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Influence of the cold storage time of raw beef meat and grilling parameters on sensory quality and content of heterocyclic aromatic amines

Arkadiusz Szterk [a, *], Katarzyna Jesionkowska [b]

[a] Department of Food Analysis, Prof. Waclaw Dabrowski Institute of Agricultural and Food Biotechnology, Rakowiecka 36, 02-532 Warsaw, Poland
[b] Department of Functional Food, Organic Food and Commodities, Faculty of Human Nutrition and Consumer Sciences, Warsaw University of Life Sciences, 159 c Nowoursynowska, 02-776 Warsaw, Poland

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ABSTRACT

Heterocyclic aromatic amines (HAAs) are one of the most cancerogenic and mutagenic compounds present in food products. Thus, the researches on discovering new factors influencing formation of HAAs in food as well as the ways of reduction these compounds in food products are one of the main directions in food science. These investigations should led to the production of tasty food with significantly decrease content of HAAs. Unfortunately, there is no scientific research which are investigating the concentration of HAA together with the sensory properties of the products (e.g. grilled beef meat). The performance of the typical grilling process (180 °C and 280 °C) of beef meat (rib eye, sirloin and roast beef) stored in a raw state during 5 and 15 days in cooling condition and, than sensory evaluation and determination of correlation between overall sensory quality and content of HAA was the objective of presented study. This experiment confirmed that through proper selection parameters of culinary processing parameters such as thermal meat processing, the type of the culinary element of the beef meat and time of its storage in cooling conditions it is possible to ensure high overall sensory quality along with low content of HAA forming during grilling.

1. Introduction

The majority of heterocyclic aromatic amines (HAA) has strong mutagenic and cancerogenic properties as they are included into the group of indirect mutagens/cancerogens – subjecting to the metabolic activation due to influence of cytochrome P450. HAA are compounds mainly responsible for the mutagenicity and carcinogenicity of food products both of plant and animal origin, preserved in high temperatures during roasting, grilling or frying. Numerous experiments revealed that HAAs can cause target site mutation, deletion, insertion and breaking the hydrogen binding of DNA chain. The tumorigenic transformation and the mutations blocking the genome replication are the effect of mutations resulted from HHA activity (Sugimura, Wakabayashi, Nakagama, & Nagao, 2004; Turesky, 2007). Many animal studies revealed their strong carcinogenetic properties (Shirai et al., 1997; Sichler et al., 2002). The broaden and detailed examination of biological activity of HAAs indicated that MeIQ and PhIP shows strong mutagenic properties and Trp-P-1, Trp-P-2, Glu-P-1, Glu-P-2, AαC, MeAαC, IQ, MeIQ, MeIQx and PhIP are characterized by strong cancerogenic properties against mammal cells (Sugimura et al., 2004; Turesky, 2007).

It is well known that HAAs are formed in food during thermal processing in the temperature over 100 °C. Generally, their concentration and profile in food are mainly function of temperature and time of the thermal processing. One of factors recently discovered is time of cold storage of raw meat before processing (Polak, Andrensek, Zlender, & Gasperlin, 2009; Polak, Dosler, Zlender, & Gasperlin, 2009; Szterk et al., 2012; Szterk & Waszkiewicz-Robak, 2014). In experiments conducted by Szterk and Waszkiewicz-Robak (2014) the increase of concentration of nitric bases and their nucleotides during cold storage of raw meat lasting 15 days was observed. These compounds are substrates for HAAs creation but also are very important precursors of flavor. During cold storage, the textural properties are also shaped, thus, time of raw meet refrigerated storage has a significant influence on sensory quality after thermal processing of meat. However, this

* Corresponding author.
E-mail address: szterkarkadiusz@gmail.com (A. Szterk).

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factor never was correlated with content of HAAs and sensory quality of meat treated with high temperature (Gibis & Weiss, 2010; Olsson, Skog, Lundstrom, & Jagerstad, 2005).

In the majority of researches published during last twenty years, the very drastic parameters of thermal processing were applied (high temperature, long time and thin piece of meat). It is unknown whether such products are edible, because the experiments concerning of HAAs formation during thermal processing usually luck the evaluation of sensory quality of processed meat.

It was hypothesized that too drastic thermal processing conditions promotes the formation of the HAAs simultaneously with deterioration of the sensory quality of processed meat (Alaejos & Afonso, 2011). Thus, optimization of thermal processing parameters together with other factors essential for HAAs creation during grilling process (the time of the meat storage in cooling conditions) leading to minimization of HAAs content together with the maintaining of high sensory quality of the grilled beef meat, is possible.

The main objective of this study was performance of grilling process with maintenance of parameters typically reached at home but in controlled condition in laboratory what enabled the detailed characterization of process parameters and their precise multiple repetition. The sensory quality of the grilled meat in the accredited sensory laboratory followed by the determination of HAAs concentration was conducted. The correlation between the content of the HAAs and the sensory quality of the grilled meat was performed. The influence of meat storage in the cool conditions on the sensory quality and HAAs content was also determined.

