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OXIDATIVE STABILITY OF CHICKEN MEAT DURING STORAGE INFLUENCED BY THE FEEDING OF ALFALFA MEAL

Jana Tkáčová, Mária Angelovičová, Peter Haščík, Marek Bobko

ABSTRACT

The aim of our experiment was to determine the effect of the alfalfa meal component in feed mixtures of Ross broiler chickens on oxidative stability of meat. Proportion of alfalfa meal in feed mixtures was 4 and 6%. The results were compared to the control group without alfalfa meal in feed mixtures. At the end of the experiment (day 38), 6 pcs of broiler chickens from each group with an average live body weight over 1 800 g were randomly selected. The samples for chemical analysis consisted of identical proportion of breast and thigh muscle, and about 1 cm^2 of skin with subcutaneous fat. Fat from the meat was obtained after the samples drying. A fat was determined by extraction by means of laboratory instrument Det N Gras Selecta P. The oxidative stability of meat on the basis of acid number of fat was determided by chemical analysis. Chicken meat was stored at -18 °C for 12 months and 18 months. The acid number of fat of stored meat for 12 months was 7.38 mg KOH per g in the control group, 7.42 mg KOH per g in the group with a proportion of 4% alfalfa meal, and 11.18 mg KOH per g in the group with proportion 6% alfalfa meal. An acid number of fat of stored meat for 18 months was 5.90 mg KOH per g in the control group, 4.65 mg KOH per g in the group with a proportion of 4% alfalfa meal, and 7.07 mg KOH per g in the group with a proportion of 6% alfalfa meal. Chicken meat is notably sensitive to lipid oxidation because of its high content of polyunsaturated fatty acids. Legislation in Title 5 of Part 3 of the Codex Alimentarius of the Slovak Republic and the Government Regulation No. 286/2003 Coll. in the Annex 4 in Part B provides the requirements for animal fats and meat products. Regulation of the European Parliament and Council (EC) No. 853/2004 lays down specific hygiene rules for food of animal origin. In particular, determination of free fatty acids content of rendered animal fat (tallow, lard, other animal fat). Legislative regulation does not contain requirements for the quality of chicken meat, the acid number of fat of fresh or frozen chicken meat, respectively. Chicken meat is preferred over other kinds of meat. It is characterized by certain dietary and nutritional properties that consumer prefers. A price of this kind of meat remains attractive. In terms of human health, oxidative stability of chicken meat is important, especially of stored meat. In general terms, the various food additives are currently used to maintain the food stability. Great attention is currently paid to additives of natural origin. Similar focus is presented in our study. We can state, on the basis of the oxidative stability results of chicken meat, that natural feed component has its justification. This issue requires further research.

Keywords: broiler chicken; alfalfa meal; stored meat; oxidation; acid number of fat.

INTRODUCTION

In recent years, poultry meat becomes more preferred food not only in Europe but especially in China, Brazil and India. Consumption of poultry meat increased even in Africa. If we compare global production of meat, total poultry production nearly converges to total pork production. According to forecasts, poultry production of China will increase by 37% and ten times in India by 2020. Its popularity is mainly due to the efficiency of fattening, short time of fattening and because of small area for breeding, relatively. Of course, even for dietetic properties of poultry meat. Lipids have an important role in food product quality, making them more desirable by improving the organoleptic properties of flavour, colour and texture. In addition, they confer nutritive value on the product, constituting a source of metabolic energy, essential fatty acids and fat-soluble vitamins. On the other hand, the lipid components are susceptible to attack by molecular oxygen **(Baggio, 2006)**. Poultry meat is in terms of dietary properties and nutritional value very interesting because of the high content of protein, minerals and vitamins and low percentage of fat. Meat poultry contains an average of 19.7 to 22.3% of protein, 1.4 to 22.16% of lipid, 57 to 75.25% of water, 1.00 to 1.07% of ashes **(Benková, 2009).** The composition of the meat varies depending on the type of animal, breed, gender, age and nutrition. Structure and composition of muscle depends on the method of meat processing, which affects biochemical, organoleptic and technological properties of the meat **(Pipek, 1998; Brezina et al., 2001; Benková, 2009).** The biological value of dietary fats is assessed as their digestibility, content in fat-soluble vitamins, essential fatty acids, cholesterol, and according to the proportion of each type of fatty acids. The quality and type of fat affects the

