OXIDATIVE STABILITY OF MEAT AND MEAT PRODUCTS AFTER FEEDING OF BROILER CHICKENS WITH ADDITIONAL AMOUNTS OF VITAMIN E AND ROSEMARY

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SUMMARY

Lipid oxidation is a major cause of meat quality deterioration. Lipid oxidation is an important determinant of shelf life of meat and meat products. Antioxidants are natural or synthetic substances used to prevent lipid oxidation. Meat protection, primarily against lipid components, is possible by addition of antioxidants to feed mixes. This is the way to ensure oxidative stability of meat fats during the post-slaughter processing of carcasses and storage of meat. The aim of our project was to investigate the protective effect of natural antioxidants rosemary (500 mg.kg^{-1}) and vitamin E (40 mg/bird/day) added to feed mixes and fed to broiler chickens from day 21 to day 42 of life. Afterwards, oxidative stability of ground meat and poultry sausages was determined. After meat processing, samples were collected, frozen and stored in a freezer at – 18°C for 14 days. Frozen breast muscles of individual groups of chickens were ground and stored in a refrigerator at 1°C for 14 days. Poultry sausages were produced from frozen thigh muscles of individual groups of chickens. Synergistic effect of vitamin E added to feed mixes and rosemary added to ground meat was evaluated. Before grinding of meat, meat samples of broiler chickens saturated with vitamin E were enriched with 50 mg.kg^{-1} of rosemary powder FlavorGuard P (Christian Hansen, Denmark). The addition of vitamin E and rosemary powder FlavorGuard P to feed mixes for broiler chickens resulted in increased oxidative stability of fats in meat. Extra addition of rosemary to meat enriched with vitamin E suppressed more significantly the oxidative processes and increased oxidative stability of poultry meat and poultry sausages. Use of frozen meat and low storage temperature (1°C) of ground meat also significantly suppressed oxidative processes.

Key words: *Broiler meat, lipid oxidation, malondialdehyde, rosemary, vitamin E*

INTRODUCTION

Lipids are important components of food and they have insufficient status in human nutrition. Lipids influence nutrition, energy and sensory value of food. Food with a higher fat content is preferred, because fat improves taste and aroma of foods and makes them more attractive for consumers. Lipids are also the source of lipophilic vitamins (A, D, E, K) and polyunsaturated fatty acids. Important are dominantly ω3-polyunsaturated fatty acids. Nutrition enriched with ω3-polyunsaturated fatty acids (PUFA), reduces the risk of development of cardiovascular diseases, hypertension and arthritis (Echarte et al., 2001).

Apart from these positive factors, fat consumption in food is accompanied by some negative factors with direct or indirect impacts on human health. Among these negative factors, in the first place should be mentioned additives, which are added to fats to correct their properties (stabilisers, emulsifiers, antioxidants, colouring agents, etc.) and also the consumption of adverse metabolites present in fats or formed from fats (Bystrický and Dičáková., 1998).

Lipids and PUFA are mainly sensitive to various

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changes as hydrolysis and oxidation. Lipid oxidation is an autocatalytic process occurring in food and biological membranes, which leads to significant damage of food quality (Eriksson and Na, 1995). This process is supported by various factors, the most important being the content of PUFA. They can be used as substrates for initiation of oxidative process. The main catalysts of oxidation are highly reactive free radicals as superoxide anion (O^{2}-), hydroxyl radical (OH°), proxyl radicals (ROO°), which contain one or more free electrons. Other factors, which support lipid oxidation, are substances as myoglobin, hemoglobin, cytochromes and transport metals (Fe). They are produced during the process of meat ripening. Level of oxidation is also increased, because of the damage of muscle membranes during processes such as removal of bones, meat grinding or cooking (Ladikos and Lougovois, 1990).

