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Antioxidant Effect of Vitamin E and Selenium on Omega-3 Enriched Poultry Meat

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Abstract. Oxidation of the meat lipids can be effectively controlled with antioxidants. The objective of this research was to examine the effect of addition of selenium and vitamin E in broilers diet enriched with n-3 fatty acids on meat quality. The dietary treatments were based on the administration of vitamin E (200 mg a-tocopheryl acetate/kg diet) alone or in addition with selenium (0,3 mg sodium selenite/kg diet) to a 15% flax seed diet. The oxidative stability, evaluated by thiobarbituric acid reactive substances (TBARS), was determined after 120 days of storage at –20 °C. In addition, fatty acids composition and vitamin E level were evaluated. The addition of antioxidants in broilers diet did not affect carcass characteristics, with the exception of abdominal fat weight. The flax seed from diet increased the concentration of the omega-3 fatty acids 18:3. PUFAs were higher when vitamin E and Se were supplemented to the broiler diets. The concentration of α-tocopherol in poultry meat increased with the increase in its dietary supplementation. TBARS values decreased when antioxidants were added, but no differences between treatments containing the two antioxidants were observed. The results of this study showed that vitamin E and selenium can be added to broiler diet to protect against meat oxidation during storage.

Keywords: oxidation, lipids, α-tocopherol, selenium, poultry meat, n-3 PUFA

INTRODUCTION

A strategy to increase n-3 polyunsaturated fatty acids (n-3 PUFA) intake in the human diet includes consumption of enriched products. However, by using high levels of unsaturated fatty acids in animals diet, the degree of unsaturation of muscle membrane lipids increases, leading to a reduced oxidative stability of the meat. In general, oxidation products are considered as inducers of cardiovascular and atherogenesis problems. Lipid oxidation contributes to undesirable changes in a number of meat quality parameters, including loss of texture, flavor, water-holding capacity (Morrissey and Kiely, 2006) and to development of rancid odors and flavors, being the major causes of meat quality deterioration during storage and shortening the shelf life. As a result, such meat is not assumed to be fresh by the consumer.

In raw meat products, the primary factors that influence lipid oxidation include fatty acid composition, dietary fat quality, endogenous prooxidative and antioxidative constituents, water activity and nonmeat additives (prooxidative and antioxidative) (Rojas and Brewer, 2007). Antioxidants have been successfully added to livestock feeds in order to increase meat oxidative stability. There is a tendency towards the use of natural antioxidants in detriment of

synthetic ones. It has been suggested that a combination of different antioxidants may be more effective in retarding lipid oxidation rather than the use of a single antioxidant (Barroeta, 2007).

In this respect, dietary antioxidants such as vitamin E, carotenoids, herbal extracts and selenium (Se) are beneficial in preventing the detrimental effects of PUFA-enriched diets. Vitamin E and Se are key components of the antioxidant system, reducing lipid peroxidation. Supplementation of vitamin E significantly improved the meat stability against oxidative deterioration. Vitamin E is the primary lipid-soluble antioxidant found in foods and human blood and tisuess. It is well known that vitamin E inhibits the process of lipid peroxidation in oils and in the biological lipid-protein complexes such as biological membranes or circulating lipoproteins (Fellenberg and Speisky, 2006). Selenium plays an important role in the antioxidant defence system due to its requirement by the Se-dependent GSHPx, which is involved in cellular antioxidant protection. It has been suggested that there is a synergistic relationship between Se and vitamin E, because GSHPx continues the work of vitamin E by detoxifying hydroperoxides (Stanley, 1998).

The aim of this study was to investigate the individual and combined effect of different types of dietary antioxidants, vitamin E and sodium selenite, on lipid oxidation of n-3 enriched broiler meat during storage.

MATERIALS AND METHODS

Animals and diets

A total of 20 broiler chickens one day old, obtained from a commercial hatchery, were used for the experiment. Over a period of 21 days the birds were fed a starter diet, having free access to feed and water. Starting from 21 days, the birds were weighed and randomly divided; five birds per pen were allocated to 4 pens. Each of the 4 floor pens was equipped with wood shaving as litter. All the birds in each pen were allocated to one of four dietary treatments, resulting 4 groups: 1) control group fed with basal diet, 2) FS experimental group fed with basal diet and 15% flax seeds, 3) FS+E experimental group fed with basal diet supplemented with 200 mg α-tocopheryl acetate and 15% flax seeds, 4) FS+E+Se experimental group fed with basal diet supplemented with 200 mg α-tocopheryl acetate and 0,3 mg sodium selenite and 15% flax seeds. The nutrient composition of the experimental diets is given in Table 1. Performance data were taken weekly.

