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## CORRECTION OF OXIDATIVE METABOLISM OF ANIMALS BY MEANS OF BIOLOGICALLY ACTIVE COMPLEX OF ANTIOXIDANT DEFENCE

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### ABSTRACT

Nowadays one of the most burning issues in manufacturing of high-quality hog products is reduction of biological value of meat. The reasons for that are low quality of feed stuff, huddling, limited activity of animals, and contamination of the feed stuff with xenobiotics of technogenic nature. All the factors result in development of animals' stress that firstly triggers activation of free-radical process. Thus, the research on correction of oxidative stress of animals is considered to be essential. To eliminate negative effect of lipid peroxidation among fattening pigs, biologically active complex of antioxidant defence, including vitamin-mineral, oily and plant components, was used. Content of malondialdehyde in blood serum and content of amino acids in longissimus was determined.

The use of biologically active complex of antioxidant defence in diet of the experimental pigs made it possible to reduce negative influence of LP metabolites on organism. Besides, in the experimental group the number of animals with the meat parameters of biological value close to the expected level according to genetic potential, increased.

**Disciplinary:** Animal Sciences, Biosciences/Biotechnology.

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## 1. INTRODUCTION

Under the conditions of large industrial livestock breeding complexes, the influence of various stress factors on the organism of agricultural animals has a synergetic effect. According to a number of authors, low quality of hog products, as well as veterinary problems in agricultural enterprises in this sphere of livestock breeding, are mostly caused by stress factors [3, 7].

One of the basic components of organism adaptation to living conditions is oxidative metabolism

and its supporting homeostatic mechanisms. The improper physiological balance between intensity and speed of free-radical reactions on one side and activity of antioxidant systems on the other side leads to metabolic changes in the organism, i.e. to “oxidative stress” [5]. Hypersynthesis of various bioradicals, such as reactive oxygen species (ROS) and reactive nitrogen species (RNS) is accompanied by quick depletion of reserves of the antioxidant system [6]. On the molecular-cellular level, it is characterized by activation and intensity of the LP process that results in both changes in properties of biomembranes and in the complete cellular dysfunction [4]. Transition LP products are able to induce oxidative transformation of nucleic acids, and protein and lipidic macromolecules. Under the conditions of oxidative stress, the speed of reparation of those macromolecules is much less than the speed of the formation of bioradicals [4, 5]. Oxidative modifications mostly affect amino acids included in the composition of proteins, such as cysteine, methionine, histidine, arginine, tryptophan, tyrosine, and phenylalanine [3, 6].

According to both Russian and foreign authors, the influence of stress factors on porcine organisms firstly results in a reduction of the expected genetic level of production [1, 2]. Issues of correction and regulation of processes of oxidative metabolism in porcine organisms are still considered to be essential in veterinary theory and practice.

This research concentrated on the influence of alimentary biologically active complex of antioxidant defense on the amino acid composition of porcine muscles under the conditions of oxidative stress.

## 2. MATERIALS AND METHODS

Dynamics of the MDA level in blood serum of fattening pigs was studied under the conditions of research and production experiment. Blood of animals was taken from ear's marginal vein on the 1<sup>st</sup>, 30<sup>th</sup> and 60 days of research. Quantification of MDA in biological material, such as blood serum, and samples of muscles, heart, liver, kidneys, and lungs was done according to standard methods. Amino acid profile was identified in homogenate consisting of equal weights of longissimus, and shoulder, round and iliac muscles, according to 17 parameters. Analysis of amino acid composition was done on a liquid chromatograph LC-20 Prominence (Shimadzu production) with spectrophotometric detector (254 nm); reverse stationary phase column C18 (Analysentechnik 250 x 4.6 mm, 5 µm, Germany) and proper precolumn; mobile phase – mixture of sodium acetate liquor (0.06 mmol/dm<sup>3</sup>, pH≤5.5 and 4.05 and 1% liquor 2-propanol in acetonitrile).