appearance, taste and especially energy and nutrient content of food **(Jurkovičová, 2008).** The lipids are presented in meat especially as fatty acid esters of glycerol. They contain lipophilic vitamins, lipids, mainly phospholipids, and essential fatty acids. The fats are rated negatively for their high energy. Fat of meat influenced a tenderness and fragility of meat **(Ingr et al., 1993; Pipek 1995)**. Fat has an important role in the formation and in the texture of meat. Fat is a source of energy and the fat also affects tastiness properties of the meat **(Pipek, 1991; Pipek, 1998)**. Poultry fat contains higher amounts of polyunsaturated fatty acids (PUFAs) than other fat of animals for slaughter. PUFAs are responsible for a lot of oxidative changes, for example as changes of organoleptic properties and shelf life **(Korimová et al., 2000; Turk et al., 2000; Bou et al., 2001)**. Long chain polyunsaturated fatty acids are conditionally essential nutrients for adequate growth, development and function in humans **(Gill, 2012)**. Chicken meat had a lower proportion of saturated (36.4 ±3.6%; p <0.001) and a higher proportion of PUFAs (21.3 ±3.5%; p <0.001) **(Almeida, 2006)**. Because chicken meat has high content of PUFAs **(Botsoglou et al., 2002)**, is notably sensitive to lipid oxidation. Thigh meat, as compared to breast meat, is particularly vulnerable because of its higher fat content **(Jensen et al., 1998)**. Among them, omega-3 PUFAs (ω-3 PUFAs) have gained popularity due to their various health promoting and diseases preventing attributes. For example, ω-3 PUFAs are reported to be highly effective against cardiovascular diseases, cancer and other metabolic diseases **(Wang, 2012; Gulhan, 2014)**. Long chain ω-3 PUFAs eicosapentaenoic and docosahexaenoic were observed only in dark chicken meat (23.0 ±3.0 and 14.0 ±1.0 mg per 100 g **(Almeida, 2006)**. The oxidation of fats is one of the major problems in the meat industry due to a decrease in quality flavor and loss of nutritional value **(Ladikos and Lougovois 1990; Ahn et al., 1992)**. Lipid oxidation causes loss of nutritional and sensory values as well as the formation of potentially toxic compounds that compromise meat quality **(Cortinas, 2005)**. Oxidation leads to oxidative rancidity and involves oxygen attack on glycerides whereas hydrolysis leads to hydrolytic rancidity and it involves hydrothermal or enzymic (lipase) hydrolysis to free fatty acids and other products. There are some factors affecting the development of rancidity such as the degree of unsaturation of the oils, heat, prooxidants, light, certain enzymes (lipoxygenases), moisture content and availability of oxygen **(Özogul et al., 2006)**. Secondary oxidation products such as aldehydes, ketones and esters, are responsible for the increased depreciation and deviation from the natural flavors **(Ladikos and Lougovois, 1990)**. During storage maturation occurs in deep frozen raw meat over time **(Pipek, 1992)**. When the meat is stored for a long time, maturing phase passes into a deep autolysis. This action is undesirable. There is degradation of the proteins on oligopeptides and amino acids. Meat acquires an unpleasant taste and undergoing to hydrolysis of fats **(Kadlec et al., 2002)**. Oxidation might also play a role in controlling proteolytic activity of enzymes and could be linked to meat tenderness. The oxidative stability of meat depends upon the balance between anti- and pro-oxidants, including the concentration of PUFAs **(Mercier, 2004).** Fatty acids are released by the hydrolysis of fats **(Velíšek, 2009).**

