

INTERNAL ORGANS OF PIGS AS A SOURCE OF BIOLOGICALLY ACTIVE SUBSTANCES

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Abstract – The aim of this study was to investigate the certain physico-chemical characteristics of the individual lyophilized extracts of the pigs' internal organs and complex preparations Kolimak and Dinormin prepared on their basis. When using the above mentioned preparations, the positive effects were observed in the laboratory animals with the model of diarrhea. The paper presents the data of amino acid composition, electrophoretic examination, HPLC, thin layer chromatography of the preparations. Considerable quantity of the low molecular weight components were found in the individual extracts and complex preparations. It has been proposed that these substances can have a protective antimicrobial effect.

Key Words – Pigs' organs, Extract, Electrophoresis, Chromatography

I. INTRODUCTION

The growth in the demand for meat produce determines constantly increasing volumes of meat and meat products production which, in turn, lead to increase in quantity of secondary raw materials. In other words, processing of meat carcasses requires complex utilization.

Nowadays, the problem of processing of low-value raw material presents a challenge. From the authors' point of view, the term "low-value raw material" is quite misleading. Any raw material (tissues, organs, hides obtained in the process of farm animals processing) could serve as a source of valuable biologically active substances. Many years GNU VNIIMP named after V.M. Gorbатов has been developing the technology of obtaining medicines from endocrine-enzyme and special raw materials [1,2].

Organs of gastrointestinal tract and immuno-competent tissues of animals can serve as a source of biologically active substances. In experiments on laboratory animals with a model of antibiotic

associated diarrhea, a clear therapeutic effect was demonstrated when using Kolimak and Dinormin preparations [3,4].

The aim of the work was to study physico-chemical and biological properties of the extracts of the pigs' internal organs.

II. MATERIALS AND METHODS

The subjects of research were the preparations obtained by extraction of pig organs and tissues: pyloric and fundic sections of the stomach, pancreas, duodenum, spleen, mesenteric lymph nodes and thymus. On the basis of the pooled aqueous-salt extracts of these groups of tissues, two complex preparations Kolimak and Dinormin have been derived, which are recommended for treatment of diarrhea in weaned and sucking pigs [4].

Amino acid content was determined in the individual extracts and complex preparations on the Biotronik-IC-2000 analyzer (Germany) in mode of 3-buffer elution.

Electrophoretic examination was carried out in 12.5 and 15% polyacrylamide gels in the presence of 0.1% sodium lauryl sulfate when comparing with the standards of molecular masses in the range of 170 to 10 kDa.

The investigation of the preparations by high performance liquid chromatography (HPLC) was conducted on the chromatograph Agilent (USA) (column Phenomenex C18, 5 µm, detection at 280 nm, potassium phosphate buffer pH 3.4, flow rate 1.0 ml/min, temperature of the column thermostat 30°C, elution mode – isocratic).

Protein content was measured by Bradford method [5].

Thin layer chromatography on silica gel (Silufol F254, isopropanol-ammonia-water 14:1:5) was performed.

III. RESULTS AND DISCUSSION

Amino acid composition of the Kolimak and Dinormin preparations is presented in figures 1 and 2. It can be presumed that peptide-protein components being constituents of their composition belong to the globular proteins. As a result of two independent detections, quite close data on the amino acid composition were obtained.

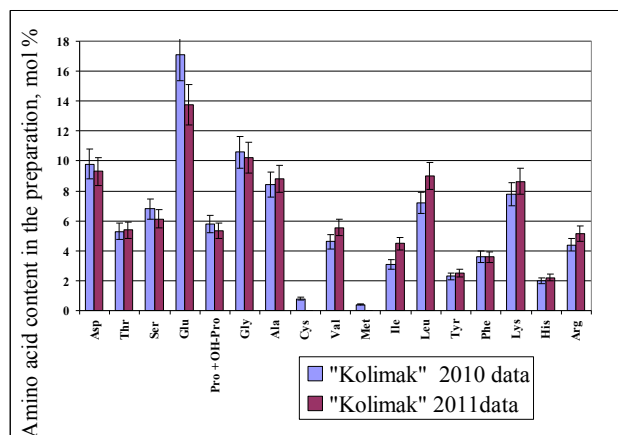


Figure 1 Amino acid composition of Kolimak preparation

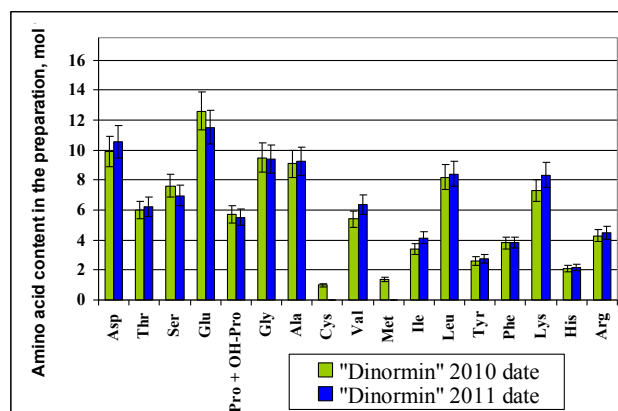


Figure 2. Amino acid composition of Dinormin preparation

Electrophoretic separation of the examined samples shows that they are all multi-component mixtures containing proteins in a wide range of molecular masses. Molecular masses of the

components being constituents of the individual extracts are in a range of 10 to 60 kDa (Fig. 3).

