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Symbiotic Probiotic Lactobacillus Acidophilus + Streptococcus Lactis In The Correction Of Microecological Disorders Of The Intestines Of Pigs.

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ABSTRACT

For the correction of intestinal biocenosis, as well as for the prevention and therapy of dysbiosis in animals and humans the main role is given to probiotics, the effectiveness of which is related to the inhibition of toxins, the formation of the barrier of the intestinal mucosa, preventing the attachment of pathogens, increasing the body's defenses and improving the balance of the intestinal microflora. Correction of the intestinal flora of the macroorganism should include complex probiotic preparations, which include a lactic acid liquid leaven (LAL) based on Streptococcus lactis and Lactobacillus acidophilus. The experience of the use of leaven for piglets in the amount of 20 ml per 1 head / day in mixture with the mixed fodder during the period of growing and fattening (from the second to the sixth months) contributes to an increase in body weight gain by 9,9%, allowing to receive additional weight gain from 8 to 11 kg per animal. The leaven removes the negative impact of contaminated with molds fodder on the immune system, improves the parameters of intestinal microbiocenosis. The amount of lactobacilli, lactococci and bifidobacteria in the intestine increases in more than 100 times in comparison with the parameters of the control group. Moreover, the addition of LAL to the poorly contaminated with molds mixed fodders contributes to the complete absence of fungal spores in the organs of animals (liver, heart) having the greatest nutritional value and used for the preparation of child nutrition.

Keywords: pigs, ration, symbiotic probiotic, normoflora, immunity, microbiocenosis, meat products

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INTRODUCTION

The disturbance of the normal microflora of the biological systems of the organism of animals and humans has a general ecological character associated with the global ecological changes in the environment. Evolutionally developed biocenosis between microorganisms and macroorganism is destabilized under modern conditions, causing various pathological conditions of the organism, passing into the clinical stage with the manifestation of various symptom-complexes [1].

In the necessary correction and prevention of dysbacteriosis the main role is given to probiotics. Probiotics are bacterial preparations of living microbial cultures, the effectiveness of which is associated with a beneficial effect on the body due to the inhibitory effect on toxin producers, the formation of a full barrier of the intestinal mucosa, preventing the attachment of pathogens to it, modulating of protective mechanisms of the body and improving the balance of the intestinal microflora [2]. This is achieved through the direct bacterial antagonism and the initiation of a specific immune response in intestine. Probiotics include not only lactic acid bacteria, but also other non-pathogenic species and strains: *E. coli*, bifidobacteria, etc. The most commonly used probiotics are strains of lactobacilli, bifidobacteria, yeast *Saccharomyces cerevisiae*, some strains of *Escherichia coli*. Bifido- and lactobacilli belong to the obligate (resident) intestinal microflora, whereas staphylococcus, streptococcus and yeast-like bacteria are not capable of prolonged existence in the body and enter the body from the environment. It is a transient microflora and it can be dangerous and undesirable for a living organism [3]. Correction of the intestinal flora of the macroorganism should include probiotic preparations, which are now generally divided into four main groups: probiotics – living microorganisms (liquid on milk whey and dry, lyophilized); prebiotics – substrates for maintaining the vital activity of normal microorganisms; synbiotics – probiotic + prebiotic substrate; symbiotics – complex of two or more probiotic cultures [4,5,6].

The effects of probiotics on the human and animal organism are manifested in several directions:

- general effects – control of potentially pathogenic microflora and reduction of endotoxins;
- humoral effects – inhibition of IgE synthesis with simultaneous stimulation of IgA;
- cellular effects that promote growth and regeneration of body cell.

It has been determined that probiotics activate tissue and humoral immunity of animals (T and B lymphocyte system) and affect the production of immunoglobulins. The effect of probiotic preparations can be considered as the introduction into the body of additional amounts of antigens, acting as modulators of the immune system, activating the specific defense of the organism and increasing the secretion of immunoglobulins of various classes [7, 8].

Feed additives based on lactic acid microorganisms possessing probiotic properties are able to have a positive effect on the animal organism and its intestinal microflora, as well as on the quality of meat products [9, 10].

METHODOLOGY OF RESEARCH

The scientific and economic experiment was carried out on two-month pigs of SM-1 breed in Breeding Plant "Leninsky Put" in the Novokubansky District of the Krasnodar Territory. Scheme of the research and production experiment is presented in Table 1.