At 42 days, all birds were finally weighed and slaughtered to determine carcass characteristics. All birds were fasted for 12 h prior to slaughter. The weight of the plucked birds was recorded and the abdominal fat pads removed. Carcass yield (without feathers, head, feet and digestive organs) was expressed as a percentage of the live body weight just before slaughter, and carcass components (breast, thigh and wing) were expressed as percentage of carcass weight.

Fatty acid analyses

Fatty acid composition of muscle was determined by gas chromatography. Lipid extraction was performed according to Folch, on 1 g of muscle tissue. Subsequently, the fatty acid methyl esters were analyzed using a SHIMADZU GC-17A gas chromatograph with a 100 m capillary column.

Vitamin E level

α-tocopherol extraction was determined according to Schuep and Rettenmeier in 1994 and high performance liquid chromatographic (HPLC) conditions were performed with a Shimadzu VP Series for the α-tocopherol analysis. Tocopherols were separated on a Alltima RP C-18 column (250 x 4.6 mm i.d., 5 μm particle size) and the column temperature was 25^0

C. The mobile phase was a mixture of acetonitril : methanol (50 : 50 , v / v) with flow rate set at 1.0 mL /min .

Oxidative stability of poultry meat

Lipid peroxidation was estimated based on TBARS as described by Tarladgis et al. (1960) after 120 days of storage at –20 °C. Samples were evaluated for malondialdehyde (MDA) production using a spectrophotometric assay (Jasco V-530). TBARS value was calculated by multiplication of sample absorbance with 7.843.

Statistical Analysis

The differences between treatments were determined by ANOVA for significant differences (P< 0.05)

RESULTS AND DISCUSSION

Productive performance

The results of dietary treatment with different antioxidant source regarding the performance and productivity parameters are presented in table 1, respectively 2. In the present study, the antioxidant supplementation to diets enriched with n-3 PUFAs improved (P <0.05) BW and BWG of the broilers at 42 days. The same significant improvement (P ≤ 0,05) was observed relative to the feeds supplemented with flax seeds. This is due to dietary supplementation with polyunsaturated fats base from both of the flaxseed and vitamin E. Similar results have been reported by Lopez-Ferrer et al. in 2001, using 4% fish oil supplement as polyunsaturated fatty acids. No significant improvement resulted from the supplementation of vitamin E and Se at 0.30 mg/kg compared with the vitamin E alone treatment.

Tab. 1

Performance parameters of broilers[1,2] (21-42 day)

Variables	Control	FS	FS+E	FS+E+Se	*P*
Initial body weight	957	1034	1013	978	-
Final body weight	1833.33	1971.42	2071.42	2103	*
Body weight gain	876.33	937.42	1058.42	1125	*

[1] Values are expressed as means of 5 broilers/group
[2] Values are expressed in g/broiler
* P ≤ 0.05

Tab. 2

Carcass yield parameters of broilers[1]

Variables	Carcass[2]		Abdominal fat		Thighs		Breasts		Wings	
	g	%[a]	g	%[b]	g	%[b]	g	%[b]	g	%[b]
Control	1550	84.54	23	1.48	412	26.58	474	30.58	169	10.90
FS	1575	79.89	24,5	1.55	436.75	27.73	535	33.96	171	10.85
FS+E	1520	73.37	45*	2.96	429.25	28.24	498.25	32.77	153.75	10.11
FS+E+Se	1545	73.46	41*	2.65	423.71	27.42	489.98	31.71	161.23	10.43

[1] Values are expressed as means of 5 broilers/group
[2] Carcass yield without feathers, head, feet and digestive organs
[a] percentage of the live body weight
[b] percentage of carcass weight
* P < 0.05

Few studies have investigated the effects of dietary antioxidant supplementation on cut-up yields of broilers. Although the average carcass weight of FS+E group is the lowest, abdominal fat weight exceeds the parameter values of other groups. In the studies related to Se supplementation, it is noted that form of dietary supplementation also appears to affect cut-up yields of broilers. For example, Edens (2001) noted that the percentage of carcass weight increased when organic Se was added to the diet. However, *pectoralis major* yield decreased as a result of organic Se supplementation in the same study. In this study, the supplementation of Se to n-3 PUFA-enriched diets did not affect ($P > 0.05$) meat yield (table 2).