Electrophoresis images of the complex preparations revealed practically all components found in the tracks of the individual extracts. Considerable quantities of low molecular weight fractions with molecular masses about 10 kDa or lower were detected in the preparations of pancreas and thymus. It can be presumed that these fractions contain low molecular weight peptides and free amino acids, which, as well known from literature, can have a protective antimicrobial effect [6].

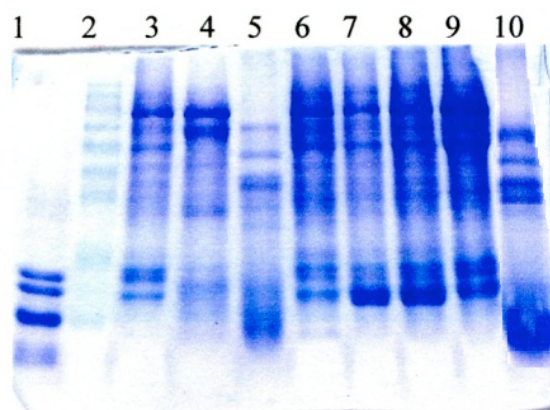


Figure 3. SDS-electrophoresis in 15% PAG:

1. standard of molecular mass for peptides (16.95-8.160 kDa)
2. standard of molecular mass (130kDa-10kDa)
3. extract of pyloric and fundic sections of the stomach
4. extract of duodenum
5. extract of pancreas
6. complex preparation Kolimak
7. extract of spleen
8. extract of mesenteric lymph nodes
9. complex preparation Dinormin
10. extract of thymus

The comparison of the data obtained by HPLC and electrophoresis in PAG confirms the presence of the components of a protein nature in a wide range of molecular masses.

The concentrations of proteins in extracts of pancreas, duodenum and mesenteric lymph nodes were 34.02, 10.73 and 15.58 μg , respectively. Chromatography of the preparations Kolimak and Dinormin is shown in figures 4, 5.

Multi-component nature of the individual extracts and the preparation Kolimak was also confirmed by thin layer chromatography on silica gel (Fig. 6).

IV. CONCLUSION

High heterogeneity of the individual extracts being constituents of the complex preparations Kolimak and Dinormin, the presence of the substances of a protein nature in a wide range of molecular masses were confirmed by electrophoresis and liquid chromatography. With that, the extracts of the organs which are a part of the gastrointestinal tract are quite individual, while the extracts of spleen and thymus shows significant similarity in the area

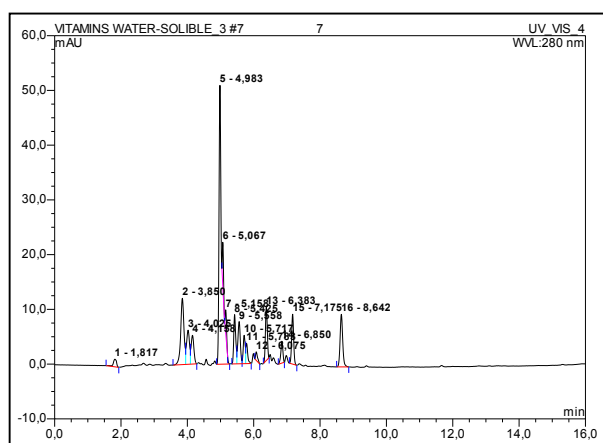


Figure 4 Chromatography of the Kolimak preparation

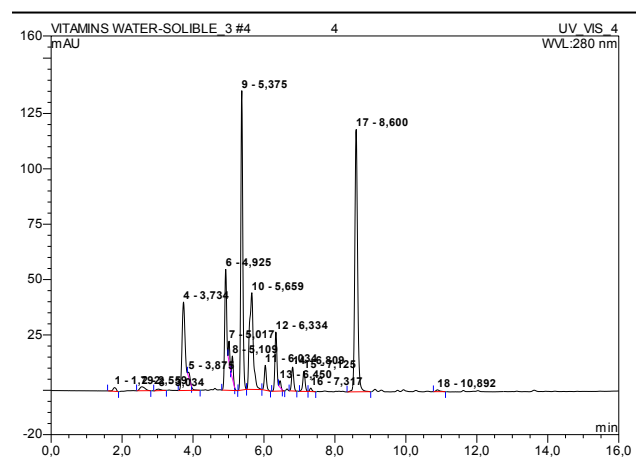


Figure 5 Chromatography of the Dinormin preparation

of the high molecular weight proteins, which could be explained by the closeness of their

physiological functions as immunocompetent organs.

A distinguishing characteristic of the studied extracts is the presence of considerable quantities of low molecular weight components in them.

Effective action of the preparations Kolimak and Dinormin is a sum of the physiological actions of each of the incoming component and exclusion of any of them can influence the spectrum of activity of the preparations.



A 1 2 3 4 5 6 7
Figure 6. Chromatogram of the examined samples

A – alanine (as a marker)

1. pyloric and fundic sections of the stomach
2. duodenum
3. pancreas
4. complex preparation Kolimak
5. spleen
6. lymph nodes
7. complex preparation Dinormin

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