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## Polycyclic Aromatic Hydrocarbon Content of Breast and Leg's Meat of Indonesian Native Duck

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**Abstract:** Duck's leg and breast of Indonesian native duck were analyzed in laboratory to determine polycyclic aromatic hydrocarbon (PAH) such as *naphthalene*, *acenaphtene*, *phenentrene*, *flourantene*, *pyrene*, *benzoantrasene* and *perylene*. The result showed that *naphthalene*, *flourantene*, *pyrene*, *benzoantrasene* and *perylene* in leg's duck was higher than breast duck. Total fat of legs (5.87%) were higher than breast (3.31%). This result showed that total fat of raw duck meat influence PAH formation. Antioxidant activity of leg was higher than breast's duck (8.79% : 5.1%). This result showed that endogenous antioxidant did not able to prevent PAH formation.

**Key words:** PAH, duck meat, antioxidant

### INTRODUCTION

Duck meat is a source of fat and protein for humans. Indonesia have many native duck's species such as Tegal, Magelang, Mojosari, Bali and Alabio. The most part of Indonesian native ducks are layer, but when egg's production were low they were used as broiler.

Indonesian people are very interested in duck's meat which is cooked in charcoal grilling. This cooking method can conduce many dangerous compound. PAH is one of potential carcinogenic compound which are formed in charchoal grilling methode of duck's meat. Lin *et al.* (2011) reported that Benzo (a) pyrene alone, or the total amount of major PAHs with carcinogenic potential for humans, in Peking duck samples, were detected at levels of 8.7 and 54.7 Lg/kg, respectively.

The availability of high fat and protein results in duck meat is susceptible in oxidation. Oxidation process can lead to the emergence of various novel compounds that are harmful to human health are known as free radicals. Free radical is a molecule or a compound containing one or more unpaired electrons in its outer orbital. The existence of unpaired electrons cause the highly reactive compounds looking for a partner in an offensive way and electron binding molecules in the surrounding areas (Pokorny *et al.*, 2001).

Oxidation can be prevented by antioxidant compounds, so that the formation of harmful compounds do not occur. According to Huang, Ou and Prior (2005), antioxidant is a substance in foods that significantly decreases the adverse effects of reactive species, such as reactive oxygen and nitrogen, on normal physiological functions.

An oxidant is an electron-accepting compound or compounds that can attract electrons. Fresh duck meat

contains antioxidant compounds in very small quantities, so it is not able to prevent oxidation at ambient temperature. Some compounds are classified as endogenous antioxidant such as tocopherol, carnosine, lipoic acid and some enzyme systems (Decker and Mei, 1996).

The content of antioxidant compounds are affected by the availability of food components, so the difference in the amount of fat or protein can lead to differences in the antioxidant content. The main targets of antioxidant are proteins, unsaturated fatty acids and lipoproteins and DNA elements including carbohydrates (Surai, 2002). Oxidation of fatty acids produce hydroperoxides and aldehydes that contributes to the occurrence of protein oxidation (Estevez *et al.*, 2009).

Antioxidants can neutralize free radicals become harmless compounds. Polycyclic aromatic hydrocarbons (PAH) is one of the potentially cancerous compounds that formed from food processing. According to Chung *et al.* (2011), charcoal produces higher PAH than roasted and grilling. Hydrophobic components of PAH consisting of 2 or more aromatic chain. PAH is an environmental pollutant that can be formed during food processing (Agerstad dan Skog, 2005). The formation of PAH in grilled meat allegedly caused by contaminated smoke and charcoal combustion.

Duck's leg and breast are part of duck meat which is commonly consumed by Indonesian people. Several previous studies have proved that the leg and breast containing different total fat, thus allegedly antioxidant content is also different. The difference of raw meat antioxidant activity can result the difference in PAH content of grilled meat.

The research aims to compare PAH content between leg and breast meat of duck. The result of this research will be necessary to correct cooking methods of duck's meat. The healthy duck's meat can be produced by right methods of cooking.