Fatty acids profile

Fatty acids composition of poultry breast and thigh is presented in table 3. The omega-3 fatty acid ALA is higher in muscle of flaxseed treated birds. The ratio between LA and ALA (18:2 n-6 and 18:3 n-3) is significantly much lower following the FS treatment, giving a more favorable omega-3 status in the FS-broilers. The lipid composition of broiler meat is influenced by fatty acids present in their diet. As the diet becomes richer in PUFA, there is an increase in the PUFA/saturated fatty acid balance in the carcass (Grau *et al.*, 2001).

Dietary supplementation with vitamin E may have reduced fatty acid oxidation. For instance, PUFAs were higher (2.49%, 7.78% respectively) when vitamin E was supplemented to the broiler diets. Similarly, increases in LNA (5.25%, 104.61% respectively) and LA (16.07% for breast) were found in meat from birds fed vitamin E. Total n-3 and total n-6 deposition in breast muscle was higher in birds fed vitamin E, thus it may prevent oxidative loss of n-3 PUFA in poultry meat.

Tab. 3

Fatty acids composition of poultry meat *

Fatty acid	Thigh meat				Brest meat			
	Control	FS	FS+E	FS+E+Se	Control	FS	FS+E	FS+E+Se
Palmitic	20.1	18.49	19.16	19.12	20.46	20.41	18.42	18.27
Palmitoleic	2.92	2.63	1.72	1.77	0.96	0.69	1.82	1.74
Heptadecanoic	n.d.	n.d.	0.17	0.15	n.d.	0.87	0.57	0.49
Cis-10-Heptadecanoic	n.d.	n.d.	0.14	0.17	n.d.	1.26	0.43	0.51
Stearic	9.14	8.38	10.37	10.56	10.72	11.66	10.22	10.13
Oleic	26.12	26.83	24.4	24.13	18.82	15.82	22.27	22.35
Vaccenic (Isomer)	1.87	1.48	1.67	1.59	1.82	2.15	1.73	1.66
Linoleic	23.38	28.28	27.93	28.03	26.43	24.57	28.52	28.33
α-linolenic	1.9	5.33	5.61	5.59	1.38	2.17	4.44	4.58
Cis-11,14-Eicosadienoic	4.43	3.12	4.13	4.17	8.55	8.96	5.52	5.47
SFA	29.24	25.87	29.7	29.83	31.18	32.94	29.21	28.89
MUFA	30.91	30.94	27.93	27.66	21.6	19.92	26.25	26.26
PUFA	29.71	36.73	37.67	37.79	36.36	35.7	38.48	38.38
UFA	60.62	67.67	65.6	65.45	57.96	55.62	64.73	64.64
Omega-3	1.9	5.33	5.61	5.59	1.38	2.17	4.44	4.58
Omega-6	27.81	31.4	32.06	32.2	34.98	33.53	34.04	33.8
Omega-9	26.12	26.83	24.4	24.13	18.82	15.82	22.27	22.35
n-6/n-3	14.63	5.89	5.71	5.76	25.34	15.45	7.66	7.37
SFA/UFA	0.48	0.38	0.45	0.45	0.53	0.59	0.45	0.44

* % methyl esters of fatty acids

Dietary supplementation with selenium also increased levels of most fatty acids, although it cannot be seen any significant differences between FS+E and FS+E+Se treatment. In contrast to vitamin E alone, the addition of dietary selenium did not increase LNA in poultry meat.

α-tocopherol content in poultry meat

The concentration of α-tocopherol in the tissues is presented in table 4. Even though there are no differences regarding the amount of α-tocopheryl among treatments (FS+E and FS+E+Se), there are clear differences in α-tocopherol deposition in the whole body of broilers. The lower concentrations were found in the treatment with no α-tocopherol supplementation, but also in treatment with high inclusion levels of PUFA alone (FS group). The high-fat treatment rich in PUFA predisposes tissues to lipid peroxidation processes, where vitamin E plays a protective role. Several authors have found that feeding diets rich in PUFA results in lower α-tocopherol concentrations in tissues in poultry (Surai and Sparks, 2000). These results suggest that there is some α-tocopherol destruction between absorption and deposition, or during deposition, due to the high PUFA content of the high-fat diets, which would result in a high PUFA deposition in the body. These results imply an important role of vitamin E in the increase of poultry meat shelf life when highly unsaturated diets are fed to these animals.

