

LIPID OXIDATION IN CHICKEN MEAT AFTER APPLICATION OF BEE POLLEN EXTRACT, PROPOLIS EXTRACT AND PROBIOTIC IN THEIR DIETS

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ABSTRACT

In the experiment, the effect of the addition of bee pollen, propolis extract and probiotic in a feed mixture for chicken broilers Ross 308 on oxidative stability of breast and thigh muscles during chilled storage was investigated. In the experiment were included 180 pieces of one day-old chicks, which were divided into 4 groups (control, E1, E2 and E3). Chickens were fed by *ad libitum* system until the age of 42 days. These feed mixtures were made without antibiotics preparation and coccidiostats. Bee pollen extract in amount of 400 mg.kg⁻¹ (E1), propolis extract in an amount of 400 mg.kg⁻¹ (E2) was added into feed mixtures and probiotic (*Lactobacillus fermentum*) (E3) in an amount 3.3 g added daily to the water given the experimental group. During whole period of chilled storage were higher values of MDA determined in control group compare to experimental groups. The higher average MDA value determined in breast muscles of broiler chicken hybrid combination Ross 308 was in samples of control group (0.129 mg.kg⁻¹) compared to experimental groups E1, E3 (0.125 mg.kg⁻¹) and E2 (0.115 mg.kg⁻¹) after 7-day of chilled storage. Significantly higher values of MDA were determined in control group compare to second experimental group on the end of storage. Trend of thigh muscle oxidation stability of chicken hybrid combination Ross 308 was during 7 days of chilled storage similar than in breast muscle. The higher average MDA value determined in thigh muscles was in samples of control group (0.142 mg.kg⁻¹) compared to experimental groups E1 (0.137 mg.kg⁻¹), E2 (0.125 mg.kg⁻¹) and E3 (0.138 mg.kg⁻¹) after 7-day of chilled storage. We have not determined statistically significant differences between testing groups on the end of storage. Higher amount of MDA in thigh muscle compare to breast muscle is due to by higher amount of fat occurred in thigh muscle.

Keywords: oxidative stability; meat; broiler chicken; propolis; bee pollen; probiotic

INTRODUCTION

Consumer concerns on the quality of meat and meat products have greatly increased during past decades. „Quality“ and „healthfulness“ were reported to be one of the most important factors for influencing consumers choice for food (Lennernas et al., 1997).

Lipid oxidation is one of the primary causes of quality damages related to flavour, colour, taste and nutritional composition of meat and meat products (Kanner, 1994; Gray et al., 1996; Marcinčák et al., 2005; Min et al., 2008).

Many factors can contribute to the initiation and development of lipid oxidation process in meat, such as fat content and fatty acid profile, degree of processing, storage conditions, and the balance between tissue pro and antioxidants content (Jensen, et al., 1997). Poultry meat is notably sensitive to lipid oxidation because of its high content of polyunsaturated fatty acids (Botsgolou, et al., 2002) and thigh meat, as compared to breast meat, is particularly vulnerable because of its higher fat content (Jensen, et al., 1998).

Lipid oxidation products have harmful biological effects and some have been related to the etiology of various neurodegenerative and cardiovascular diseases as well as different types of cancer (Cohn, 2002; Schroepfer, 2002).

Thus, it is important to not only improve the nutritional value of foods but also to minimize lipid oxidation to provide healthy food products. Consumers reject some antioxidants that are very effective in controlling lipid oxidation whereas they accept natural products with antioxidant activity since they are often perceived as safer and more nutritious than food containing additives or food coming from animal feed ingredients of a non-natural origin. While natural products are desired by many consumers, these products can be difficult to define since some man-made food additives and feed ingredients can be completely identical to those present in nature, slightly different, or modified for a better use (Bou et al., 2009). The negative consequences of lipid oxidation of meat and meat products can be overcome by the use of antioxidants in the diets (Kazimierczak et al., 2008; Haščík et al., 2012; Elimam et al., 2013). In recent period, after ban of antibiotics and coccidiostats in poultry nutrition in EU,

different alternative supplements e.g. probiotics, plant essential oils and their extract, enzymatic preparations and bee pollen products (pollen, propolis or their extracts), have begun to use for their positive influence on health state, feed utilization, nutritional and sensory quality of product as well as economics of poultry industry production (Wang et al., 2004; Shalmany and Shivazed, 2006; Seven et al., 2008).

The aim of the experiment was to determine the oxidative stability in the most valuable parts of chicken carcasses (Ross 308 hybrid combination) during the cold store (7 days) after application of bee pollen extract and propolis extract added to feed mixtures and probiotic added into drinking water.

