard (Figure 4).

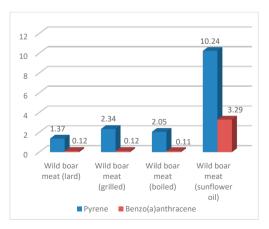


Figure 4. Pyrene, benzo[a]anthracene from wild boar meat (in lard), wild boar meat (grilled), wild boar meat (boiled), wild boar meat (in sunflower oil), (µg/kg)

Choosing an appropriate type of thermal process, respectively the raw material with the lowest fat content, can be an effective option to reduce the amount of PAHs inside meat preparations (Ciecierska et al., 2023).

We find the highest average values for chrysene, benzo[k]fluoranthene, benzo[a]pyrene, dibenzo[a,h]anthracene, benzo [ghi]perylene

and indeno[1,2,3-cd], in the case of the process of frying the venison sample in sunflower oil, according to Figure 5. The undetectable values were in the case of the following indicators: benzo[b]fluoranthene, benzo[a]pyrene, dibenzo[a,h]anthracene and benzo [ghi]perylene from the samples subjected to the processes frying in lard, boiled, grilled and frying in sunflower oil (Figure 5).

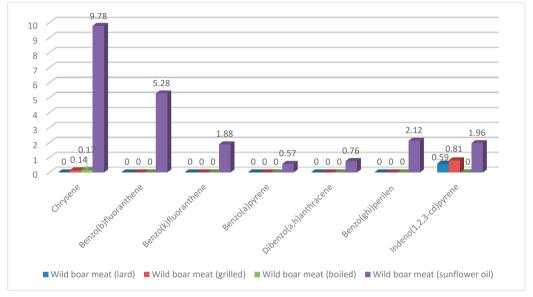


Figure 5. Chrysene, benzo[b]fluoranthene, benzo[k]fluoranthene, benzo[a]pyrene, dibenzo[a,h]anthracene, benzo[ghi]perylene and indeno[1,2,3-cd], pyrene from wild boar meat (in lard), wild boar meat (grilled), wild boar meat (boiled), wild boar meat (in sunflower oil) (µg/kg)

### **CONCLUSIONS**

Game meat, in this case wild boar, can contain different concentrations of PAHs, with the highest average values in the frying process in sunflower oil, according to our study. The highest average value for benzo[a]anthracene is in the case of frying in sunflower oil. It is remember essential to that consumption of game meat subjected to various thermal treatments, which give the average values of PAHs above the limits allowed by the legislation, over a prolonged period of time, may present a possible carcinogenic risk for consumers. The method and technique chosen for preparing the meat is important. The consumer is concerned about the special sensory value that is obtained as a result of the

various thermal processes of meat preparation, but these compounds that result from the thermal processes and which are obviously not friendly to the human body must be taken into account.

Among the PAHs studied by us, the highest values are for acenaphthalene with an average value of 46.55 ( $\mu$ g/kg), phenanthrene with an average value of 95.43 ( $\mu$ g/kg), fluoranthene with an average value of 18.16 ( $\mu$ g/kg), pyrene with an average value of 10.24 ( $\mu$ g/kg) and chrysene with an average value of 9.78 ( $\mu$ g/kg), all these indicators being with higher average values in the case of game meat subjected to the frying process in sunflower oil compared to the other three processes used, boiling, frying in lard and grilling, where the average values were lower.

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