Table 4

Effect of dietary α-tocopheryl supplementation on α-tocopherol content in poultry meat

Variable	**α-tocopherol**[a]
Control	1,3
FS	1,6
FS+E	2,4
FS+E+Se	2,5

[a] expressed in mg / kg breast meat

Regarding vitamin E tissue concentration, it is well described the linear increase of α-tocopherol concentration in chicken tissues with the increase in its dietary supplementation (Flachowsky et al., 2002). The type and the amount of dietary fat influence the digestion and absorption of fatty acids. Given that vitamin E is a fat-soluble vitamin, it is believed that dietary fat also influences the intestinal uptake of this vitamin.

Oxidative stability of poultry meat

The TBARS values (expressed as mg MDA/kg) for poultry meat are showed in figure 1. It can be seen that TBARS values increased with an increase in supplementation of diet with PUFA. Lipid oxidation in meat from FS+E and FS+E+Se treatments were 2.17 and 2.34 times lower than those from FS treatment ($P<0.05$). TBARS values directly depend on the PUFA content respectively, amount of antioxidants. The only meat that can be assigned as good quality meat (table 6) is that from poultry which received antioxidant treatment. These results indicate that dietary α-tocopherol and Se supplementation can be an effective way to prolong oxidative stability of poultry meat enriched with n-3 fatty acids.

Oxidative changes in muscle foods are generally quantified by the measurement of secondary degradation products. It is accepted that TBARS numbers correlate well with sensory scores of oxidised and warmed-over flavour in muscle food. TBARS numbers greater than 1 correlate significantly with oxidised scores obtained by trained panellists for meats stored under frozen conditions (Buckley and Morrissey, 1992).

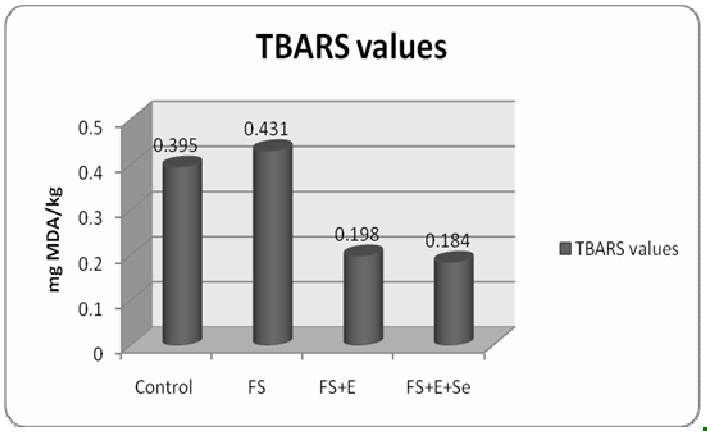

Fig. 1. TBARS values for poultry meat

Tab. 6
Approximate scale for interpretation of TBARS values in meat (Frigg, 1992)

TBARS values	**Interpretation**
≤ 0,2	Good quality
0,2 - 0,5	Limited, tolerable
0,5 – 1,5	Somewhat oxidized
1,5 – 5	Oxidized
≥ 5	Rancid

Sheldon in 1997 observed that meat obtained from animals given a feed with added α-tocopherol contained significantly less volatile oxidation products compared to meat from the control group. This also resulted in a beneficial sensorial characteristic of the meat as expressed by its tenderness and juiciness (De Winne, 1997). The inclusion of tocopherols in the membranes is critical to stabilize meat.

CONCLUSIONS

- Supplementation of vitamin E at 200 g/kg and Se at level of 0.30 mg/kg to broiler diets enriched with n-3 PUFAs significantly increased body weight gain, but did not affect carcass characteristics, with an exception of abdominal fat weight.
- The ratio of n-6 to n-3 fatty acids decreased with increased vitamin E alone or in addition with Se supplementation.
- Both α-tocopherol and oxidative stability of breast and thigh muscles were significantly increased by vitamin E supplementation in diet.
- A high level of dietary polyunsaturated fatty acids negatively influences α-tocopherol content in poultry meat.
- Antioxidative effects of vitamin E and Se were evident in diets enriched in n-3 PUFAs, by reducing TBARS value but no additional benefit was observed in feeding Se supplement in combination with the vitamin E supplement.

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