Deep autolysis of slaughtered animals is not desired, but it is not possible to completely prevent or eliminate the microbial proteolysis. Deep autolysis catalyzed by the native enzyme can take place and to continue in relatively isolated state (**Steinhauser et al., 1995)**. The negative consequences of lipid oxidation can be overcome by the use of antioxidants in the diet **(Cortinas, 2005)**. Meat composition of PUFAs changed at the animals, which were fed with diets with increased unsaturated fats **(Lin et al., 1989; Ajuyah et al., 1993; Ahn et al., 1995; Mooney et al., 1998; Lo'pez Ferrer et al., 1999)**.

The aim of the study was to investigate the oxidative changes of stored meat at -18 °C depending on feeding of alfalfa meal in the broiler chickens.

MATERIAL AND METHODOLOGY

The feeding experiment was performed in commercial poultry farm with a final hybrid chickens Ross 308, which is used for meat production. A space for the purpose of the experiment was situated in front of the hall, with a deep bedding system of breeding. This space was divided into three equal parts to meet the requirements of the standard distribution. 100 pieces of one-day-old chickens were placed within each group. Conditions corresponding to standards of Decgree of Ministry of Agriculture of the Slovak Republic no. 2136/2004-100 of 23 August 2004 were created for our feeding. The broiler chickens were fed by standard feed mixtures of soy-cereal type *ad libitum*. Standard feed mixtures, usually used in practical conditions, were used in the control group of broiler chickens. In the 1st and 2nd experimental groups were used similar feed mixtures as in control group, only feed mixtures of 1st experimental group was enriched by 4% proportion of alfalfa meal at the expense of the wheat, and feed mixtures of 2nd experimental group was enriched by 6% proportion of alfalfa meal at the expense of wheat. The experimental period was divided into 3 phases: starter with starter feed mixtures for broiler chickens at the age of 1 to 18 days, grower with grower feed mixtures for broiler chickens at the age of 19 to 31 days, finisher with finisher feed mixtures for broiler chickens at the age of 32 to 38 days. The feed mixtures were produced according to law no. 440/2006 Coll. At the end of the experiment (day 38), 6 pcs of broiler chickens from each group with an average live body weight over 1 800 g were randomly selected. A slaughtering of broiler chickens was realized by human rapid cut of the carotid artery (*Ateria carotis communis*). Subsequently, feathers as well as internal parts of broiler chickens were mechanically removed. A carcass was prepared. The slaughtering was carried out at the Department of Evaluation and Processing of Animal Products, Faculty of Biotechnology and Food Sciences, SUA in Nitra. Chicken carcasses were packaged in plastic containers and stored at -18 °C for 12 and 18 months. Chemical analysis of samples was realized after the storage period at the Department of Food Hygiene and Safety FBFS SUA in Nitra. The samples for chemical analysis consisted of identical proportion of breast and thigh muscle, and about 1 cm^2 of skin with subcutaneous fat. Fat from the meat was obtained after samples drying.

A fat was determined by extraction by means of laboratory instrument Det N Gras Selecta P. An acid number of fat was determined from obtained fat. Acid value of fat was determined after dissolution of fat in the extract ethanol-diethyl ether in a 1:1 alkalimetric titration against phenolphthalein. The extracted fat was slightly heated and fat was dissolved in 25.0 ml of ethanol-ether. The content in extraction flask was titrated with a few drops of the indicator with the potassium hydroxide solution until it turned to slight pink color. An acid number of fat was expressed in mg KOH per g. A fat acid value is the number of mg of potassium hydroxide required to neutralize free fatty acids per gram of fat extracted from the extracting agent. Mathematical and statistical evaluation of the results was realized by the SAS Enterprise Guide Version 1.5 system program.