Products formed during the process of fat degradation must be considered very carefully, because of their negative impact on human health. They are included into mechanisms of such diseases like arteriosclerosis, cancer, suppression of immunity, Parkinson's disease, Alzheimer's disease and numerous cardiovascular diseases (Patthy et al., 1995).

Presently, great attention is being paid to lipid protection against oxidation. There are many ways to achieve this aim: decrease of ambient temperature, prevention of air access, elimination of factors supporting oxidation and addition of antioxidants (Bystrický and Dičáková, 1998). Nowadays, the most effective way of inhibition of oxidative processes is with the use of antioxidants (Turek et al., 2000). Antioxidants are used for the prolongation of shelf life of food, and to ensure health safety. They interfere with the lipid oxidation process and other oxidative labile substances in following way:

a) React with free radicals (primary antioxidants) or reduce formed hydroperoxides (secondary antioxidants),

b) Bind with metals into complexes, which support oxidation processes,

c) Eliminate oxygen.

Primary antioxidants are, for instance, ascorbic acid and its derivatives, tocopherol, phenol substances and propyl gallate. During recent years, antioxidants based on rosemary extracts have become very popular all over the world in the field of meat processing and meat products production. Anti-oxidative effect in selected types of preparations is ensured by natural phenol substances present in spices (Turek et al., 2000).

The aim of our project was to evaluate the influence of natural antioxidants, applied in feed mixes, on oxidative changes in fats of ground poultry meat and poultry sausages during storage under freezing conditions. The synergetic effect of vitamin E used in feed mixes and rosemary powder added to meat before grinding and their influence on oxidative stability of fats was also determined.

MATERIAL AND METHODS

In our experiment, meat of broiler chickens (n = 120), fed from day 21 of the study with feed mixes saturated with natural antioxidants, was used. In group R (n = 40) rosemary powder FlavorGuard P (Christian-Hansen, Denmark) was added to feed mixes at the dose of 500 $mg.kg^{-1}$. In group E (n = 40) vitamin E (Hydrovit E forte, PHARMAGAL, Slovak Republic) was incorporated in the drinking water at the daily dose of 40 ml/bird. The control group (K, n = 40) was fed feed mixes without the addition of antioxidants. Immediately after the slaughtering process, bones and skin were removed from poultry meat.

Breast muscles were packed into PE bags (1 kg), frozen and stored for 14 days at -18°C. Frozen samples were cut on the cutter and ground for five minutes. For the evaluation of the synergistic effect of vitamin E added to feed mix and rosemary powder FlavorGuard P added before grinding, rosemary powder at the dose of 50 $mg.kg^{1}$ was mixed into meat saturated with vitamin E. Ground meat was packed in PE bags and kept in a refrigerator at 1°C for 14 days.

The thigh muscles of individual broiler chickens were frozen at –18°C, stored for fourteen days and used for production of poultry sausages. The following four groups of samples were collected: K - control group; E - meat of broiler chickens fed feed mixes saturated with vitamin E; R - meat of broiler chickens fed feed mixes saturated with rosemary

Table 1. Poultry sausage – composition

Ingredients (g)	K	R	E	E+R
Meat	2000	2000	2000	2000
Flour	90	90	90	90
Salting mixture	55	55	55	55
Water	300	300	300	300
Black spice	5	5	5	5
Red spice	4	4	4	4
FlavorGuard P (mg)	-	-	-	100

K - control; E - meat of broiler chickens fed vitamin E (40 mg/bird/day); R - meat of broiler chickens fed rosemary powder (500 mg.kg^{-1}); E + R - meat of broiler chickens fed vitamin E (40 mg/bird/day) and with addition of rosemary powder (50 mg.kg^{-1}) before grinding

Table 2. Determination of TBA, presented as a malondialdehyde, in ground poultry meat (breast, n = 6) stored at 1°C, after addition of antioxidants in feed

