

## **Modification of meat raw materials by manipulation with animals**

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*Abstract* – the paper presents the results of comparative investigation of a peptide composition of pharmacological preparations “Tserebrolizin™”, “Tserebrolizat™” and “Tserebral™”, currently present on the Russian market. The analysis of low molecular protein fractions and polypeptides of the preparations “Tserebrolizin” and “Tserebrolizat” and of the muscular and brain tissue of pigs, which had endured insult, shows that in the case of hemorrhagic insult the total concentration of peptide components in these tissues of the operated animals increases, reaching the values of the preparations being investigated. In neurological tests a similar rehabilitation effect of the meat of post-insult pigs and “Tserebral” was demonstrated. The test results on laboratory animals confirm the anti-insult effect of the meat of convalescent pigs.

***Index Terms:* hemorrhagic insult, meat, electrophoresis, bioactive peptides**

### **I. INTRODUCTION**

Most strategies of animals feeding which are currently successfully used are “passive”, i.e. they are based on enrichment or incorporation of particular feed ingredients or chemical compounds through animals’ gastrointestinal tract, thus influencing through the absorption of these components the chemical, biochemical or technological characteristics of meat raw materials. In recent decades the most urgent challenge for food industry is the development of food products with pre-determined curative-preventive properties. A search of food components preventing widely spread diseases and modulating physiological systems of human beings in response to pathological states is of particular interest.

Di-, tri- and polypeptides, possessing hypotensive, antioxidant and antistress properties were isolated from raw meat (Arihara, 2006). An evident geroprotective effect when introducing synthetic dipeptides of thymus – thymogen and vilon (L-Lys-L-Glu) - in the experiments on mice was revealed; a reduction in frequency of spontaneous tumors was also marked in rats (Miller, 2001).

Study of the prospects for purposeful changes in raw materials properties by direct manipulations with animals is of much current interest.

Earlier (Chernukha et al, 2009) we made an assumption that alongside with brain other tissues of pigs can accumulate (though to less extent) biologically active substances, possessing curative effect with regards to hemorrhagic insult. The meat of such animals can become raw materials for the production of functional food products having a high risk of cerebrovascular diseases and the people who had endured insult.

Preliminary investigations on rats have shown good prospects for use of the meat of animals, who had recovered from insult, and its influence on the acceleration of rehabilitation period.

The brain of pigs, which had recovered after modeling of hemorrhagic insult, serves as the raw material for production of medicinal anti-insult preparation ‘Tserebral’ (Patent №2151605, 2000).

The objective of the present investigation was the development of innovation technologies for production of endogenous bioactive peptides, possessing modulating and effector’s action as a meat product’ component

## II. MATERIALS AND METHODS

“Tserebrolizin” used for curing ischemic and hemorrhagic insult contains a complex of peptides obtained from the brain of intact pigs. The active fraction of “Tserebrolizin” is presented by biologically active neuropeptides, the molecular weight of which is not higher than 10000 Da. The closest analogue of it is a neuroprotector medicine “Tserebral”, which has specific traumatrophic peptides with molecular mass 400 – 1500 Da. “Tserebral” has a positive effect on inhibition of neurodegenerative processes, possesses neuroprotective effect and improves general condition of post-insult organism. An analogue of the “Tserebrolizin” preparation has been developed in Russia under the trade name “Tserebrolizat”. The preparation is a hydrolised proteolytic enzyme from *Aspergillus terricola st. H-20*, cattle brain, and contains free amino acids and peptides.

Low molecular fractions of proteins and polypeptides of neuroprotector preparations, meat and brain of pigs were studied by gel-electrophoresis method. The electrophoresis was carried out in polyacrylamide gel with 16,5% T and 6% C using tris-tricin electrode buffer with pH 8.25, i.e. on gels, having 16.5% cross links.

The meat was obtained from pigs, which had safely endured one-sided autohemorrhagic insult and slaughtered on the 52<sup>nd</sup> day after operation.

A method of Makarenko (Patent № 1767518, 1990) was used to reproduce the hemorrhagic insult in animals. A one-sided acute hemorrhagic insult was modelled on laboratory animals (white nondescript male rats at 3 month age) under ether anesthesia. Manipulations with the animals and their handling in vivarium were carried out according to International rules of humane treatment of animals.

Four groups were formed, 7 animals per each. In groups I, II and III modelling of insult was carried out.

