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Reducing effect of artichoke extract on heterocyclic aromatic amine formation in beef and chicken breast meat

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ARTICLE INFO

Chemical compounds studied in this article:
IQ (PubChem CID: 53462)
IQx (PubChem CID: 105041)
MeIQ (PubChem CID: 62274)
MeIQx (PubChem CID: 62275)
4,8-DiMeIQx (PubChem CID: 104739)
PhIP (PubChem CID: 1530)
Harman (PubChem CID: 486-84-0)
Norharman (PubChem CID: 244-63-3)
AαC (PubChem CID: 62805)
Trp-P-2 (PubChem CID: 5284476)

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ABSTRACT

The aim of this study was to investigate the inhibitory effect of different levels of artichoke extract (0, 0.5, and 1.0%) on the formation of heterocyclic aromatic amines (HAAs) in beef and chicken breast meat cooked by either pan-frying or oven-roasting. All meat samples were cooked at three different temperatures (150, 200, and 250 °C) and the levels of 12 HAAs (IQ, IQx, MeIQ, MeIQx, 4,8-DiMeIQx, 7,8-DiMeIQx, PhIP, harman, norharman, AαC, MeAαC, and Trp-P-2) were assessed. The total HAA content in beef and chicken breast ranged from not detectable to 49.26 ng/g, and not detectable to 83.06 ng/g, respectively. The inhibitory effects of 0.5 and 1.0% artichoke extracts on total HAAs levels were found to be 6–46% and 25–98% in beef, and 5–97% and 14–95% in chicken breast, respectively. The present study showed that artichoke extracts could mitigate HAA formation especially in oven-roasted beef and chicken breast meat.

1. Introduction

Meat is an important part of our diet and contributes valuable nutrients, such as high quality proteins, all essential amino acids, niacin, vitamin B6, vitamin B12, iron, zinc, selenium, phosphorus, endogenous antioxidants, and other bioactive substances, including taurine, carnitine, carnosine, ubiquinone, glutathione, and creatine, which are beneficial to our health (Oz, Kaban, & Kaya, 2010; Williams, 2007). However, several epidemiological studies have shown that high intake of meat and cooking practices are associated with an increased risk of various human cancers, including colon, breast, prostate, and pancreatic cancers (Hasnol, Jinap, & Sanny, 2014; Knize & Felton, 2005; Persson, Graziani, Ferracane, Fogliano, & Skog, 2003; Viegas, Amaro, Ferreira, & Pinho, 2012). Heterocyclic aromatic amines (HAAs) are formed in muscle foods frequently cooked at high temperatures via the Maillard browning reaction with creati(ni)ne, amino acids, and sugars as the precursors, and are considered as highly mutagenic and potentially carcinogenic compounds (Murkovic, 2004). Many factors influence this complex reaction, and due to concerns about risk factors for human cancers, human exposure to these compounds should be avoided (Gibis, 2007; Johansson & Jägerstad, 1996; Murkovic, 2004). The quantity of HAAs depends on several factors, such as meat type, the levels of doneness, cooking methods, cooking temperature and time, pH, water activity, concentration of precursors heat, mass transfer, lipid level, or use of antioxidants (Dundar, Sarıcoban, & Yılmaz, 2012; Kizil, Oz, & Besler, 2011).

Artichoke (*Cynara scolymus L.*) is a herbaceous plant of the Asteraceae family, and represents an important component of the Mediterranean diet (Lattanzio, Kroon, Linsalata, & Cardinali, 2009). Additionally, artichoke is perceived as a health-protective plant due to

Abbreviations: HAA, heterocyclic aromatic amine; nd, non-detected; IQ, 2-amino-3-methylimidazo[4,5-*f*]quinoline; IQx, 2-amino-3-methylimidazo[4,5-*f*]quinoxaline; MeIQ, 2-amino-3,4-dimethylimidazo[4,5-*f*]quinoline; MeIQx, 2-amino-3,8-dimethylimidazo[4,5-*f*]quinoxaline; 4,8-DiMeIQx, 2-amino-3,4,8-trimethylimidazo[4,5-*f*]quinoxaline; 7,8-DiMeIQx, 2-amino-3,7,8-trimethylimidazo[4,5-*f*]quinoxaline; PhIP, 2-amino-1-methyl-6-phenylimidazo[4,5-*b*]pyridine; AαC, 2-amino-9H-pyrido[2,3-*b*]indole; MeAαC, 2-amino-3-methyl-9H-pyrido[2,3-*b*]indole; Norharman, 9H-pyrido[3,4-*b*]indole; Harman, 1-methyl-9H-pyrido[4,3-*b*]indole; Trp-P-2, 3-amino-1-methyl-5H-pyrido[4,3-*b*]indole; 4,7,8-TriMeIQx, 2-amino-3,4,7,8-tetramethylimidazo[4,5-*f*]quinoxaline