Duration of storage (day)	Malondialdehyde (mg.kg^{-1})			
	K	R	E	R+E
0	0.031 ± 0.005	0.029 ± 0.004	0.030 ± 0.004	0.025 ± 0.003
3	0.039 ± 0.008	0.030 ± 0.005	0.031 ± 0.007	0.027 ± 0.003
7	0.068 ± 0.019	0.056 ± 0.007	0.052 ± 0.002	0.031 ± 0.007
9	0.079 ± 0.007	0.043 ± 0.013	0.042 ± 0.003	0.032 ± 0.004
14	0.227 ± 0.020	0.177 ± 0.008	0.151 ± 0.009	0.081 ± 0.010

K - control; E - meat of broiler chickens fed vitamin E (40 mg/bird/day); R - meat of broiler chickens fed rosemary powder (500 mg.kg^{-1}); E + R - meat of broiler chickens fed vitamin E (40 mg/bird/day) and with addition of rosemary powder (50 mg.kg^{-1}) before grinding

Table 3. Determination of malondialdehyde in poultry sausages (n = 6) after addition of antioxidants

Duration of storage (day)	Malondialdehyde (mg.kg^{-1})			
	K	R	E	R + E
1	0.444 ± 0.003	0.378 ± 0.011	0.397 ± 0.007	0.326 ± 0.009
7	0.430 ± 0.012	0.391 ± 0.008	0.342 ± 0.008	0.319 ± 0.011
14	0.525 ± 0.008	0.399 ± 0.015	0.359 ± 0.012	0.304 ± 0.005

K - control; E - meat of broiler chickens fed vitamin E (40 mg/bird/day); R - meat of broiler chickens fed rosemary powder (500 mg.kg^{-1}); E + R - meat of broiler chickens fed vitamin E (40 mg/bird/day) and with addition of rosemary powder (50 mg.kg^{-1}) before grinding

powder, and E+R - meat of broiler chickens fed feed mixes saturated with vitamin E and with the addition of rosemary powder at the dose of 50 mg.kg-1 before grinding.

Meat (2 kg of each group) was cut on the frozen meat cutter. After the addition of a salting mixture, and in group E+R also rosemary powder, meat was ground for two minutes. The addition of ingredients according to recipe (Table 1) followed, and meat was ground for additional three minutes. After mixing of meat with ingredients, filling into casing of pork intestines followed. Centre of sausages was subjected to thermal treatment at 70°C for ten minutes. Total time of thermal treatment was 3.5 hours.

The decomposition of fats was evaluated by measurement of the thiobarbituric acid value (TBA), and the expressed amount of malondialdehyde calculated for one kilogram of sample. Evaluation of TBA was performed according to Grau et al. (2000) and measured by spectrophotometric method at 532 nm (Helios γ, v. 4,6, Thermo spectronic, Great Britain). The examination of samples was carried out at 0, 3, 7, 9 and 14 days of storage at 1°C. The values of selected parameters, recorded in tables, are average values derived from calculation of six samples of meat and meat products.

RESULTS AND DISCUSSION

The lipids in poultry exhibit a higher degree of unsaturation compared with red meat, because of a relatively high content of phospholipids. The degree of unsaturation of phospholipids in subcellular membranes is an important factor in the determination of oxidative stability of meats. The oxidative potential increases as the degree of unsaturation of lipids in meat increases (Coetzee and Hoffman, 2001). In relation to the character of the auto-oxidation process, the effect of antioxidants is more significant the sooner they are applied. The ideal situation is when fats are protected immediately after the slaughter of animals (Turek et al. 1999, Pípová et al., 1995). Such protection can be achieved by feeding animals with feed containing antioxidants (Coetzee and Hoffman, 2001). The oxidation of lipids is influenced by the addition of antioxidant substances. The practical application of antioxidants can be difficult from the point of view of hygiene and technology. It is much better when natural antioxidants are incorporated in feed mixes (Kušev et al., 1996). Feeding of poultry with a higher level of natural dietary antioxidants provides the poultry industry with a simple method for improvement of the oxidative stability, sensory quality, shelf life and acceptability of poultry meats. Vitamin E and rosemary belong to substances with significant anti-oxidative activity. They are important for the stabilisation of optimal health status of poultry and as prevention against diseases. Higher concentrations of vitamin E in feed mixes have a positive impact also on feed conversion, increased daily weight gain and average weight of broilers, and also cause reduced feed consumption (Coetzee and Hoffman, 2001).