Possibility of correction of oxidative stress was evaluated by means of implementation of a biologically active complex of antioxidant defense (BACAD), consisting of vitamin-mineral, oily and plant components, into the porcine organism. The complex included soy isolate, pyridoxine, niacin, vitamin C, magnesium citrate, a complex of calcium with methionine hydroxy analog (MHA), lysine, threonine, valine, tryptophan, flaxseed, olive and jinjili oils (gingili/gingelly oils), and leaves of *Urtica dioica*.

Two groups of landrace breed of pigs were formed on the principle of analogs (n=40). The animals from the control (C) and experimental (E) groups were kept under the usual conditions of the pig-breeding farms. The pigs from the experimental group were given BACAD for 14 days.

The authenticity of the results was proved using statistic techniques with averaging and standard deviation calculation using Student's t-test. The results were considered to be verified by p<0.05.

Processing of the obtained data was done using Microsoft Excel®.

The research on the dynamics of the MDA level in blood serum of pigs has shown that only 8.75% of animals had background concentration within physiological standards that was  $0.853 \pm 0.015$  mcIU/l. In 15% of cases MDA content increased by 59.5% ( $1.276 \pm 0.023$  mcIU/l). Most of the pigs under research (76.25%) had MDA content on the level of  $1.045 \pm 0.019$  mcIU/l that on average was higher than physiological standards by 30.6% ( $p \leq 0.05$ ). For the whole period of research and production experiment in the Control group of pigs MDA level increased by 7.2-8.5% as compared with background parameters. The obtained data proved that the pigs under the conditions of industrial breeding technologies suffer from depression of system of antioxidant defence. Organisms of animals are subjected to constant intensive influence of toxic products of lipid peroxidation. In the Experimental group of pigs having the diet with added BACAD, there was positive dynamics in reduction of MDA level ( $p \leq 0.05$ ). By the 30<sup>th</sup> day of research, MDA content in blood serum was  $0.967 \pm 0.020$  mcIU/l. by the 60<sup>th</sup> day of research  $0.834 \pm 0.022$  mcIU/l. that was not different from physiological standards of that kind of animals.

**Table 1:** MDA concentration in samples of tissue and organs of the experimental pigs, mcIU/l.

№	Sample of the researched biomaterial	MDA concentration. mcIU/l	
		The Control group	The Experimental group
1.	Blood serum	$1.386 \pm 0.036$	$0.834 \pm 0.022$
2.	Muscle tissue	$1.296 \pm 0.027$	$1.195 \pm 0.031$
3.	Heart	$1.432 \pm 0.024$	$1.367 \pm 0.026$
4.	Liver	$2.180 \pm 0.032$	$2.053 \pm 0.035$
5.	Kidney	$1.955 \pm 0.026$	$1.986 \pm 0.028$
6.	Lung	$1.733 \pm 0.022$	$1.420 \pm 0.032$