RESULTS AND DISCUSSION

An acid number of fats were determined in the stored chicken meat for 12 months, which ranged from 4.72 to 10.50 mg KOH per g of fat in the control group. Due to the large margin of determined values, the coefficient of variation was 36.86%. In the experimental group with proportion of 4% alfalfa meal, the acid number of fat was determined in the range of 4.69 to 10.49 mg KOH per g with coefficient of variation 28.04%. In the group with proportion of 6% alfalfa meal, the acid number of fat was determined in the range of 5.56 to 20.61 mg KOH per g fat and the coefficient of variation was 47.99%. The differences in acid number of fat among the groups were not statistically significant (p >0.05) after 12 months of meat storage. An acid number of fats determined in the stored meat for 18 months varied from 4.35 to 6.56 mg KOH per g fat with coefficient of variation 17.60% in the control group of broiler chicken, which were fed by standard feed mixtures. The acid number of fats in the range of 4.01 to 10.69 mg KOH per g fats was obtained in the meat of broiler chickens, which were fed by feed mixtures with proportion 4% of alfalfa meal in the feed mixtures. If proportion of alfalfa meal formed 6% of feed mixtures, the acid number of fat was measured in the range of 3.29 to 10.33 mg KOH per g fat, and the coefficient of variation was 38.68%. The differences in acid number of fat among the groups were not statistically significant (p >0.05) after 18 months of meat storage. Slovak legislation in Title 5 of Part 3 of the Codex Alimentarius of the Slovak Republic and the Government Regulation No. 286/2003 Coll. in the Annex 4 in Part B set out the requirements for animal fats and meat products. Regulation of the European Parliament and Council Regulation (EC) No. 853/2004 lays down specific hygiene rules for food of animal origin. In particular, determination of free fatty acids contents of rendered animal fat (tallow, lard, other animal fat). Legislative regulation does not contain requirements for the quality of poultry meat, the acid number of fats of fresh or frozen poultry meat, respectively. It was demonstrated in several studies that feeding of oxidized diets to broilers resulted in negative effects on bird performance **(Cabel et al., 1988; Engberg et al., 1996),** on oxidative stability of tissues and membranes **(Asghar et al., 1989; Lin et al., 1989; Jensen et al., 1997; Grau et al., 2001a)** and on shelf-life of meat during storage **(Sheehy et al., 1994; Rhee et al., 1996; Sheldon et al., 1997; Grau et al., 2001b)**. Based on studies by many authors, it can be assumed that the composition of the feed mixture has a significant effect on

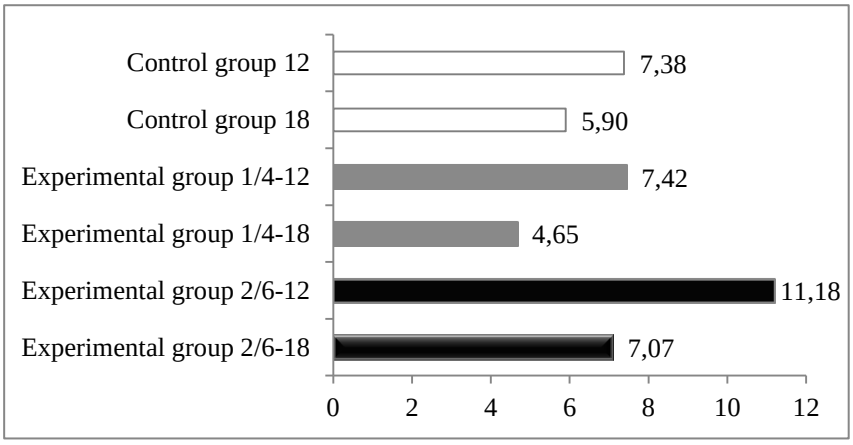

Figure 1 An acid number of fat of stored meat depending on proportion of alfalfa meal in feed mixtures of broiler chickens.