## MATERIALS AND METHODS

**Material:** About 24 Tegal's male ducks were slaughtered and cutted become leg and breast part. Meat and skin of each part were mixed and homogenized so that in order to obtain the smooth mixture.

**PAH determination:** The Gas Chromatography (GC) instrument (Shimadzu mod. GC-17A), mass spectrometer (Shimadzu, quadrupole detector mod. GC-MS-QP5000) equipped with an acquisition data system (Shimadzu, CLASS 5000). The MS detector could be operated in two different modes: scanning (TIC) and selected ion monitoring (SIM). The data were acquired operating in the SIM mode that allows quick identification and quantification of the preselected ion peaks. On the other hand, non-preselected peaks (e.g. interferences) are not quantified. SIM is a more sensitive technique for trace quantitative analysis and can result in as much as a 500-fold increase in sensitivity than TIC.

The used column was an Equity-5 (30 m 0.25 id, 0.5 mL) fused-silica capillary column from Supelco (Milano, Italy) that had a high efficiency, thus the components of environmental or food extracts could be much better separated than by using HPLC. Ultra pure (99.999%) helium was used as a carrier gas (20.6 mL/min). The 1 L solutions of the extracts were injected in the splitless mode at 0.61 min split delay. The injection of both extracts from samples and standard solutions was performed by hand. The use of an injector equipped with electronic pressure control allows the maintenance of a constant flow rate during the entire separation. The injector temperature was maintained at 280°C.

The GC temperature program was: from 40°C (2 min) to 100°C at 40°C/min, to 200°C at 10°C/min, to 325°C (8 min) at 30°C/min. Identification of the components of the standard mixture was carried out by comparing retention times for each component of the mixture with those of pure components analyzed under the same experimental conditions. Identification was confirmed by comparing the spectra of the single components with those stored in the acquisition system library (NIST).

The identification of PAHs in the solutions extracted from samples was carried out on the basis of previously determined retention times and confirmed by using previously acquired mass spectra. The content of each single analyte in the sample was quantified relatively to the perdeuterated PAHs added to the dry residue. The complete calibration for all the analytes was made by injecting, every 2-3 days, five solutions containing

analytes at known concentrations. Most of the analytes have response factor of the same order of magnitude. The response of the GC-MS instrument was checked every morning using a solution containing only four compounds.

**Sample extraction:** For analysis of possible traces of PAHs in the duck meat, preliminarily, some samples were extracted using a solid-phase (SPE) by means of SPE cartridges containing a hydrophobic sorbent. The tested sorbents were C18-silica (ENVI-18, 0.5 g, supplied by Supelco, Milano). All cartridges were conditioned with 5 mL of MeOH and 5 mL of water. PAHs were eluted using fractions of 2 mL of the eluting solvent. Applying this extraction method to two meat samples, we obtained lower recovery of analytes (30-60%) than that described below and used for analysis of 13 samples. A known volume (150 L) of the surrogate standard solution (anthracene-d10 and benz[a]anthracene-d12) was added to 15 mL of duck meat and the sample was digested for 3 h under reflux after addition of 50 mL of a methanolic solution of KOH (2 mol/L). After cooling and addition of 20 mL of water, the solution was liquid-liquid extracted three times with 10 mL of hexane.

**Antioxidant activity determination:** The DPPH assay was done according to the method of Thaipong *et al.* (2006). The stock solution was prepared by dissolving 24 mg DPPH with 100 mL methanol and then stored at -20°C until needed. The working solution was obtained by mixing 10 mL stock solution with 45 mL methanol to obtain an absorbance of 1.170.02 units at 515 nm using the spectrophotometer.

Fruit extracts (150 mL) were allowed to react with 2850 mL of the DPPH solution for 24 h in the dark. Then the absorbance was taken at 515 nm. The standard curve was linear between 25 and 800 mM Trolox. Results are expressed in mM TE/g fresh mass. Additional dilution was needed if the DPPH value measured was over the linear range of the standard curve.