**Table 2:** Amino acid composition of porcine muscle tissue under the conditions of oxidative stress, (mg/100g).

№	Parameters	Performance standard [2]	Parameter among the pigs from Control group (n=20)	Parameter among the pigs from Experimental group (n=20)
<b>Irreplaceable amino acids</b>		<b>7801</b>	<b><math>5840.2 \pm 51.7</math></b>	<b><math>7736.5 \pm 35.4</math></b>
Including:				
1	Valine	1135	$668.2 \pm 22.3$	$917.7 \pm 14.5$
2	Isoleucine	970	$763.3 \pm 12.9$	$1226.5 \pm 17.3$
3	Leucine	1538	$845.3 \pm 18.4$	$1194.8 \pm 20.4$
4	Lysine	1631	$1526.4 \pm 92.9$	$1857.2 \pm 25.1$
5	Methionine	478	$515.5 \pm 15.4$	$402.8 \pm 11.7$
6	Threonine	961	$818.2 \pm 20.3$	$1251.5 \pm 19.9$
7	Tryptophan	274	$138.6 \pm 16.9$	$156.8 \pm 13.8$
8	Phenylalanine	814	$565.1 \pm 15.2$	$728.7 \pm 14.4$
<b>Non-essential amino acids</b>		<b>10832</b>	<b><math>9569.6 \pm 94.8</math></b>	<b><math>10954.2 \pm 69.4</math></b>
Including:				
9	Alanine	1213	$1219.3 \pm 62.4$	$1194.3 \pm 57.8$
10	Arginine	1223	$1020.1 \pm 63.7$	$1372.5 \pm 35.3$
11	Asparagine acid	1895	$1801.8 \pm 70.5$	$2197.5 \pm 52.8$
12	Histidine	773	$705.6 \pm 19.4$	$764.7 \pm 14.1$
13	Glycine	864	$698.0 \pm 14.7$	$792.6 \pm 18.3$
14	Glutamine acid	3385	$2899.7 \pm 113.5$	$3136.8 \pm 103.7$
15	Oxyproline	50	$48.6 \pm 5.4$	$38.4 \pm 4.8$
16	Serine	743	$641.5 \pm 17.6$	$822.5 \pm 10.6$
17	Tyrosine	695	$535.0 \pm 14.8$	$710.3 \pm 11.2$
<b>Total number of amino acids</b>		<b>18633</b>	<b><math>15409.8 \pm 101.6</math></b>	<b><math>18690.2 \pm 62.2</math></b>

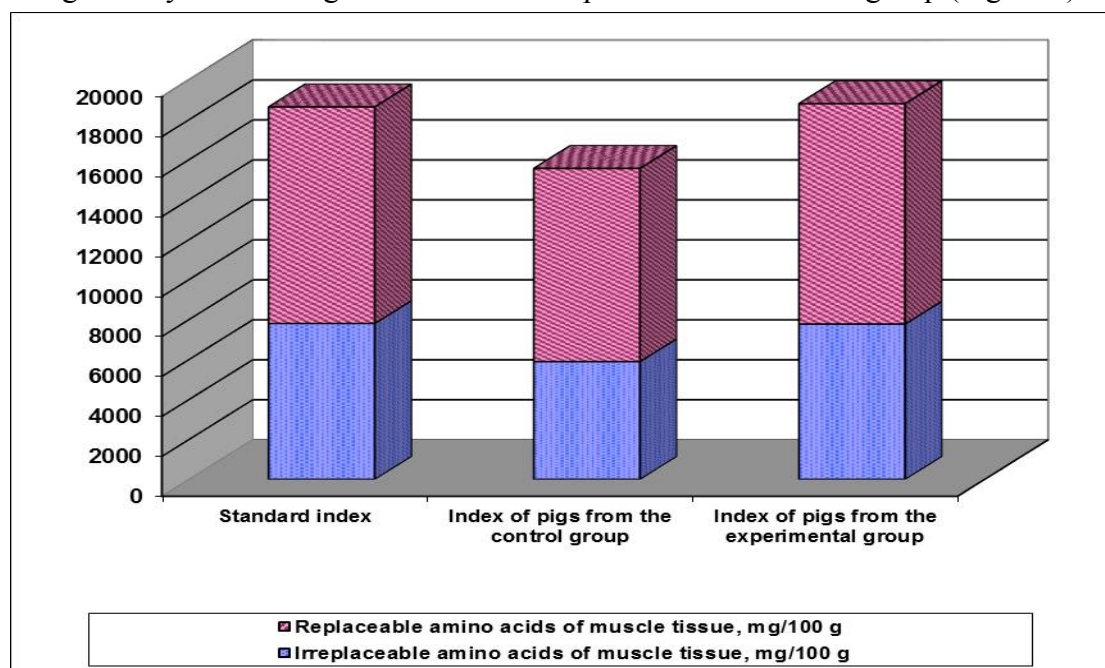
The research on MDA level in samples of porcine tissue and organs have shown that in homogenate of liver of animals from both the Control and Experimental group, MDA concentration was the highest -  $2.180 \pm 0.032$  mcIU/l and  $2.053 \pm 0.035$  mcIU/l, respectively (Table 1).

