lipid peroxidation in the organism of carp affected by aeromonosis and in the treatment of the drug "Flyumek" and its complex with milk thistle seeds.

Experiments were carried out on patients with aeromonosis of two-year-old carps, which, by analogy, were divided into three experimental groups, the control group was clinically healthy fish, with 4 individuals in each. The first group, clinically healthy fish, received 3% starch suspension, the second group received only 3% starch suspension, the third group, the experimental group, which received an antibacterial drug "Flumek" at the rate of 10 mg/kg of fish consisting of 3% starch suspension for 7 days, in the fourth experimental group, besides the similar dose of the antibacterial drug, 5% of the ground Milk thistle seeds was given spotted (Silybum marianum).

The results of the research showed that aeromonosis disease of carps by leads to an increase in the intensity of the processes of lipid peroxidation in blood plasma of fish and aldehyde and ketone derivatives of oxidation-modified proteins, as evidenced by a significantly higher level of their indicators in the sick fish compared to healthy. The introduction of the drug "Flumek" to carp which afected of aeromonosis, separately and in combination with the seeds of Milk thistle caused a possible decrease in the content of aldehyde and ketone derivatives of oxidative modification of proteins, lipids hydroperoxides and TBA-active products.

Keywords: carp, aeromonosis, «Flyumek», flumequin, Milk thistle, oxidatively modified proteins, lipid peroxidation

УДК 619:616 - 097.3

## PROTECTIVE ACTIVITY OF A COMPLEX ANTIGEN FOR THE DESIGN OF ASSOCIATED VACCINE WITH THE PURPOSE OF CONTROL OF BACTERIAL INFECTIONS OF POWDER

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**Annotation.** Significant damage to pig farms with traditional and sometimes industrial technology, the maintenance of association with bacterial infections - salmonella, pastereliosis, esherichiosis. Previously, for the control of the well-being of farms, a commercial PPD vaccine (pasteurellosis, paratyphoid, diplococcal infection) was used. Currently, the drug loses its

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protective capacity due to the proliferation of microorganisms in aetibic families with altered antigenic structure. To create a commercial drug of a new specimen, a number of studies have been carried out on its design and testing on a sensitive livestock. An experimental sample of an associated antigen proved to be quite competitive in immunological activity compared with commercial PPD vaccine. The protective activity of blood serum of animals after its application was 92 %, the level of T- and B-lymphocytes increased by 39.5 % and 22.7 %, respectively, the phagocytic activity in experimental group pigs increased by 7.1 % in comparison with the control (4.8 %). The survival of the pigs used by the experimental antigenic specimen in the groups after weaning was 99.5 % compared to the control group (75.4 %). Average daily gain of pigs the experimental group was at  $592.4 \pm 3.2 \,$ g, and at control  $-412.4 \pm 5.7 \,$ g.

Keywords: complex associate antigen, bacterial infections of piglets, projective activity of blood serum, phagocytic activity of white blood cells

**Topicality.** Pig Production in Ukraine is the leading industry in terms of providing meat products to the population. However, respiratory and gastrointestinal diseases of infectious etiology in many pig farms of Ukraine are stationary and are accompanied by significant losses, which reduces the profitability of production. In particular, it is a disease of the piglets before and after the ejaculatory age – salmonella, factor pasteurellosis and escherichia coli.

Analysis of recent research and publications. The stationary nature of these diseases leads to the carrier of microorganisms – potential stem cells [1, 2, 5]. In the scheme of preventive measures for the control of the mentioned infections, a concentrated polyvalent formolyaccine against paratyphoid, pasterellidosis and diplococcal septicemia of piglets (PPD) was used for a long time. However, with the discovery of new serological variants of pathogens of pasterelosis, salmonella, and escherichia, which present an inevitable threat to young pigs, the drug has lost its true feasibility [3, 4, 6]. In such conditions, there was an urgent need to create a new analogue drug that could fill a devastated niche in the list of so-needed biopreparations.

The purpose of the study was to compare the characteristics of the immunobiological status of the organism of piglets vaccinated with an associated antigen, which was developed on the basis of epizootic isolates of pasteurella, salmonella and escherichiae.