**Experimental design:** Data were divided into two populations namely leg and breast part. PAH, total fat and antioxidant activity were measured as parameters. The results of measurements of the parameters on the leg were compared with breast part than presented descriptively.

## RESULTS

PAH content of duck's meat were presented in Tabel 1. TBA value and total fat of duck's meat were presented in Tabel 2.

Antioxidant activity of duck's meat were presented in Tabel 3.

Table 1: Availability of PAH in duck's breast and leg

| PAH Compound   | Breast (ng/g) | Leg (ng/g) |
|----------------|---------------|------------|
| Naphthalene    | 0.001         | 0.011      |
| Acenaphthene   | 0.001         | 0.001      |
| Phenanthrene   | 0.003         | -          |
| Flourantene    | 0.013         | 0.731      |
| Pyrene         | 0.002         | 0.003      |
| Benzoantrasene | -             | 0.194      |
| Perylene       | 0.002         | 0.012      |

Table 2: TBA value and total fat of duck's breast and leg

| Meat part | Total fat (%) | TBA value (mg malonaldehyd/kg) |
|-----------|---------------|--------------------------------|
| Breast    | 3.31          | 0.107                          |
| Leg       | 5.87          | 0.110                          |

Table 3: Antioxidant activity of duck's breast and leg

| Meat part | Antioxidant activity (%) |
|-----------|--------------------------|
| Breast    | 5.10                     |
| Leg       | 8.74                     |

## DISCUSSION

The differences of PAH content between leg and breast duck were affected by the differences of total fat. Some researchers suspect that the main precursor formation of PAH is fat. According to Alomirah *et al.* (2011), PAH formation is influenced by the type of heat source, duration of roasting, use marinating sauce and fat content of food.

The process of PAH formation was thought occur in three ways. The first way, pyrolysis organic compound such as fat, protein and carbohydrate. According to Bartle (1991), the greatest concentrations of PAHs have been shown to arise from pyrolysis of fat. Fat degradation would produced many free radicals which was precursor of PAH formation. The second way, contamination from combustion and smoke. The nutrients are able to bind to free radicals derived from the combustion and smoke. The highest concentration of PAHs was detected in charcoal grilled (Fahardian *et al.*, 2010). The third way, PAH was formed by fat melting in direct contact with the heat source. PAH was brought to the meat surface in the form of smoke.

PAH is known as carcinogenic compound that potentially become cancer in human body. This could be happened when PAH accumulated in human body for many years. Several countries had standard of minimum PAH consumption everyday. For average consumers across the European countries, dietary exposure for the sum of eight carcinogenic and genotoxic PAHs (PAH8) was estimated at 1.73 mg/day (EFSA, 2008).

Table 2 showed that TBA value of duck's leg were higher than breast. It explained that duck's leg meats were easier to oxide than breast. Further, duck's leg meat produced more free radical than breast. Duck's leg fat are higher than the breast. Total fat of meat is influenced by several factor during the duck lives. According to Baeza (2006), genotype, feed and age having the same effect on fat content. Besides speed of fat oxidation after cutting can affect the total fat. When fat was heated, it

would fallen down to charcoal with the result that more smoke was produced. Compounds derived from smoke were capable of forming PAH in meat.

Table 2 also showed that the antioxidant activities of duck's leg meat was higher than the breast. Differences in nutrient content between leg and breast meat is the cause of the differences in the value of antioxidant activity. Due to degradation of fats and proteins form new components that can act as an antioxidant. Data of total duck's leg fat and breast are presented in Table 2.

**Conclusion:** Leg's meat produced more PAH than breast. Endogenous antioxidants of duck's meat are not effective to prevent formation of PAH. Fat content is directly proportional to the presence of PAH in duck's leg and breast meat.

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