Control group 12 – stored chicken meat for12 months at -18 °C,
Control group 18 – stored chicken meat for 18 months at -18 °C,
Experimental group 1/4-12 – stored chicken meat for 12 months at -18 °C of group with proportion 4% of alfalfa meal in feed mixtures,
Experimental group 1/4-18 – stored chicken meat for 18 months at -18 °C of group with proportion 4% of alfalfa meal in feed mixtures,
Experimental group 2/6-12 – stored chicken meat for12 months at -18 °C of group with proportion 6% of alfalfa meal in feed mixtures,
Experimental group 2/6-18 – stored chicken meat for18 months at -18 °C of group with proportion 6% of alfalfa meal in feed mixtures.

the chemical composition of meat broiler chickens, sensory quality and oxidative stability **(Horwitt, 1986; Wood, 2004; Hugo, 2009; Bobko et al., 2012; Gibbs et al., 2013)**. Lipid oxidation is considered the main cause of quality damages related to flavor, color, taste, and nutritional composition of meat and meat products **(Mielche and Bertelsen, 1994; Gray et al., 1996)**. The total amount of fat is a major factor of quality of fatty acids. The effects of fatty acid composition on meat quality are also reviewed. Fatty acid composition determines the firmness/oiliness of adipose tissue and the oxidative stability of muscle, which in turn affects flavour and muscle colour. Vitamin E is an essential nutrient, which stabilises PUFAs and has a central role in meat quality **(Wood, 2008)**.

Free fatty acids are produced by secondary enzymatic cleavage of triglycerides **(Koman et al., 1989)**. Higher values of acid number of fat may be according to **Sopková et al. (2007)** caused by the hydrolytic degradation of the fatty substance. **Guteriez (2013)** studied susceptibility of unsaturated fatty acids to oxidation. It is related to the degree of unsaturation, polyunsaturated fatty acids, they are more prone to oxidation than monounsaturated fatty acids. Equally he demonstrated higher susceptibility of polar lipids to oxidation as compared with neutral lipids. Lipolysis can be responsible for a decrease of content polar lipids and releasing free fatty acids. Few authors deal with the assessment of chicken fat by acid number of fat. The authors are more concerned with assessing the quality of fats by thiobarbituric acid reactive substances (TBARS) values. Physical agents (heat, oxygen, light) and chemical factors (content of certain metals) play an important role in the development of oxidation **(Ozturk and Cakmakci, 2006)**. Top of lipoperoxidation is derived from the free radical, such as nitrogen dioxide **(Kanner et al., 1987)**. A speed fat oxidation of meat also depends on the presence of a prooxidants and antioxidants **(Tichivangana and Morrissey, 1985; Ruiz et al., 1999)**.

CONCLUSION

Chicken meat is preferred over other kinds of meat. It is characterized by certain dietary and nutritional properties that consumer prefers. A price of this kind of meat remains significant. In terms of human health, oxidative stability of chicken meat is important, especially of stored meat. In general terms, the various food additives are currently used to maintain the food stability. Great attention is currently paid to additives of natural origin. Similar focus is presented in our study. We can state, on the basis of the oxidative stability results of chicken meat, that natural feed component has its justification. This issue requires further research.

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Contact address:

Ing. Jana Tkáčová, Ph.D., Slovak University of Agriculture in Nitra, Tr. A. Hlinku 2, Faculty of Biotechnology and Food Sciences, 949 76 Nitra, Slovakia, E-mail: tkacova.j@gmail.com

prof. Ing. Mária Angelovičová, Ph.D., Slovak University of Agriculture in Nitra, Tr. A. Hlinku 2, Faculty of Biotechnology and Food Sciences, 949 76 Nitra, Slovakia, E-mail: maria.angelovicova@gmail.com

associate prof. Peter Haščík, Ph.D., Slovak University of Agriculture in Nitra, Tr. A. Hlinku 2, Faculty of Biotechnology and Food Sciences, 949 76 Nitra, Slovakia, E-mail: peter.hascik@uniag.sk

Ing. Marek Bobko, Ph.D., Slovak University of Agriculture in Nitra, Tr. A. Hlinku 2, Faculty of Biotechnology and Food Sciences, 949 76 Nitra, Slovakia, E-mail: marek.bobko@uniag.sk.