

## **The effect of improper feeding on the lipid peroxidation of meat animals**

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*Summary:* Pigs of 50 kg bodyweight were fed without premix for two months before slaughtering. Their loin samples showed a lower degree of lipid peroxidation (indicated by a higher antioxidative capacity) than other pig groups which had consumed various premix levels.

Malonaldehyde concentration increased in the blood sera and livers of 7-week-old chickens treated with a high dose of an ionophoric coccidiostatic agent, Salinomycin. Premix deprivation resulted only in a trend to decrease lipid peroxidation.

The possible effects of lipid peroxidation and its degradation products on the physico-chemical characteristics of meat and human health is discussed. The results urge that improper feeding practices should be avoided and optimal premix applications should be found which correspond to the actual requirements.

*Zusammenfassung:* Schweine von 50 kg Körpergewicht wurden vor dem Abschlachten 2 Monate lang ohne Premix (Vitaminzusatz zum Grundfutter) gefüttert. Die Lendenstücke der Schweine zeigten erhöhte antioxidative Fähigkeit, d. h. niedrige Peroxidationsgrade, im Gegensatz zu anderen Schweinegruppen, denen verschiedene Mengen an Premix gefüttert wurden.

Der Gehalt an Malonaldehyd in Blutserum und Leber von 7wöchigen Hühnern, denen hohe Dosen an Salinomycin verabreicht wurden, ist gestiegen. Als Resultat von Premix-Entzug zeigte sich eine abnehmende Tendenz in Lipidperoxidation.

Es werden die möglichen Einflüsse der Lipidperoxidation und der Zersetzungsprodukte auf die physikalisch-chemischen Eigenschaften von Fleisch und auf die menschliche Gesundheit diskutiert. Diese Ergebnisse drängen uns, Möglichkeiten einer optimalen Ernährung mit optimaler Premix-Versorgung zu finden, die dem aktuellen Bedarf entspricht.

*Key words:* premix, lipid peroxidation, coccidiostatic agent, pigs and chickens, antioxidative capacity

### **Introduction**

In recent years, detrimental effects – due mainly to improper feeding and animal keeping practices – have been observed in the physico-chemical characteristics and composition of slaughtered meat, e.g. the formation of DFD (dark-firm-dry) and PSE (pale-soft-exsudative) meat. Among the phenomena the yellowing of bacon has caused many problems (5). Of the detrimental effects we considered the following factors:

1. During the keeping of meat animals the requirement of vitamins, essential macro- and micro-elements is met by a premix form, which means the addition of an adequate mixture of pure vitamins and metal salts to

the feed. We suppose that the premix requirement depends on the period of growth, however, in practice the amount of the added premix does not change during the life of the animals.

2. In the circumstances in which they are kept some infectious diseases may develop in meat animals and in many cases very high doses of medicines are necessary to overcome the illness. This veterinary treatment is often coupled with toxic side-effects.

In particular, the yellowing of bacon is associated with the oxidative decomposition of fats in the living tissues. This degradation means not only an organoleptic impairment but its characteristic process, lipid peroxidation, may form some compounds harmful to human health (9).

Our aim was to investigate the effect of certain dietary factors including premix concentration, and an ionophoric coccidiostatic agent, Salinomycin on some characteristics of lipid peroxidation in pigs and chickens.

### Animal feeding conditions and methods

Pigs of the race Hungarhyb were kept on livestock farms and received the same feed until they attained 50 kg bodyweight. Then the pigs were divided into three groups: the first (group P) received no premix in the growing and fattening period; group G received a premix containing only essential macro- and micro-elements and group C was fed according to the routine animal keeping practice, i.e. with a premix containing trace elements and vitamins. The amount of premix was 2 % of the foodstuff in all cases. The composition of the two premixes is shown in Table 1.

After feeding the animals for 2 months, the pigs were slaughtered and samples were taken from the loins (musculus psoas minor).

Table 1. The composition of 1000 g premix given to pigs of group C and G.

