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Effect of protein-xanthophylls (PX) concentrate of alfalfa supplementation on physico-chemical properties of turkey breast and thigh muscles during ageing

M. Karwowska [a,*], J. Stadnik [a], Z.J. Dolatowski [a], E.R. Grela [b]

[a] Department of Meat Technology and Food Quality, University of Life Sciences in Lublin, Skromna 8, 20-704 Lublin, Poland
[b] Institute of Animal Nutrition, University of Life Sciences in Lublin, Akademicka 13, 20-950 Lublin, Poland

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ABSTRACT

The objective was to evaluate the effects of a dietary protein-xanthophylls (PX) concentrate of alfalfa to turkey diets (at 15 and 30 g kg^{-1} feed) on the physico-chemical properties of breast and thigh muscles during ageing. The experiment involved 120 turkeys (Big-6 type) allotted to 3 groups (group I—control group; group II—with 1.5% supplementation of the protein-xanthophylls (PX) concentrate; group III—with 3% supplementation of the protein-xanthophylls (PX) concentrate). Measurements of pH, water holding capacity, color, oxymyoglobin content, TBARS and oxidation–reduction potential showed that the addition of protein-xanthophylls (PX) concentrate of alfalfa to a turkey diet did not cause deterioration of breast and thigh meat quality. In addition, changes in color, oxymyoglobin content, TBARS and oxidation–reduction potential values suggested that the inclusion of the concentrate to turkey diets acts as an antioxidant in the raw meat.

1. Introduction

The quality of feed components has a great affect on animal health and meat quality (Pettigrew & Esnaola, 2001). It is well established that in the case of monogastric animals such as turkey many nutrients as well as feed additives are transferred from the feed to the muscle and fat tissues. Therefore, a number of studies have dealt with the effect of dietary vitamin E (Mercier, Gatellier, Vincent & Renerre, 2001) or α-tocopheryl acetate (Botsoglou, Grigoropoulou, Botsoglou, Govaris & Papageorigiou, 2003; Higgins, Kerry, Buckley & Morrissey, 1998; Mercier et al., 2001) supplementation of turkey diets on the storage stability of meat and meat products. In addition, dietary fat has been reported to be effective in improving the oxidative stability of chicken meat (Botsoglou et al., 2003; Mercier et al., 2001).

Dietary supplementation with herbs and herb extracts proved to be a simple method to introduce a natural antioxidant into chicken meat. Several reports suggested that alfalfa and its products can be used as components of animal feed (Grela, Semeniuk & Florek, 2008; Güçlü et al., 2004; Heitman & Meyer, 1959; Hopkins & Nicholson, 1999; Karwowska, Dolatowski & Grela, 2007; Karwowska, Dolatowski and Grela, 2008). It has been shown that alfalfa is a source of natural antioxidants (xanthophyll and its derivatives), vitamins (C, D, E, K, and P) and minerals (especially iron and copper) (Ben Aziz, Grossman, Budowski, Ascarelli & Bondi, 2006; Grela et al., 2008; Heitman & Meyer, 1959; Stochmal, Simonet, Macias & Oleszek, 2001). Furthermore, several studies have reported the hypocholesterolemic and antiatherosclerotic activities of alfalfa meals. Saponins are suggested to be responsible for the reduction of cholesterol and prevention of atherosclerotic plaque formation and exhibit antioxidants activity (Avato et al., 2006; Dixit & Joshi, 1985; Francis, Kerem, Makkar & Becker, 2002; Khaleel, Gad, El-Maraghy, Hifnawy & Abdel-Sattar, 2005). Alfalfa contains phytoestrogens, including flavonoids, isoflavonoids, coumestans and lignans with high levels of estrogenic activity (Lamsal, Koegel & Gunasekaran, 2007).

The literature contains limited information regarding supplementation of animal as well as poultry diets with alfalfa products (Güçlü et al., 2004; Krauze & Grela, 2008). The analysis of turkey blood by Krauze and Grela (2008) showed that a protein-xanthophylls (PX) concentrate of alfalfa significantly reduced concentrations of total protein and cholesterol. The results of Güçlü et al. (2004) indicated that addition of 9% alfalfa meal into a laying quail diet improved eggshell quality and reduced serum triglycerides and serum and egg yolk cholesterol without any adverse effect on performance. Generally, little attention has been paid to the effects of alfalfa addition on the physico-chemical and oxidative stability of meat.

The objective of this study was to evaluate the effects of a dietary protein-xanthophylls (PX) concentrate of alfalfa on the water holding capacity, oxidation–reduction potential, color stability and lipid oxidation of turkey breast and thigh muscles during ageing.

2. Materials and methods

2.1. Animals and diets

One hundred and twenty 42-day turkey poults (Big-6 type) were selected at random and divided into three groups of 40. A control

* Corresponding author. Tel.: +48 81 4623344; fax: +48 81 4623345.
E-mail address: malgorzata.karwowska@up.lublin.pl (M. Karwowska).

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group (group I) was fed according to the Poultry Feeding Standards (NRC, 1994). The other groups were fed this diet supplemented with 1.5% (group II) and 3.0% (group III) of a protein-xanthophylls (PX) concentrate of alfalfa, instead of the extracted soybean extracted meal. Alfalfa concentrate was prepared by compacting the juice of alfalfa leaves (Grela et al., 2008). Food and water were provided *ad libitum*. After 11 weeks, the turkeys with an average live weight of 16.9 ± 0.5 kg were slaughtered under commercial conditions. The breast and thigh muscles were taken, packed into the HDPE bags and stored at 4 °C until use. The samples were examined at 48; 96 and 120 h *post mortem*.