The diets of groups II and III contained pig meat (5 g per animal): group II – raw meat of pigs having had insult; group III – raw meat of intact pigs. The animals of group I and the control IV consumed common vivarium diet. All the diets were balanced over major nutrients. Animals of group I also received Tserebral preparation intranasally. The influence of meat and the preparation on dynamics of disturbances development, induced by HI, and also on the change in mass, was studied during two weeks, with the observation of the state of animals on the 2<sup>nd</sup>, 3<sup>rd</sup>, 5<sup>th</sup>, 6<sup>th</sup>, 10<sup>th</sup>, 13<sup>th</sup> and 15<sup>th</sup> days after operation.

Were evaluated: (1) weight change according to the results of their daily weighing; (2) orientation and exploring behavior and motor activity - in test “open field”; (3) muscular tension - test on a horizontal bar; (4) neurological deficiency - on the scale Stroke-index McGrow in Kulchikov modification (Patent № 2005133748/14, 2005).

## III. RESULTS AND DISCUSSION

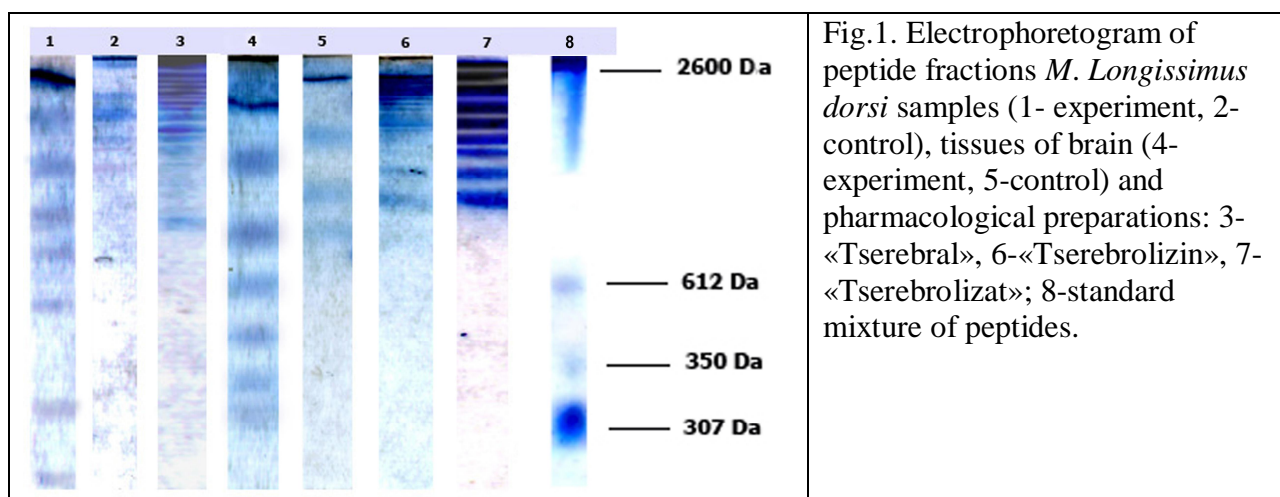
Comparative amino acid composition of the preparations “Tserebral™”, “Tserebrolizat™” and “Tsebrilizin™” is presented in the Table. As is seen from the

Table, the preparations from the brain of cattle by the content of threonine, serine, methionine, lysine and arginine are significantly different from the preparation of pigs' brain.

Table 1. Amino acid composition of the preparations, possessing neurprotector effect

Amino acid	Amino acid content in mg/ml**			
	Limits of detection	Tserebral	Tserebrolizat	Tsebrilizin
Aspartic	2.40 - 3.60	2.67±0.22	2.43±0.41	2.10±0.23
Threonine	0.21 - 0.39	1.65±0.12	1.18±0.27	1.63±0.21
Serine	0.21- 0.39	1.37 ±0.26	0.65±0.25	1.83±0.30
Glutamic	3.20 – 4.80	4.86 ± 0.50	3.78± 0.48	3.54 ±0.52
Proline	1.60 – 2.40	1.25±0.35	1.48± 0.25	1.46 ±0.31
Glycine	1.20 – 1.80	1.35 ± 0.32	1.68 ±0.25	1.19 ±0.23
Alanine	2.40 – 3.60	2.66± 0.56	2.45± 0.61	2.46±0.58
Valine	1.60 – 2.40	2.14±0.65	2.62± 0.68	2.22± 0.63
Methionine	0.35 – 0.65	1.02 ±0.15	0.71±0.09	1.14± 0.17
Isoleucine	1.60 – 2.40	1.72± 0.22	2.11± 0.31	1.78± 0.26
Leucin	4.80 – 7.20	3.75± 0.37	4.10± 0.31	3.89± 0.35
Tyrosine	0.35	0.27± 0.08	0.77 ± 0.09	0.37± 0.08
Phenylalanine	1.60 – 2.40	1.30 ± 0.11	1.45 ± 0.15	1.72 ± 0.20
Histidine	1.04 – 1.56	1.18± 0.06	1.03 ± 0.09	1.00 ± 0.05
Lysine	4.80 – 7.20	1.91± 0.25	1.94± 0.31	1.89± 0.38
Arginine	0.30 – 1.10	1.65 ± 0.27	1.82± 0.32	1.65± 0.31

The analysis of electrophoretic separation (Fig.1) of peptide mixtures has shown qualitative and quantitative differences in the peptide composition of muscular and brain tissue of control and experimental animals.