Group C			Group G		
Retinol	IU	1,000,000	Zinc	mg	2,800
Calcipherol	IU	100,000	Iodine	mg	50
Tocopherol	IU	1,250	Selenium	mg	3
Phyllochinon	mg	165	Manganese	mg	1,200
Thiamine	mg	60	Copper	mg	500
Riboflavine	mg	220	Iron	mg	1,950
Pyridoxine	mg	158	CaCO <sub>3</sub>	g	350
Cyanocobalamine	mg	1.8	NaCl	g	200
Niacin	mg	600			
Pantothenic acid	mg	500			
Butylated-hydroxytoluene	mg	400			
Zinc	mg	9,600			
Iodine	mg	198			
Selenium	mg	12			
Manganese	mg	4,800			
Copper	mg	1,980			
Iron	mg	7,800			
Phosphorus	g	130			
Calcium	g	170			

Healthy broiler hybrid chickens were also kept on livestock farms and received the routine feed supplemented with premix. At the age of 3 weeks the chickens were divided into two groups, one of which was chosen to be treated with Salinomycin (120 mg/kg fodder) for a week. Then part of both groups of 4-week-old chickens were killed and blood sera, chest muscle and liver samples were used for chemical analysis.

The experiment was continued and the Salinomycin treatment went on using the same dose till the age of 7 weeks. Another group of the same age was fed with standard chicken premix added at a level of 0.5 % to the foodstuff. The third group received premix-free foodstuff. The last group also received premix-free fodder but it was supplemented with 300 mg  $\alpha$ -tocopherol/kg feed. On the 7th week all the chickens were killed and their various organs were used for chemical analysis.

The determination of antioxidative capacity indicates the levels of fat-soluble antioxidants and the activity of peroxide-metabolising enzymes. Malonaldehyde can be formed from the lipid-peroxides. As it is considered mutagenic and tumourigenic (9), we decided to determine its level in the tissues in some cases.

Determination of antioxidative capacity (3) was based on measuring the oxygen absorption in a water-sunflower oil emulsion system.

The antioxidant capacity (A.C.) is calculated from the equation

$$AC = \frac{T_a - T_c}{T_c}$$

where  $T_a$  is time elapsed for 50 % reduction of the available gaseous oxygen in the sample containing the antioxidative material, and  $T_c$  is the corresponding time interval in the control, without sample.

Table 2. Antioxidative capacity of loin samples of pigs.

	n	Slaughter 15. 9. 1983	n	Slaughter 10. 10. 1983
Premix-deprived group (P)	5	5.52 $\pm$ 3.52	7	9.81 $\pm$ 3.71*
Group fed with microelement premix (G)	5	3.27 $\pm$ 0.95	6	4.95 $\pm$ 1.77
Group fed with vitamin- and microelement-premix (C)	6	1.73 $\pm$ 0.78*	6	5.02 $\pm$ 1.27

\* =  $p < 5\%$

Table 3. Antioxidative capacity of the organs of 4-week-old chickens.

	Serum n = 7	Chest muscle n = 6	Liver n = 6
Control group	2.12 $\pm$ 1.56	2.31 $\pm$ 1.36	1.31 $\pm$ 0.12
Group fed with Salinomycin	0.92 $\pm$ 0.85	0.44 $\pm$ 0.19	2.53 $\pm$ 0.71

Table 4. Antioxidative capacity (AC) and malonaldehyde (MA) content of the organs of 7-week-old chickens.

	Serum n = 5		Liver n = 5	
	AC	MA nM/l	AC	MA nM/l
Group fed with Salinomycin	2.67 ± 1.54	34.5 ± 10.1	8.04 ± 7.1	43.3 ± 2.0
Group fed with 0.5 % premix	1.69 ± 0.65	19.4 ± 2.6**	6.47 ± 1.12	30.9 ± 1.3***
Group without premix	2.28 ± 0.95	13.4 ± 3.6***	11.0 ± 7.4	37.0 ± 2.7*
Group without premix, supplemented 300 mg/kg tocopherol	2.86 ± 1.4	14.2 ± 2.9***	5.65 ± 1.8	33.8 ± 5.1**

\* =  $p < 1\%$ \*\* =  $p < 0.2\%$ \*\*\* =  $p < 0.1\%$

Malonaldehyde assay was carried out with the thiobarbituric acid colour reaction after  $\text{Fe}^{\text{II}}$ -ascorbic acid initiation, in n-butyl alcohol, at 532 nm, according to Ohkawa et al. (7).