2.2. Proximate analysis

Lipid contents were measured by extraction, in a Soxhlet apparatus with methanol and chloroform according to Blight and Dyer (1959). Protein content was determined by the Kjeldahl method (AOAC 2000) and moisture content by drying samples (5 g) at 102 °C (AOAC 2000).

2.3. Measurement of pH

10 g of minced meat was homogenised with 100 ml of distilled water for 1 min using a homogenizator (IKA ULTRA-TURRAX T25 Basic, Germany). The homogenate was filtered through filter paper and the pH of the filtrate measured with a digital pH-meter CPC-501 (Elmetron) equipped with a pH electrode (ERH-111, Elmetron). Measurements were carried out on four replicates.

2.4. Determination of water holding capacity (WHC)

Measurement of WHC was performed using a centrifugation method (Wierbicki, Tiede & Burrell, 1962). 50 g of minced meat samples was homogenised with 50 ml of distilled water for 1 min using a homogenizator (IKA ULTRA-TURRAX T25 Basic, Germany). The homogenates were then centrifuged at 1500 g for 20 min using a MPW-350R centrifuge (MPW Med-Instruments, Poland). Water holding capacity was calculated as: $WHC = (M_1 - M_2)/M_3 \times 100\%$, where: M_1—weight of added water (g); M_2—weight of supernatant after centrifugation; M_3—weight of meat in homogenate (g). Four replications were made on each sample.

2.5. Oxidation–reduction potential (ORP)

Oxidation–reduction potential of muscles was determined according to the method of Nam and Ahn (2003) with slight modification. Meat samples (10 g) were homogenised with 30 ml of de-ionized water for 1 min using a homogenizator (IKA ULTRA-TURRAX T25 Basic, Germany). ORP measurements of the homogenates were carried out in four replications for each sample using a digital pH-meter CPC-501 (Elmetron) set to the millivolt scale and equipped with redox electrode (ERPt-13, Elmetron).

2.6. Color measurements

Color parameters (CIE L*a*b*) were measured on the freshly cut surface of a muscle using 8200 Series reflection spectrocolorimeter (X-Rite) with a D65 illuminant and a 10° standard observer (AMSA, 2005). In addition, reflectance measurements were taken from 580 to 630 nm and the reflectance ratio 630 nm/580 nm used to estimate the oxymyoglobin proportion of the myoglobin pigment (Hunt et al., 1991). Prior to use, the spectrocolorimeter was calibrated against white and black standard tiles. Six measurements were taken on each sample and averaged for statistical analysis.

2.7. Lipid oxidation

Lipid oxidation was determined as thiobarbituric acid reactive substances (TBARS) values following the procedure of Pikul, Leszczyński and Kummerow (1989). 10 g of ground meat sample was homogenized with 34.25 mL of cold (4 °C) 4% perchloric acid and 0.75 mL of BHT in ethanol (0.01%). The sample was filtered through Whatman No. 1 filter paper and the filtrate adjusted to 50 mL with 4% perchloric acid. Then, 5-mL of the filtrate was transferred to a test tube and mixed with 5 mL of 0.02 M TBA solution in distilled water. The tube was heated in a boiling water bath for 1 h and then cooled for 10 min with cold tap water. The rose-pink color obtained by the reaction between malondialdehyde and 2-thiobarbituric acid was measured at 532 nm (Nicole Evolution 300, Thermo Electron Corporation). Results are reported as milligram of malondialdehyde (MDA) per kilogram of the sample. Measurements were carried out in four replications for each sample.

2.8. Statistical analysis

The data were analyzed using the Statgraphics v. 5. Analysis of variance was carried out. Significance of differences between the feeding groups at the same ageing time and the same group at different ageing times was tested (at the significance level $p \leq 0.05$) using *T*-Tukey's test.

3. Results and discussion

3.1. Chemical composition and pH of meat

The chemical composition of turkey breast and thigh muscle was not influenced by the diet (Table 1).

Incorporation of protein-xanthophylls (PX) concentrate of alfalfa in turkey diets had no significant ($p \leq 0.05$) effect on their pH during 120 h of storage (Fig. 1). The pH values of breast muscles were lower than those of the thigh muscles. pH values did not vary during storage.

3.2. Water holding capacity of the meat

WHC of fresh meat is important since it affects major characteristics of the cooked meat such as technological quality and sensory properties. The protein-xanthophylls (PX) concentrate of alfalfa supplements in the turkey diet had no significant effect ($p \leq 0.05$) on the water holding capacity of the breast and thigh muscles during storage (Fig. 2).

3.3. ORP of the samples

Dietary supplementation with protein-xanthophylls (PX) concentrate of alfalfa had no significant ($p \leq 0.05$) effect on the oxidation reduction potential of turkey breast muscle (Fig. 3A) at 48 and 96 h *post mortem*. Results for redox measurements of the breast muscles from the group with 1.5% supplementation of PX 120 h after slaughter

Table 1
Proximate analysis of turkey breast and thigh muscles (means ± standard error).

Samples		Chemical compositions		
		Moisture (%)	Protein (%)	Fat (%)
Group I (control)	Breast muscle	73.0 ± 0.8	24.26 ± 0.4	1.52 ± 0.3
	Thigh muscle	76.1 ± 1.2	19.49 ± 0.1	3.27 ± 0.8
Group II 1.5% PX)	Breast muscle	72.2 ± 1.3	24.19 ± 0.6	2.21 ± 0.2
	Thigh muscle	75.5 ± 0.1	19.52 ± 0.6	3.65 ± 0.9
Group III (3% PX)	Breast muscle	73.8 ± 1.0	23.49 ± 0.3	1.90 ± 0.6
	Thigh muscle	76.1 ± 0.5	19.81 ± 0.1	3.19 ± 0.8