In the dialysate of brain of the operated animals 3 bands in the interval of molecular masses 2600 – 600 Da and 5 bands with molecular masses < 600 Da are found. In the dialysate of brain of the control animals - 2 peptide bands in the range of molecular masses 2600 – 600 Da and 3 bands with molecular masses < 600 Da are found.

The results show that in the case of hemorrhagic insult the total concentration of peptide components in the tissues of animals on which the operation had been performed, increases. In the muscular tissue of pigs, due to hemorrhagic insult, the polypeptides

similar to those found in the preparations, possessing curative and recovery effect, are being formed.

The results of weighing have shown that the animals which had in their diets the meat of post-insult pigs had a sustained weight gain reaching the weight of intact rats (Fig.2). Fluctuations in the mass on critical 6<sup>th</sup> and 10<sup>th</sup> days were marked.

In rats to which Tserebral has been introduced, the weight gain was less pronounced but stable, the critical days were not evident. The animals of group III, which had received the meat of intact pigs, demonstrated the worst result.

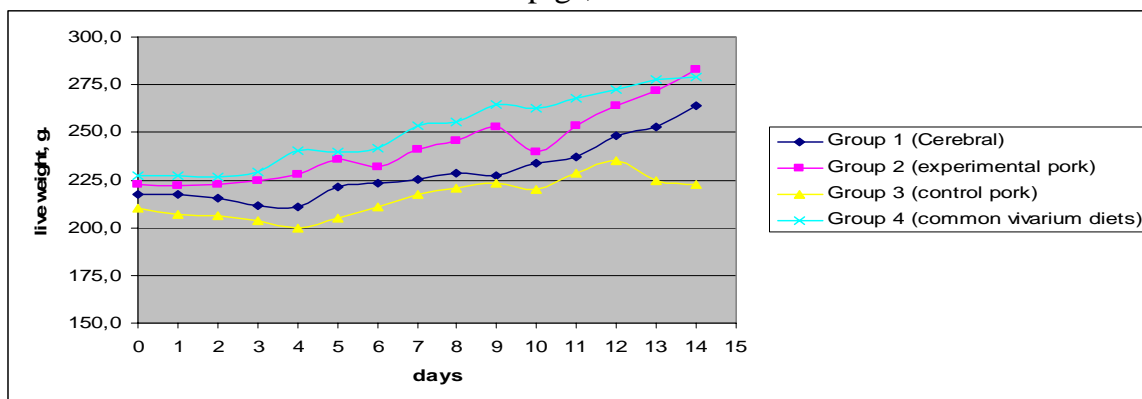


Fig.2. Animal live weight gain

In the analysis of neurological status of the animals on the 2<sup>nd</sup> day of the experiment, the maximum number of animals with light neurological disorders was marked in the 3<sup>rd</sup> group (62%), for the control group this index was 30%.

The animals which consumed meat, recover more quickly, than the animals of the control group, the indices of neurological deficiency on McGrow scale improve.

The incorporation into the rats' diets the meat of pigs in which intracerebral posttraumatic hematoma had been reproduced led to significant decrease of manifestation of post-insult disorders.

The analysis of neurological status of the revived animals has shown that "Tserebral" reduced manifestation of neurological deficiency in operated animals at the early stages of recovery while in the group of animals with HI, having consumed the meat of post-insult pigs this index was surely decreasing during the whole observation period.

The preparation "Tserebral" and the meat of post-insult pigs possess a similar therapeutic effect - they increase the total indices of motor activity and "the effect of the hole".

### III. CONCLUSION

The obtained results confirmed our assumptions about the accumulation of biologically active substances having anti-insult properties in muscular tissue of animals. Testing on laboratory animals indicate that the meat of convalescent pigs and the neuroprotector "Tserebral" possess a similar rehabilitation effect during reproduction of hemorrhagic insult.

These data prove a possibility to obtain meat with pre-determined curative properties due to manipulations with animals. These raw meats can be used for creation of the products for patients with acute disturbance of cerebral circulation (hemorrhagic insult).

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