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its hepatoprotective, anticarcinogenic, and hypocholesterolemic properties, and hence is widely used as an ingredient in soups, stews, and salads (Ceccarelli et al., 2010; Gaafar & Salama, 2013; Garbetta et al., 2014; Pereira, Calhelha, Barros, & Ferreira, 2013). Artichoke extracts are commercially produced from plant leaves and contain high levels of minerals, low levels of lipids, vitamin C, and dietary fiber, and are a rich source of inulin (Gaafar & Salama, 2013; Lattanzio et al., 2009; Pereira et al., 2013). The major antioxidant compounds in artichoke extracts are cafinulin and feoylquinic acid derivatives, such as cynarin and chlorogenic acid and other flavonoids, including luteolin and apigenin (Ceccarelli et al., 2010; Garbetta et al., 2014; Pereira et al., 2013; Wang et al., 2003).

Numerous strategies have been used to limit the formation of HAAs, including decreased cooking time and temperatures and pre-cooking of meat in the microwave. However, due to the free radical scavenging activity of certain compounds with antioxidant potential, their addition to meat strongly suppresses the formation of HAAs (Knize, Dolbeare, Carroll, Moore, & Felton, 1994; Vitaglione & Fogliano, 2004). Antioxidant compounds have been incorporated by spreading onto meat surfaces (Balogh, Gray, Gomaa, & Booren, 2000) or by addition to ground meat (Shin, Rodgers, Strasburg, & Gray, 2002). Also, prior to cooking, the effect of marinades containing dietary antioxidant compounds on HAA levels has been previously studied, showing that meat marinades are effective in reducing HAA levels (Busquets, Puignou, Galceran, & Skog, 2006; Gibis & Weiss, 2012; Smith, Ameri, & Gadgil, 2008). Finding promising strategies to decrease HAA formation in foods is important to minimize HAA exposure and the effect of artichoke extract on the reduction of HAA formation has not been investigated previously. In this regard, the objective of this study was to evaluate the potential inhibitory effects of artichoke extract on the formation of HAAs in pan-fried and oven-roasted beef and chicken breast meat at different cooking temperatures.

2. Material and methods

2.1. Materials

2.1.1. Raw materials

The chicken breast meat and beef *longissimus lumborum* were purchased from a local market in Ankara, Turkey. They were transported to the laboratory on ice. The commercial artichoke extract (*Cynara scolymus L.*) was obtained from Balen, Ankara, Turkey.