Results of the analysis of malondialdehyde in frozen breast muscles, ground for five minutes and stored at 1°C are shown in table 2. Immediately after grinding, the MDA content in all samples was low. That was probably due to the fact that the temperature of meat after grinding of -3.5°C and during the period of storage in a refrigerator did not exceed 1°C. During the first nine days of storage, only a slight increase in MDA concentrations was recorded in all samples. The highest amount of MDA on day 9 of storage was in the control group (0.079 mg.kg-1). The lowest amount of MDA in samples and the best anti-oxidative effect were found in meat samples with a combination of antioxidants, vitamin E and rosemary powder. Subsequent storage of samples caused an increase in MDA concentrations. The addition of antioxidants to feed mixes increased the oxidative stability of lipids in meat in comparison with the control group. Better oxidative stability was also evident in meat samples of broilers given in feed vitamin E and with the addition of rosemary alone to meat. The most significant effect was achieved by a combination of vitamin E saturated in feed mixes and rosemary powder added to meat before grinding. A significant anti-oxidative effect of added antioxidants was confirmed.

The method of meat processing and temperature of storage have a significant effect on the oxidative stability of lipids (Ladikos and Lougovois, 1990; Bystrický et al., 1993). Reduction of the ambient temperature results in a slower process of all

chemical reactions, including lipid oxidation. By the combination of freezing and addition of antioxidants, primarily a combination of vitamin E and rosemary (Korimová et al., 1998; Marcinčák et al., 2003), it is possible to significantly suppress the process of lipid oxidation.

The manufacturing processes of meat products (cutting, grinding, and mixing) cause degradation of the muscle membrane system and have a strong impact on the oxidation of intracellular fat, primarily phospholipids (Bystrický and Dičáková, 1998). Thermal treatment makes oxidative processes faster, what significantly changes the value of thiobarbituric acid. The level of oxidative damage of lipids in the manufacture and storage of poultry sausages is presented in table 3. Immediately after the production of sausages, a higher level of oxidative damage of lipids was recorded. That can be attributed to heat treatment of poultry products. The highest levels of MDA were recorded in products produced from meat of broilers of the control group (0.444 mg.kg^{-1}). The lowest oxidative damage of lipids was recorded in sausages produced from meat of broiler chickens fed vitamin E and with addition of rosemary powder before grinding. Storage of products at 4°C influenced only the MDA concentrations in the control group, when a significantly increased concentration was recorded (0.525 mg.kg^{-1}). On the other hand, an opposite effect was noted in other groups (E, E + R) or a stabilisation (R) of the MDA levels.

The anti-oxidative effect of rosemary is based on its ability to inactivate free radicals produced during the auto-oxidation process (Pokorný et al., 1998). Rosemary extracts, as antioxidants, were successfully used for the stabilisation of smoked products (Korimová et al., 2000), canned meat (Guntensperger et al., 1998), pork meat (Marcinčák et al., 2003), poultry meat and products (Karpinska et al., 2000). Results obtained by these authors are consistent with the results recorded in our study. Anti-oxidative activity is significantly evident in systems containing tocopherol (Pokorný et al., 1998).

CONCLUSION

Use of vitamin E and rosemary powder FlavorGuard P in feed mixes given to broiler chickens from day 21 of life increased the oxidative stability of fats in meat. Significant improvement of the fat oxidative stability was achieved with the addition of rosemary powder to meat before grinding in case of broiler chickens given vitamin E in feed. By the combination of freezing and addition of antioxidants, primarily a combination of vitamin E and rosemary, it is possible to suppress significantly the oxidation process during the production of meat products.