2. Materials and methods

2.1. Samples

Sirloin – TDR, roast beef – STR and rib eye (steak) – CUB acquired from 12 different 20–23 month old bulls (*Limousine breed*) were used in this study. Meat samples taken in a slaughterhouse three days after the slaughter were divided into pieces (every piece of meat has a weight of about 2 kg) and kept in vacuum packaging at 2 °C (tolerance ± 0.2 °C). Analytical samples were taken after 5 and 15 days of storage. The samples were submitted to the following thermal treatment: three meat slices of at least 2.5 cm thickness taken from each animal were grilled at 180 °C and 280 °C.

2.2. Culinary processing

2.2.1. Grilling

Three 2.5 cm-thick steaks were cut off from each sirloin – TDR, roast beef – STR and rib eye (steak) – CUB sample and were grilled in an electrical grill at 180 °C and 280 °C after 5 days and 15 days of slaughter. Grilling were made accordance with Szterk and Waszkiewicz-Robak (2014).

2.3. Heterocyclic aromatic amines determination

HAAs were determined in the grilled meat samples in accordance with Szterk et al. (2012) and Szterk (2013).

2.3.1. Procedure

25 g of meat was placed in a glass bottle with a volume of 100 ml covered with a cap with Teflon seal, to it 40 ml of 1 M NaOH and 25 μL of internal standard (150 pg – 13C2 and 15N Trp-P 1) was added. The sample was inserted in a shaking incubator at 80 °C for approx. 2 h. After the alkaline hydrolysis the sample was cooled and mixed with 50–60 g of diatomaceous earth (Diatomaceous earth Sigma Aldrich 392,545). Powdery sample was transferred to a catheter syringe with volume of 160 ml (at the bottom of it was placed a small amount of glass wool to prevent loss of the sample), and then the whole was slightly compacted in order to level the material. Syringe was placed in a suction flask with volume of 250 ml, which was sealed and then 400 ml of ethyl acetate was extracted using a slight vacuum (700 mbar). The ethyl acetate solution was filtered through fluted filter paper containing about 15 g of anhydrous sodium sulphate in order to achieve the possible dehydration of the sample, extracting a small amount of water, which might enter into it during the extraction of sample mixed with diatomaceous earth with ethyl acetate. The whole was filtered directly into the 500 ml flask with round bottom, and then the solvent was evaporated to a volume of approx. 5 ml using a vacuum evaporator. The prepared sample was than filtered through a fluted filter paper directly into the activated OASIS MCX 150 mg, ion – exchange stanchion 6 ml manufactured by Waters, additionally fitted with container increasing its capacity to 50 ml. The sample in the flask was washed with ethyl acetate several times, using about 40 ml of solvent and then it was put on the stanchion. For purification of the sample by aforesaid stanchion, the method described by Szterk et al. (2012) was used. 100 ml of the prepared sample was put on the chromatographic column.

2.3.2. Chromatographic separation

Chromatographic separation was according with Szterk et al. (2012). Heterocyclic aromatic amines were separated using high performance liquid chromatography in the reversed-phase configuration (Thermo Scientific UPLC/ACCEL) and determined using the mass spectrometer LCQ Fleet using chemical ionisation at atmospheric pressure – APCI (Thermo Scientific).

2.4. Sensory analysis

The detailed sensory assessment of the samples of grilled beef meat was performed with implementation of the Quantitative Descriptive Analysis (QDA) according to procedure described by Stone and Sidel (1985). This way of sensory profiling was chosen as method widely used for evaluation of different food products providing information about whole product profile which can be easily analysed statistically and graphically presented (Cadena et al.; 2013; Lawless & Heymann, 1999; Meilgaard, Civiele, & Carr, 2006; Morais, Cruz, Faria, & Bolini, 2014). The analytical procedure of sensory evaluation was conducted in accordance to ISO standards 1399 (ISO, 2010a), in the accredited Laboratory of Sensory Analysis at Warsaw University of Life Sciences which is a member of European Sensory Network.

2.4.1. Samples preparation and presentation

The individual samples of each type of grilled beef (weighting around 15 g) were placed in transparent, odorless, plastic boxes (125 ml) covered with thigh lids in order to display the aroma uniformly in the sample's headspace. The samples of grilled beef meat were not salted or flavoured with spices. The samples were evaluated at room temperature (22 °C). The unsweetened tea (60 °C) was used between samples as the neutralizer.

The samples sets for each evaluator were coded individually with three-digit numbers to avoid the carry – over effect (the influence of the previous sample on the evaluation of the further one) the samples were presented randomly and the order of samples in the first session was different then in the second one.

2.4.2. Assessors selection and training

The assessors engaged in sensory evaluation of grilled beef meat samples were chosen from sensory experts trained in accordance with ISO standards 8586-2 (ISO, 2008) and having broaden experience in different food products. Firstly 15 evaluators participated in 5 preliminary sessions performed according to ISO standards 1399