According to the results shown in Table 1, distribution of toxic LP products in other tissues under research is equal enough. Difference in MDA concentration in homogenate of muscle tissue between the Control group and Experimental group was 7.8%.

The results of defining amino acid composition of muscle tissue of the experimental animals are shown in Table 2.

Comparative analysis of the obtained data has stated that total number of amino acids in samples of muscle tissue of the pigs from the Control group was 17.3% lower than the performance standards for muscle tissue of this kind of animals [2]. Total number of amino acids in samples of muscle tissue of the pigs from the Experimental group was within performance standards.

In the amino acid composition in samples of muscle tissue of the pigs from the Control group the content of tryptophan was 49.4% lower; the content of leucine - 45% lower; the content of valine - 41.1% lower; the content of phenylalanine - 30.6% lower ( $p < 0.05$ ). In muscle tissue from the Experimental group shortage of tryptophan stayed stable (42.8%). However, it was not so prominent (22.4%). the content of valine was 19.2%. and the content of phenylalanine was 10.5% ( $p \leq 0.05$ ). It is worth to note that content of isoleucine, lysine and threonine in samples from the Experimental group was generally 24-25% higher than in the samples from the Control group (Figure 1).

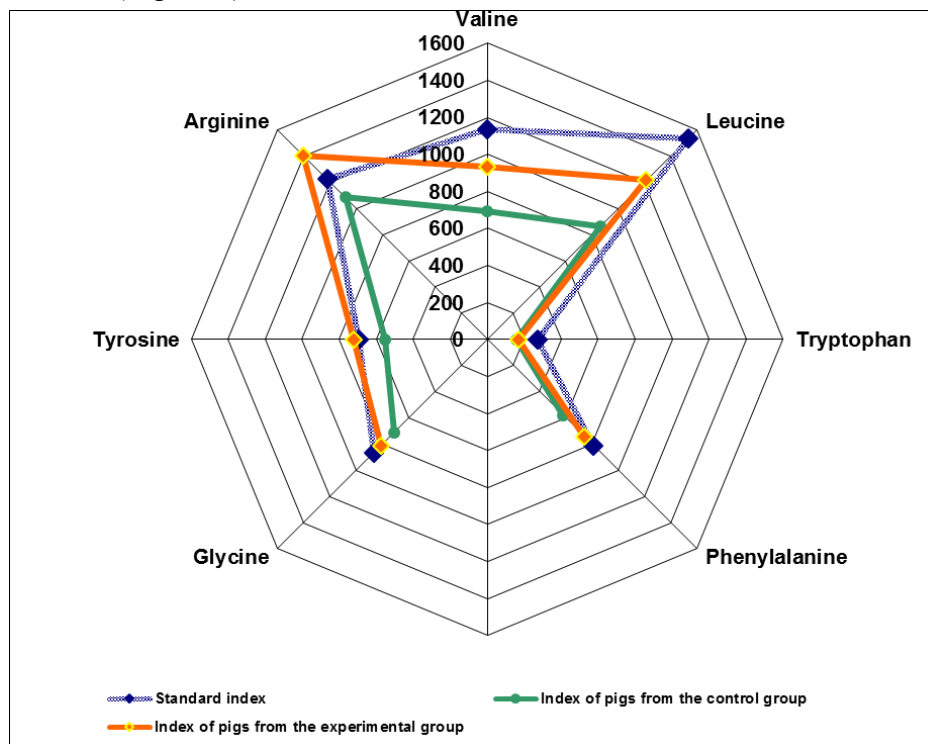


**Figure 1:** Indexes of amino acid composition of muscle tissue of Landrace breed of pigs.

Shortage of non-essential amino acids – arginine, glycine, glutamine acid, serine and tyrosine – in the samples from the Control group was from 12.6 to 23%, as compared to both performance standards and data obtained in the Experimental group. No prominent differences between the data obtained from the Experimental group and performance standards regarding content of non-essential amino acids in porcine muscle tissue have not been found ( $p \geq 0.05$ ). In all the samples of porcine muscle tissue under research disbalance in proportion tryptophan/ oxyproline (index of protein value) was identified. It was most prominent in the Control group  $2.85 \pm 0.16$ , performance standard 5.2-5.8 [2]. In the Experimental group index of protein value was  $4.13 \pm 0.09$ .

In samples of muscle tissue of the pigs from the Control group changes in amino acid composition were identified in 82.5% of cases. and in 17.5% of cases the data were within low limit of performance standards. Shortage of amino acids in muscle tissue of the pigs from the Experimental group was identified in 27.5% of cases. and in 72.5% cases the data met performance standards.

Thus, the research done have shown that under the conditions of industrial breeding technologies in 85-90 % of cases pigs suffer from oxidative stress. Local signs of oxidative stress in porcine organism were identified by increased MDA content in tissues of liver, kidneys and lungs. Concentration of that LP metabolite in organs were 1.5-2 times higher than its concentration in blood serum that may be indirect implication of oxidative damage of cell structure and disorder in process of reparation of protein macromolecules [9], as well as of trigger action of oxidative stress on whole organisms of animals (Figure 2).



