PE9.29 Influence of Post-Shock Modification of Pork on Amino-Acid Composition of Muscle Tissue and Composition of Extracted Low Molecular Peptides 192.00

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Abstract

Important quantitative changes in amino acid composition of the muscular tissue of pigs neck cut were found after artificial apoplectic shock of the animals. Together with these, changes in qualitative and quantitative composition of low-molecular (molecular mass < 3500 Da) compounds of peptide nature extracted from muscle tissue were observed. One can suppose that the modeled insult initiates in the revived animals biosynthesis de novo of muscle proteins and bioactive low-molecular peptides with large spectrum of neuroprotective, modulating and effectors action.

Key words: pork, apoplectic shock, muscle tissue of neck cut, amino acid analysis, peptides, electrophoresis

I. INTRODUCTION

Current strategy for the development of meat products with functional curative-preventive properties is based on the incorporation into initial raw materials or final products of exogenous bioactive components – food fibers, proteases, hydrolysates of dairy or meat proteins, probiotics or prebiotics of microbial origin [1]. Quite different approach to enrichment of meat raw materials with endogenous bioactive peptides, possessing a wide spectrum of protective, modulating or effectors action and based on the use of meat obtained from animals with modeled pathological states is of great interest.

II. MATERIALS AND METHODS

The objective of this work was study of amino acid composition of meat of pigs (neck cut) modified by apoplectic shock, and the electrophoretic analysis of extracted from muscular tissue low molecular peptide compounds with molecular mass < 3500 Da.

Animals. The experiments were carried out on muscle tissue of neck cut of the control and experimental pigs, subjected to apoplectic shock by the method of Makarenko A.E., et al [2].

Amino acid analysis. Amino acid analysis of hydrolysates of muscle tissue was carried out according to standard method on amino acid analyzer PMA GmbH Aracus

Isolation of low molecular peptide fraction. The muscular tissue was homogenized in a solution of 0.01M Tris-HCl + 0.15 M NaCl, pH 7.4, the supernatant was dialysed through dialysis membrane Pierce-Snake Skin 3500 MWCO against distilled water and then lyophilized. Total concentration of peptides was determined by the method of Bradford M.M. [3].

SDS-PAGE. Peptide composition of the control and experimental samples of dializates was analyzed by the method SDS PAGE, as described earlier [4]. Melittin (2600 Da), oxidized glutathione (612 Da), tripeptide Gli-Trp-Gli (350 Da) and reduced glutathione (310 Da) were used as markers of molecular masses.

III. RESULTS AND DISCUSSION

Study of the extracts of muscle tissue has indicated an increase in the solvable fraction of proteins in the experimental group as compared to the control: by 11 and 42% for water and salt-soluble proteins,

respectively, and the content of free amino acids in the experimental group by 1.4 times exceeded that in the control. The results show that in the case of apoplectic shock the total concentration of peptide components in the tissues of the operated animals increases 1.8 times.

Comparison of the data of amino acid composition analysis of the control and experimental samples of muscle tissue shows pronounced quantitative changes in the composition of bound amino acids (Table 1).

Table 1. Amino acid composition (bound amino acids) of the 'shock' and control meat (pork, neck cut)

Amino acids	Control, %	Experiment, %
Aspartic	**1.07**	**1,47**
Threonine	0.58	0.68
Serine	**0.49**	**0.63**
Glutamic	**1.61**	**2.47**
Proline	**0.62**	**0.88**
Glycine	0.60	0.64
Alanine	**0.64**	**0.96**
Valine	**0.62**	**0.78**
Methionine	**0.30**	**0.41**
Isoleucine	0.58	0.69
Leucine	0.94	1.03
Tyrosine	**0.43**	**0.56**
Phenylalanine	0.49	0.54
Histidine	0.49	0.55
Lysine	1.07	1.24
Arginine	**1.29**	**0.85**
Total	**11.82**	**14.38**

1.In bold type are amino acids, which concentrations in the control and experimental samples differ by more than 20%

2. The presented values are averaged from 3 determinations

The peculiar feature of the amino acid composition of experimental samples of muscle tissue is the reduced content (by 34%) of arginine as compared to control samples. One can suppose, that it is connected with metabolic damages in the mechanisms of biosynthesis of arginine-containing proteins. Out of 16 determined protein amino acids 8 are significantly increasing (from 25% to 53%) in the experimental samples. It can be caused by biosynthesis of several proteins de novo in response to metabolic changes in post trauma organism of the animals. This is confirmed first of all by the increase by 42% of proline – a structural component of collagen.

The quantitative determination of peptides in the dializates showed growth of their total concentration in the experimental samples by 55% (36% - control, 56 – experiment).

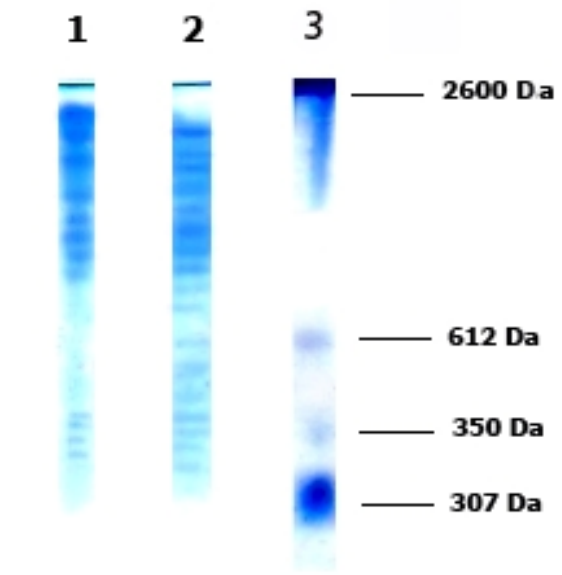
Thus, the modeled apoplectic shock induces an increase in the quantity of low molecular peptides in the muscle tissue through the synthesis de novo or autoproteolysis of muscle proteins.

Analysis of electrophoretic separation of peptide fractions in the dializates of the investigated samples has shown qualitative and quantitative differences in the peptide composition of control and experimental samples.

In all the investigated samples, there is an intensive band in the start with a molecular mass > 2600 Da. A characteristic feature of the presented results of the electrophoretic analysis (Fig.1) is a quantitative increase of peptide bands in the experimental samples as compared to the control. Thus, in the control 13 peptide bands was identified, and in the experiment – 18.

Thus, it has been shown that modelled apoplectic shock initiates in the revived experimental animals formation of new low-molecular peptides with molecular masses from 310 to 2600 Da, which, most

probably, influence the inhibition of neurodegenerative processes and possess neuroprotector effect.



a

Qualitative composition and distribution by molecular masses of peptide fractions of dializates of control and experimental muscle tissue samples

Molecular masses (Da) 2600 - 600	Sample	Molecular masses (Da) < 600
9 bands	1	4 bands
11	2	7

b

Fig.1. Separation of peptide fractions of muscular tissue samples of neck cuts (1- control, 2- experiment); 3- standard mixture of peptides a) electrophoregrams, b) table

For the purpose of comparison, a peptide composition of pharmacological preparations “cerebrolyzine”, “cerebrolyzate” and “cerebral” was analyzed. All these preparations are prepared from brains of agricultural animals (cattle and pigs). As a result it was found that in the muscular tissue of pigs, which had had apoplectic shock, polypeptides, similar to those found in the organ preparations, possessing curative and preventive effect, are formed.

IV. CONCLUSION

The modelled apoplectic shock induces in the muscle tissue of operated pig quantitative changes in amino acid composition of muscle proteins and changes in the qualitative composition of the extracted low molecular peptides.

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