MATERIAL AND METHODOLOGY

The experiment was carried out in test poultry station of Slovak University of Agriculture in Nitra. A total of 180 one day-old Ross 308 broiler chicks were randomly divided into 4 groups, namely, control (C) and experimental (E1, E2, E3) of 45 pcs chickens. During the whole period of experiment, the broiler chickens had *ad libitum* access to feed and water.

The feeding lasted 42 days. During that period, experimental broiler chickens were fed with a starter complete feed mixture HYD-01 (until 21 days of age) and a grower feed mixture HYD-02 (from 22nd to 42nd day of age). The composition of feed mixtures is given in Table 1. The feed mixtures both starter and grower were produced without any antibiotic preparations and coccidiostatics.

All the groups were fed with the same feed mixtures. However, chickens in the control group were fed with basal diet containing no special supplement, while the diet of chickens in experimental groups contained the diet supplements as follows: bee pollen extract in amount of 400 mg.kg⁻¹ added to feed mixtures given to the group E1, propolis extract in amount of 400 mg.kg⁻¹ added to feed mixtures given to the group E2, probiotic in an amount 3.3 g added daily to the water given the group E3. The groups were kept under the same conditions.

In the experiment, the probiotic preparation "Propoul" based on *Lactobacillus fermentum* (1.10⁹ CFU per 1 g of bearing medium) was used.

Bee pollen and propolis had origin in the Slovak Republic. The extracts were prepared from minced bee pollen and propolis in the conditions of the 80% ethanol in

Table 1 Composition of the diets.

Ingredients (%)	Starter (1 to 21 days of age)	Grower (22 to 42 days of age)
Wheat	35.00	35.00
Maize	35.00	40.00
Soybean meal	21.30	18.70
Fish meal (71% N)	3.80	2.00
Dried blood	1.25	1.25
Ground limestone	1.00	1.05
Monocalcium phosphate	1.00	0.70
Fodder salt	0.10	0.1.50
Sodium bicarbonate	0.15	0.20
Lysin	0.05	0.07
Methionin	0.15	0.22
Palm kernel oil Bergafat	0.70	0.16
Premix Euromix BR 0.5% ¹	0.50	0.50
Analysed composition (g.kg⁻¹)		
Crude protein	210.76	190.42
Fibre	30.19	29.93
Ash	24.24	19.94
Ca	8.16	7.28
P	6.76	5.71
Mg	1.41	1.36
Linoleic acid	13.51	14.19
ME _N (MJ.kg ⁻¹)	12.02	12.03

¹active substances per kilogram of premix: vitamin A 2 500 000 IU; vitamin E 50 000 mg; vitamin D3 800 000 IU; niacin 12 000 mg; d-pantothenic acid 3 000 mg; riboflavin 1 800 mg; pyridoxine 1 200 mg; thiamine 600 mg; menadione 800 mg; ascorbic acid 50 000 mg; folic acid 400 mg; biotin 40 mg; vitamin B12 10.0 mg; choline 100 000 mg; betaine 50 000 mg; Mn 20 000 mg; Zn 16 000 mg; Fe 14 000 mg; Cu 2 400 mg; Co 80 mg; I 200 mg; Se 50 mg.

the 500 cm³ flasks, according to **Krell (1996)**. The extraction was accomplished in a water bath at 80 °C for one hour. After that, the extracts were cooled and centrifuged. The obtained supernatants were evaporated in a rotary vacuum evaporator at bath temperature 40 – 50 °C and weighed. Residues in an amount of 40 g were dissolved in 1000 cm³ of 80% ethanol and used for 100 kg of feed mixture.

At the end of feeding (day 42th) from each group were selected 10 pieces of chicken for slaughter analysis. To determine changes in lipid degradation (determination of thiobarbiturates numbers, TBA) the samples of chickens were boned and thigh and breast muscle packed into polyethylene bags and stored for 7 days at 4 °C.

TBA value expressed in number of malondialdehyde were measured in the process of first storage day of 1st, 3rd, 5th and 7th day. TBA number was determined by **Marcinčák et al. (2004)**. Absorbance of samples was measured on UV-VIS spectrophotometer T80 (PG Limited Instruments, UK) at a wavelength of 532 nm, the translation results on the amount of malondialdehyde (MDA) in 1 kg samples.

Results of the experiment was evaluated with statistical program Statgraphics Plus version 5.1 (AV Trading Umex, Dresden, Germany), were calculated variation-statistical values (mean, standard deviation) and to determine the significant difference between groups was used variance analyse with subsequent Scheffe test.