**Materials and methods of research**. Epizootic isolates adapted to the appropriate nutrient medium were used in the work. During the manufacture and research of some properties of antigens of salmonella, pasteurens and escherichia, classic and modern immunological methods of research were used.

The full-cell antigen was prepared from the bacterial mass. Up to 20 bcm/cm³of bacterial cells were added TCU in a ratio of 1:1. During the processing of TCU, the proteins settled, and the acid-soluble polycycle complex extracted from bacteria in a refrigerator at 0 °C for 3 hours. After that, by filtration, the extract was separated from the deposited proteins and dialyzed in cellophane sacs in running

tap water for 2 days and one day in distilled water in a refrigerator. In the process of dialysis, TCU and other easily dialylated substances were removed. Polycucker complex of catfish, as a colloid, did not penetrate the walls of cellophane bags. The completion of the dialysis was noted on the negative reaction of TCU and the neutral reaction of dialyzate. The dialysate, as a rule, was slightly opalescent. To determine the antigen for dialysis, an alcohol was added in 3–4 volumes. The precipitated antigen was separated by centrifugation. It was then treated with ether and dried lyophilically.

The content of protein fractions in both structures was determined by the Lowry method in Miller's modification.

Investigation of the created antigen was carried out in conditions of one of the pig farms of the Kiev region with traditional technology of maintenance. Previous studies of antigen activity in laboratory animals and extrapolation of these results, based on theoretical calculations on the pig's body, made it possible to determine the targeted quantity and multiplicity of antigen administration to form a protective effect on the part of the organism of productive animals. Optimal scheme of antigen application for porous sows: total dose of antigen – 15 cm³ intramuscularly (5 cm³ – first introduction and 10 cm³ followed at intervals of 14 days). For pigs, the scheme of antigen application to the offspring and in the afterlife period – In an increasing quantity – 3 cm³ for 14 days before weaning, 3 cm³ – for 7 days before weaning and 3 cm³ intramuscularly after 14 days after weaning.

The experimental groups of animals included 12 sows of large white breed whose fertility exceeded 3 months, and 20 pigs that were their newborn offspring. Animals of the control group (3 sows and 5 piglets), by analogy with the experimental scheme, injected a physiological solution in volume and at the same time, coinciding with the application of the antigen to the appropriate experimental groups of animals.

Blood for research was taken in the pigs before the experiment, and then on the 3rd and 7th day after vaccination and revaccination (14th and 21st day from the beginning of the experiment). To achieve the target in the blood of experimental and control animals, indicators of cell and humoral factors of their organism were determined. The presence of T-lymphocytes in the blood of vaccinated against pasteurellosis, salmonella, and escherichia was studied by spontaneous rosette of erythrocytes of the ram (E-RUK); The number of B-lymphocytes was determined by the socket-out method of the formation of erythrocytes, sensitized with the antibody and the complement, which are associated with the corresponding receptors on these lymphocytes (EC-RUK). A bio test for the activity of the received blood serum vaccinated with antigen and non-skeletal pigs was tested in a classical test of protective proteins of white mice.

Results of the research and their discussion. Studying the state of the blood, which samples were taken after the first administration to the sows of the complex antigen, no significant changes in the number of red blood cells, leukocytes and hemoglobin were demonstrated. During the month from the beginning of the experiment, there was an unreliable increase in the number of red blood cells. Among the cells of the leukocyte series regenerative shift of the

nucleus to the left to the young with a slight basophilia was revealed, indicating a positive dynamics of the benign post-vaccination process. Comparison of the results of the study of changes in the organism of experimental and control animals as a result of the application of a complex bacterial antigen, quantitative changes from the side-phagocytic response were noted. This effect is due not only to the quantitative but also to the functional activation of neutrophils (Table 1, 2).