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SAŽETAK

OKSIDATIVNA STABILNOST MESA I MESNIH PROIZVODA NAKON HRANJENJA TOVNIH PILIĆA DODATNIM KOLIČINAMA VITAMINA A I RUŽMARINA

Oksidacija lipida je glavni uzrok smanjenja kvalitete mesa i značajan čimbenik održivosti mesa i mesnih proizvoda. Antioksidanti su prirodne ili sintetske supstancije koje se koriste za sprečavanje oksidacije lipida. Dodavanjem antioksidanata u krmne smjese moguće je osigurati oksidativnu stabilnost masti mesa tijekom klaoničke obrade i kasnijeg skladištenja mesa. Cilj našeg projekta je bio ispitati zaštitni učinak prirodnih antioksidanata ružmarina (500 mg.kg^{-1}) i vitamina E (40 mg/pilić/dnevno) dodavanih smjesama kojima su hranjeni tovni pilića od 21. do 42. dana života. Potom je određivana oksidativna stabilnost mljevenog pilećeg mesa i kobasica od pilećeg mesa. Nakon obrade mesa, uzorci su smrznuti i pohranjeni u zamrzivač na -18°C kroz 14 dana. Smrznuti mišići prsa pojedinih grupa pilića su samljeveni i pohranjeni u hladnjak na temperaturi od 1°C kroz 14 dana. Pileće su kobasice proizvedene od smrznutih mišića bataka pojedinih grupa pilića. Izvršena je procjena sinergističkog učinka vitamina E dodanog u smjese za piliće, te ružmarina koji je pridodan mjevenom mesu. Prije mljevenja, uzorci mesa tovnih pilića koji su dobivali dodatne količine vitamina E u hrani, obogaćeni su s 50 mg.kg^{-1} ružmarina u prahu FlavorGuard P (Christian Hansen, Danska). Primjena vitamina E i ružmarina u prahu FlavorGuard P u smjesama za tovne piliće rezultirala je povećanom oksidativnom stabilnosti masti u mesu. Dodatna primjena ružmarina u mesu obogaćenom vitaminom E imala je značajniji supresivni učinak na oksidativne procese i povećala oksidativnu stabilnost pilećeg mesa i pilećih kobasica. Upotreba

smrznutog mesa i niske temperature pohrane mljevenog mesa (1°C) također su imale značajan supresivni učinak na oksidativne procese.

Ključne riječi: *Meso tovnih pilića, oksidacija lipida, malondialdehid, ružmarin, vitamin E*

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VITAMINI U ANIMALNIM NAMIRNICAMA

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SAŽETAK

Vitamini su esencijalni i nužni sastojci hrane. Različitog su kemijskog sastava, a najvećim dijelom biljnog porijekla. Potpuni nedostatak nekog vitamina dovodi do karakterističnog poremećaja koji se naziva avitaminoza, ali u praksi su mnogo češća pojava djelomičnog defekta vitamina tzv. hipovitaminoze. One zavisno od stupnja vitaminskog nedostatka, dovode do manjih ili većih poremećaja važnih životnih procesa i funkcija, kao što su razmnožavanje, rast, metabolizam, funkcije pojedinih organskih sustava. Vitamini unijeti hranom ili osigurani pomoću mikroorganizama (domaće životinje) deponiraju se u iznutricama i mišićju, a domaće životinje ih izlučuju mlijekom. Otuda su i namirnice životinjskog porijekla bogat izvor vitamina. Nazočnost dovoljne količine vitamina u prehrani jedno je od načela zdrave prehrane. Prehrana i opskrba vitaminima poseban je problem u današnje vrijeme.

Ključne riječi: *vitamini, animalne namirnice, prehrana*

UVOD

Frederick Hopkins je 1906. godine ukazao na potrebu ljudskog organizma za nekim tvarima, tada

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