RESULTS AND DISCUSSION

The results of the oxidation stability measured in breast and thigh muscle of chickens Ross 308 during 7 days storage at 4 °C are shown in Table 2. Our results are in accordance with **Marcinčák et al. (2010)** who, after slaughtering and processing of poultry samples also show low values of MDA. During chilled storage of the breast and thigh muscles (7 days) were detected increased content of MDA in comparison to the first day of storage. During testing period of chilled storage were higher values of MDA measured in control group compare to experimental groups. The higher average value of MDA measured in

breast muscle of broiler chickens Ross 308 was in samples of control group (0.129 mg.kg⁻¹) compared to experimental groups E1, E3 (0.125 mg.kg⁻¹) and E2 (0.115 mg.kg⁻¹) after 7-day of chilled storage. Significantly higher values of MDA on the end of storage were determined in control group compare to second experimental group.

Trend of oxidation stability in thigh muscle of chicken hybrid combination Ross 308 was during 7 days of chilled storage similar than in breast muscle. The higher average value of MDA measured in thigh muscle was in samples of control group (0.142 mg.kg⁻¹) compared to experimental groups E1 (0.137 mg.kg⁻¹), E2 (0.125 mg.kg⁻¹) and E3 (0.138 mg.kg⁻¹) after 7-day of chilled storage. We have not found statistically significant differences between testing groups. Higher concentration of MDA in thigh muscle compare to breast muscle is due to by higher amount of fat passes into thigh muscle **Botsoglou et al. (2002)**.

Reached results of oxidation stability determined in chicken meat of hybrid combination Ross 308 after propolis extract addition in their diet are in accordance with **Betti et al. (2009)** and **Yasin et al. (2012)**. The possibilities of using alternative feed supplements containing various antioxidant active substances for poultry which increase the oxidation stability of the meat during its period of freeze storage are presented in works of **Mikulski et al. (2009)**, **Ahadi et al. (2010)**, **Marcinčák et al. (2010)**, **Karaalp and Genc (2013)**.

Ramos Avila et al. (2013) stated that the degradation pathways of fatty substances play one of the main causes of foods deterioration and unpleasant odours. This factor is also responsible for the loss of sensory properties such as flavour, texture, appearance, nutritional value of food, increases the drop losses, pigment, polyunsaturated fatty acids, fat-soluble vitamins, reduces the quality of meat intended for human consumption and ultimately reduces its stability, shelf life and safety.

Botsoglou et al. (2007) reported that a higher concentration of antioxidants in poultry meat has the effect of reducing lipid oxidation, ie there is a reduction in MDA values during chilling storage, which was confirmed by our results.

Table 2 Effect of storage (4 °C) on the concentration of malondialdehyde (mg.kg⁻¹) in breast and thigh muscle (mean ±SD).

Time of storage	Group			
	Control	E1	E2	E3
Breast muscle				
Day - 1	0.034 ±0.008 ^a	0.021 ±0.002 ^b	0.022 ±0.009 ^b	0.025 ±0.005 ^b
Day - 3	0.049 ±0.006 ^a	0.044 ±0.009 ^{ab}	0.042 ±0.011 ^{ab}	0.046 ±0.005 ^b
Day - 5	0.084 ±0.004	0.079 ±0.006	0.076 ±0.009	0.830 ±0.002
Day - 7	0.129 ±0.003 ^a	0.125 ±0.012 ^{ab}	0.115 ±0.006 ^b	0.125 ±0.010 ^{ab}
Thigh muscle				
Day - 1	0.043 ±0.002 ^a	0.030 ±0.008 ^b	0.028 ±0.002 ^b	0.033 ±0.011 ^{ab}
Day - 3	0.059 ±0.006 ^a	0.052 ±0.009 ^{ab}	0.047 ±0.006 ^b	0.050 ±0.004 ^b
Day - 5	0.097 ±0.012	0.092 ±0.014	0.087 ±0.007	0.095 ±0.011
Day - 7	0.142 ±0.019	0.137 ±0.020	0.125 ±0.008	0.138 ±0.022

Legend: Mean values in the same lines with different superscripts (a, b) are significantly different at *p* <0.05 level.

CONCLUSION

Results achieved in the experiment show that the addition of propolis extract in feed mixture for broiler chickens had positive impact on the reduction of oxidative processes in the breast and thigh muscles during chilling storage, but with the addition of bee pollen extract and probiotic has been recorded significant effect on the oxidation of fat in the breast and thigh muscles meat broiler chickens Ross308.

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