**Figure 2:** Quantative content of amino acids: targets of oxidative modifications in muscle issue of pigs.

Under the conditions of research and production experiment it was shown that the targets of oxidative modifications in porcine organism were mostly irreplaceable amino acids, such as valine, leucine, tryptophan and phenylalanine. Content of those amino acids in samples of muscle tissue was 30.6–49.4% lower. AS is commonly known, oxidative modification of irreplaceable amino acids results in changes in not only primary, but also both secondary and tertiary structure of protein. Changes in protein structure create conditions for their agglomeration that in a number of cases causes autostimulation of free-radical processes in organism [4, 5]. In the range of non-essential amino acids the most prominent quantity changes were identified regarding arginine, glycine and tyrosine. In samples of porcine muscle tissue their content was 17-23% lower. Like irreplaceable amino acids, during oxidative modification arginine and glycine cause transformation of protein macromolecules [4, 5, 6]. Modification of tyrosine leads to inhibition and blocking cell signals [5, 7]. Identified changes show that there are high chances of that under the conditions of industrial breeding technologies pigs suffer from depression of antioxidant defence of organism.

### 3. CONCLUSION

The use of BACAD in feeding diets of experimental pigs allowed to reduce the negative effect of metabolites on the organism of experimental pigs [8, 10, 11]. MDA content in the blood serum of animals reduced up to the level of physiological standard. In the tissue of the liver, kidneys, and lungs' positive dynamics were less prominent, and the MDA level was 6-8% lower than in samples of tissue of the animals from the Control group. The other result of equal importance is the standardization of most quantity indexes of the amino acid composition of muscle tissue. Content of irreplaceable amino acids – valine, leucine, and phenylalanine – was 10-20% lower than the reference value. However, as compared with the data obtained from the animals from the Control group, obtained results can be considered to tend to normalization of protein synthesis in organisms and reduction of the intensity of oxidative modification of amino acids. The positive effect of the use of BACAD was also proved by that the number of animals with the meat having a biological value close to the expected level according to genetic potential, increased by 3.2 times.

### 4. DATA AND MATERIALS AVAILABILITY

Relevant information is available by contacting the corresponding author.

### 5. CONFLICT OF INTEREST

The authors confirm that the given data do not have any conflict of interest.

### 6. ACKNOWLEDGEMENT

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