1. Indicators of non-specific resistance of the body of pigs (n = 6), P < 0.05

Indexes	Animal groups vaccinated		Checking
	Experimental Antigen	Vaccine PPD	
Leukocytes, g/l	16,1 ± 0,92	15,6 ± 1,1	12,2 ± 0,35
Phagocytic activity, %	$54,6 \pm 3,5$	$50,2 \pm 5,0$	$46,5 \pm 3,2$
Intensity of phagocytosis,%	$6.4 \pm 0.8$	$5.8 \pm 0.8$	$4,0 \pm 0,4$

After 14 days after vaccination and revaccination, the phagocytic activity was significantly intensified: in pigs vaccinated with a complex experimental antigen test, by 7.1 %. When applied commercial PPD vaccine, this figure did not exceed 4.8 %. These figures compared with the similar ones received from the control group were 17.4 % and 7.4 %, respectively. The intensity of phagocytosis has also increased: by applying a complex experimental antigen test by 23.4 % compared to the baseline and 37.5 % against the control group of animals; as a result of the use of commercial PPD vaccine, respectively 11.4 % against the baseline and 31 % against the control group of pigs.

2. Comparative characteristic of indicators of cellular immunity of the piglet organism in the application of PPD and experimental antigen (P > 0.001)

Indexes	Animal groups vaccinated		Checking
	Experimental Antigen	Vaccine PPD	
T-lymphocytes,%	38 ± 1,7	34 ± 2,4	23 ± 1,3
B-lymphocytes,%	22 ±1,3	$20 \pm 0.7$	17 ± 1,7

After application of the experimental complex antigen, an increase in the absolute and relative number of lymphocytes was determined. The activity of the antigenic complex studied increased the number of T-lymphocytes in comparison with the control by  $38 \pm 1.7$  %, while the introduction of the commercial vaccine – by  $34 \pm 2.4$  %, respectively.

Quantitative indicators of the presence of B-lymphocytes in blood samples under the influence of the experimental antigen amounted to  $22 \pm 1.3$ % (in the control group –  $17 \pm 1.7$ %). Due to the use of commercial vaccine, this figure has increased, but only by 15% compared to control.

The protective activity of the blood serum of experimental sows after the first inoculation with the pasteurial component was 1:384  $\pm$  12.8, and after the second – it was not lower than 1:584. The antibody titre after re-inoculation for salmonellosis by its component was 1:260  $\pm$  8.0, for escherichiosis – 1:380  $\pm$  20. In the control group, the titres of antibodies in the pasteurial component did

not exceed 1:4.6; for salmonellosis it is -1:8,6; for escherichiae -1:16.3 (by average titles). As a result of determining the titres of agglutinins specific to pasteurals, it was found that in the experimental group of piglets the maximum antibody titer for the 45th day was 1: 340, at 75th -1:200.

In the control group, the piglets died one week after birth, causing the death of two piglets as a result of an associated bacterial infection. In the group of 14-day pigs of the experimental group as a result of the extinction of colostral immunity, the level of antibody titers exceeded the same figures for piglets in the control group by only 3-4 %.

The protective properties of blood serum from vaccinated sows with a complex experimental antigen were at least 92 % for each component, indicating an adequate level of protection. Conservation of piglets in the group after weaning was 99.5 %, and vaccinated with a commercial vaccine - 84.2 %. When compared with the control group (not vaccinated), the safety factor did not exceed 75.4 %. The average daily gain of piglets in the experimental group was  $592.4 \pm 3.2$  g, while in the control group it was  $412.4 \pm 5.7$  g.

Conclusions and perspectives. In this way, the experimental associate antigen containing the component against pasteurellosis, salmoneeloz and escherichiosis, caused in the body of his immunized pigs imunobiological reactions, which, compared with the beginning. The following variables had the following changes: the number of T- and B-lymphocytes increased by 39.5 % and 22.7 % compared to control; 14 days after vaccination and increased phagocytic activity: y piglets vaccinated with an associated antigen - 7.1 %, vaccine the PPP by 4.8 % compared to the start-up and by 17.4 % and 7.4 %, respectively, in terms of control new group; increased intensity of phagocytosis: when applying an experimental antigen test – by 23.4 % compared but with an initial level and 37.5 % against control group of animals; as a result of vaccination PPD - by 11.4 % against the entry level and by 31 % compared with the control group of pigs. The protective activity of the serum of vaccinated sows was 92 % for each structural component, indicating an adequate level of protection. Conservation of pigs used by the experimental antigenic specimen in the groups after weaning was 99.5 % compared to the control a group where conservation was not exceeded 75.4 %. Average daily gain of pigs the experimental group was 592.4 ± 3.2 g, and in control - 412,4 ± 5,7 g. Consequently, the experimental sample of the antigen creates a sufficient protective protection and can be successful used to construct prophylactic vaccines.