## Results

Table 2 shows the antioxidative capacities of loin samples taken from the pigs slaughtered at two different times. Premix-deprived group has the highest antioxidative capacity while the others fed with different premix composition give lower levels. Only at the first slaughter was a significant difference found between the groups fed with two types of premix.

Table 3 contains the data of the first investigations on chickens. In the group fed with Salinomycin, the antioxidative capacity of serum and muscle samples is lower than in the other group. For liver samples the tendency is reversed. Due to the low number of samples it was not possible to show significant differences.

The data for the organs of 7-week-old chickens are given in Table 4. Antioxidative capacities from different groups do not deviate from each other apart from in the group without premix, where the high values are consistent with results obtained with pigs. Malonaldehyde values are the highest both in sera and livers of the group fed with Salinomycin, and the liver data differed significantly from those of other groups.

## Discussion

From the method of measuring antioxidative capacity it is clear that this method evaluates only those fat-soluble materials which can defend sunflower oil from oxidation. This group of substances primarily includes the tocopherols. An increased antioxidative capacity could mean not only a high tocopherol level but also normal activities of enzymes responsible for eliminating lipid peroxides, hydrogen peroxide and some free radicals. Superoxide dismutase (6), glutathion peroxidase (12), catalase and glutathion-S-transferase belong to the enzyme group defending the living organism from free radicals and lipid peroxidation. On the other hand, low antioxidative capacity could mean the domination of lipid peroxides over the defending mechanism.

Our investigations on pigs pointed out that the premix consumed by the animals favoured lipid peroxidation and similar processes in the last period of keeping. The premix present in the foodstuffs showed only similar trends in chickens which may be caused by the low number of experimental animals.

The effect of premix can be explained by the catalyzing activity of some macro- and microelements on lipid peroxidation (2). From Table 2 it can be seen that the premix containing an antioxidant, butylated hydroxytoluene (BHT) increased this detrimental process, probably by maintaining the  $\text{Fe}^{\text{II}}$  state of surplus consumed iron. We used the catalyzing effect of  $\text{Fe}^{\text{II}}$  for initiating lipid peroxidation for analytical purposes in the present investigations.

The enhancing effect of the high dose administered coccidiostatic agent, Salinomycin on lipid peroxidation and similar processes was demonstrated in our experiments. A rather toxic dose of this drug can destroy the protecting mechanism of the organism by many biochemical pathways. Some drugs, e.g. phenobarbital (4) and paracetamol (10) are substrates of Cytochrom-P-450, so that they increase lipid peroxidation by competitive inhibition. We have no data about Salinomycin in this respect, however the possibility of this mechanism cannot be excluded.

The lipid peroxides and some of their degradation products cause extensive damage to biological membranes, producing a decrease in electrical resistance and membrane fluidity and eventual loss of membrane integrity (8). Thus an advanced degree of lipid peroxidation in the animals might contribute to the unfavourable physico-chemical characteristics of pork and beef (exsudation etc.). The direct relationship between lipid peroxidation and the yellowing of bacon has been verified (5).

The presence of the degradation products of lipid peroxidation in meat consumed must also be considered from a food hygiene point of view. According to data from 37 countries, the occurrence of breast and colon cancer showed a strong correlation with fat intake (9). However, a causal relationship cannot be demonstrated in this case, although the role of lipid peroxidation and its degradation product (malonaldehyde) on enhancing tumourigenic processes in animals was proved (1).

These arguments underline the necessity of avoiding improper feeding practice in the keeping of animals for meat. Further investigations are necessary to select components from a premix which are responsible for the detrimental effects and to optimize premix composition to every period of animal life corresponding to the requirements.

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