2.1.2. Chemicals

Propyl gallate, ethylenediaminetetraacetic acid disodium, tetraethoxypropane, trichloroacetic acid, thiobarbituric acid (TBA), diacetyl, diethyl ether, picric acid, 1-naphthol, creatine standard, creatinine standard, potassium hexacyanoferrate, zinc sulfate, D-(+)-glucose (99.5%), and D-(+)-fructose (99%) were obtained from Merck KGaA (Darmstadt, Germany). Chemicals for HAA analysis, including ethyl acetate, methanol, acetone, sodium hydroxide, hydrochloric acid, glacial acetic acid, acetonitrile, and ammonium hydroxide solution (25%) were purchased from Merck KGaA (Darmstadt, Germany). For solid phase extraction, Extrelut NT packing material (Merck, Darmstadt, Germany), Bond Elut reservoir (Varian, Harbor City, California, USA), Oasis MCX cartridge (Waters, Milford, Massachusetts, USA), SPE manifold (Supelco Visiprep, St. Louis, Missouri, USA), and Oasis HLB cartridge (Waters, Milford, Massachusetts, USA) were used. HAA standards were purchased from Toronto Research Chemicals (Downsview, Ontario, Canada): IQ (CAS no:76180-96-6, 2-amino-3-methylimidazo [4,5-*f*]quinoline), IQx (CAS no:108354-47-8; 2-amino-3-methylimidazo [4,5-*f*]quinoxaline), MeIQ (CAS no:77094-11-2; 2-amino-3,4-dimethylimidazo[4,5-*f*]quinoline), MeIQx (CAS no:77500-04-0; 2-amino-3,8-dimethylimidazo[4,5-*f*]quinoxaline), 4,8-DiMeIQx (CAS no:95896-78-9; 2-amino-3,4,8-trimethylimidazo[4,5-*f*]quinoxaline), 7,8-DiMeIQx (CAS no:92180-79-5; 2-amino-3,7,8-trimethylimidazo[4,5-*f*] quinoxaline), PhIP (CAS no:105650-23-5; 2-amino-1-methyl-6-phenylimidazo[4,5-*f*]pyridine), harman (CAS no:486-84-0; 1-methyl-9H-pyrido[3,4-*b*] indole), norharman (CAS no:244-63-3; 9H-pyrido[3,4-*b*] indole), AαC (CAS no:26148-68-5; 2-amino-9H-pyrido[2,3-*b*]indole), MeAαC (CAS no: 68006-83-7; 2-amino-3-methyl-9H-pyrido[2,3-*b*]indole), Trp-P-2 (CAS no:72254-58-1; 3-amino-1-methyl-5H-pyrido[4,3-*b*] indole), and 4,7,8-TriMeIQx (CAS no:132898-07-8; 2-amino-3,4,7,8-tetramethylimidazo[4,5-*f*]quinoxaline). Chemicals and solvents were of high-performance liquid chromatography (HPLC) or analytical grade. All solutions, except the HPLC-grade solutions, were passed through a 0.45 μm filter (Millipore, Billerica, Massachusetts, USA) before use.

2.2. Methods

2.2.1. Sample preparation

All meat samples were sliced to a thickness of 1 cm and divided into 36 groups according to meat type (beef, chicken breast meat), cooking methods (pan-frying, oven-roasting), cooking temperatures (150, 200, and 250 °C) and artichoke extract concentrations (0, 0.5, and 1.0%). Twelve of these groups were chosen as control groups. All experiment were performed in two replicates and two slices (approximately, 100 g/slice) of meat sample were used for per treatment. The water-soluble artichoke extracts were dissolved in distilled water (1:10, w/v) and then added at concentrations of 0 (control), 0.5, and 1.0% (w/w). After adding the extract, the samples were stored at 4 °C overnight.

2.2.2. Cooking process

Both pan-frying and oven-roasting cooking methods were used. The pan-frying process was carried out with a Teflon-coated pan. The cooking time was determined based on preliminary experiments. All samples were prepared without salt, spices, fat, or oil. For pan-frying, samples were cooked for 5 min per side. For oven-roasting, samples were placed in an oven and cooked for 20 min. The surface temperature was measured using a thermometer (Testo 905-T2, Lenzkirch, Germany) to adjust the cooking temperature. Experiments were performed in two replicates. After cooking, the samples were cooled to room temperature, weighed, and homogenized using a kitchen blender (Tefal, Sarcelles, France) to produce a uniform sample. They were then stored at − 20 °C until further analysis.

2.2.3. Proximate composition and cooking loss

The proximate composition of meat samples, including protein, lipid, ash, and water content was determined according to AOAC methods (Horwitz, 2000). The lipid content was determined via the Soxhlet method and the protein content was analyzed via the Kjeldahl method. The pH of samples was measured using a digital pH meter (Hanna, Vohringen, Germany) calibrated with standard buffers of pH 4.0 and 7.0 at room temperature. The loss in sample weight during cooking was calculated from the difference in raw and cooked weight. All parameters were determined in duplicate.

2.2.4. Creatine/creatinine analyses

The concentrations of creatine and creatinine in the meat samples were determined using a method described by Polak, Došler, Žlender, and Gašperlin (2009). The results were expressed as mg/g wet weight of samples. The analyses were determined in duplicate.

2.2.5. Analysis of reducing sugars

Raw and cooked meat samples were lyophilized and the levels of glucose and fructose were analyzed using a method described by Serpen and Gokmen (2009), with minor modifications. The analyses were determined in duplicate.

2.2.6. Analysis of TBA reactive substances (TBARS)

Lipid oxidation was measured by analyzing TBARS. TBARS were determined using a modified version of the procedure reported by Kerth