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# ПРОТЕКТИВНА АКТИВНІСТЬ КОМПЛЕКСНОГО АНТИГЕНА ДЛЯ КОНСТРУЮВАННЯ АСОЦІЙОВАНОЇ ВАКЦИНИ З МЕТОЮ КОНТРОЛЮ БАКТЕРІАЛЬНИХ ІНФЕКЦІЙ ПОРОСЯТ

#### Т. В. Мазур, І. В. Пронько

Анотація. Значної шкоди свинарським господарствам з традиційною, а інколи й промисловою технологією утримання завдають асоціації бактеріальних інфекцій — сальмонельоз, пастерельоз, ешерихіоз. Раніше для контролю благополуччя господарств використовували комерційну вакцину ППД (пастерельоз, паратиф, диплококова інфекція). Наразі препарат втрачає свою протективну здатність у зв'язку з поширенням мікроорганізмів аутентичних родин зі зміненою антигенною структурою. Для створення комерційного препарату нового зразка проведено ряд досліджень з його конструювання та випробування на чутливому поголів'ї. Експериментальний зразок асоційованого антигена виявився досить конкурентоздатним за імунологічною активністю

порівняно з комерційною вакциною ППД. Протективна активність сироватки крові тварин після його застосування була на рівні 92 %, рівень Т- і В-лімфоцитів зріс порівняно з показниками контрольної групи на 39,5 % і 22,7 % відповідно, фагоцитарна активність у поросят дослідної групи зросла на 7,1 % порівняно з контрольною (4,8 %). Збереженість поросят, яким застосовували експериментальний антигенний зразок, у групах після відлучення становила 99,5% порівняно з контрольною групою (75,4 %). Середній добовий приріст поросят у дослідній групі знаходився на рівні 592,4 ± 3,2 г, а в контрольній — 412,4 ± 5,7 г.

Ключові слова: комплексний асоційований антиген, бактерійні інфекції поросят, проективна активність сироватки крові, фагоцитарна активність клітин білої крові

# ПРОТЕКТИВНАЯ АКТИВНОСТЬ КОМПЛЕКСНОГО АНТИГЕНА ДЛЯ КОНСТРУИРОВАНИЯ АССОЦИИРОВАННОЙ ВАКЦИНЫ В ЦЕЛЯХ КОНТРОЛЯ БАКТЕРИАЛЬНЫХ ИНФЕКЦИЙ ПОРОСЯТ

#### Т. В. Мазур, И. В. Пронько

Аннотация. Значительный ущерб в свиноводческих хозяйствах с традиционной, а иногда и промышленной технологией содержания обусловливают ассоциации бактериальных инфекций – сальмонеллез, пастереллез, эшерихиоз. Ранее для контроля благополучия хозяйств использовали коммерческую вакцину ППД (пастереллез, паратиф, диплококковая инфекция). В настоящее время препарат теряет свою протективную способность в связи с распространением микроорганизмов аутентичных семейств с измененной антигенной структурой. Для создания коммерческого препарата нового образца проведен ряд исследований по его конструированию и испытанию на чувствительном поголовье. Экспериментальный образец ассоциированного антигена оказался достаточно конкурентоспособным в плане иммунологической активности по сравнению с коммерческой вакциной ППД. Протективная активность сыворотки крови животных после применения находилась на уровне 92 %, уровень Т- и В-лимфоцитов вырос по сравнению с показателями контрольной группы на 39,5 % и 22,7 % соответственно, фагоцитарная активность у поросят опытной группы увеличилась на 7,1 % по сравнению с контрольной (4,8 %). Сохранность поросят, которым применяли экспериментальный антигенный образец в группах после отъема, составляла 99,5 % по сравнению с контрольной группой (75,4 %). Средний суточный привес поросят в экспериментальной группе находился на уровне  $592.4 \pm 3.2$  г. а в контрольной  $-412.4 \pm 5.7$  г.

Ключевые слова: комплексный ассоциированный антиген, бактериальные инфекции поросят, проективная активность сыворотки крови, фагоцитарная активность клеток белой крови