Table 1: Scheme of scientific and production experiment (n=35)

Group	Feeding conditions
1– control	SR (standard ration in accordance with the periods of growing and fattening, contamination with micromycetes 1×10^4 ufc / g)
2 – experimental	SR + symbiotic lactic acid leaven (LAL) on the basis of <i>Lactobacillus acidophilus</i> and <i>Streptococcus lactis</i>

During the experiment the standard rations (SR) for both the experimental and control groups of animals were the same in composition and nutritional value and were calculated to obtain 700 g of body weight gain during the period of growing and fattening. In the ration of the experimental group the lactic acid leaven (LAL) was used in an amount of 20 ml per head / day in a mixture with mixed fodder during the period of growing and fattening (from the second to the sixth months).

The lactic leaven was grown in vitro on a nutrient medium (1,5% cow milk) and pre-tested in vivo on piglets, growing and fattened pigs from the microorganisms *Lactobacillus acidophilus* and *Streptococcus lactis* picked out from the luminal microflora of early-ripened pigs of SM-1 breed. The content of lactobacilli in the liquid leaven was 1×10^{10} - 4×10^{12} ufc / ml.

The animals were monitored according to growth, immunohematological and micro-biological (qualitative and quantitative composition of the intestinal normoflora) indicators.

RESULTS

Observations of animals showed that the lactic acid leaven (LAL) in the compound of feed significantly improved the growth parameters of piglets of the experimental group (Table 2). The difference in favor of the experimental group in terms of gross increase in live weight and average daily weight gain was 6,6% and 7,0% and was statistically significant ($P < 0.001$). The analysis of hematological tests carried out during the experiment indicates that practically all studied parameters in the experimental group of pigs were higher than the values of the control analogs, although they corresponded to the values of the species norm (Table 3).

Table 2: Influence of LAL on growth rates of experimental pigs (n=35)

Group	Body weight, kg		Gross increase in live weight, kg	Average daily weight gain, g
	at the beginning of the experiment	at the end of the experiment		
1- control	21,5±0,4	100,6±1,7	79,1±0,9	658,2±11,0
2 – experimental	21,2±0,6	105,5±1,0	84,3±1,1	704,5±12,2

Table 3: Immunohematological indicators of pigs in the experiment (n=35)

Indicators	Groups	
	1 – control (SR)	2 – experimental (SR+LAL)
Erythrocytes, $10^{12}/l$	4,21±0,7	4,95±0,5
Leukocytes, $10^9/l$	9,1±0,82	9,8±0,9
Hemoglobin, g/l	114,0±6,3	124,0±7,1
Hematocrit, %	25,2±1,1	32,0±1,4
Color metric	0,80±0,07	0,93±0,05
Platelets, $10^9/l$	185,0±11,3	258,0±8,6
Basophils, %	0,5±0,01	0,4±0,01
Eosinophils, %	1,3±0,1	1,0±0,2
Band neutrophils, %	1,67±0,3	2,00±0,2
Segmented neutrophils, %	43,7±3,2	48,6±2,8
Lymphocytes, %	50,4±4,6	45,4±5,0
Monocytes, %	2,5±0,9	3,0±0,5
T lymphocytes, %	54,2±2,6	63,4±3,2
B lymphocytes, %	41,3±2,0	46,7±1,4
NBT test: average cytochemical index stimul. <i>St. aureus</i>	1,39±0,09	1,35±0,07
NBT test: average cytochemical index spontan.	1,28±0,05	1,26±0,04

Studies of the cellular and humoral units of immunity showed that the use of lactic acid leaven promoted an increase in the content of T lymphocytes due to a decrease in zero cells (OK lymphocytes). Thus, in the experimental group the number of T cells by the end of the experiment was $63,4 \pm 3,2\%$, the number of B cells was $46,7 \pm 1,4\%$, while in intact animals these values were at the level of $54, 2 \pm 2,6\%$ and $41,3 \pm 2,0$ respectively. The difference in groups was 16,9% and 13,0%.

The use of LAL allowed to decrease NBT test results (spontaneous test with Nitroblau tetrasolium, 0,1% solution of which allows to assess the oxygen state of the dependent bactericidal mechanism of phagocytes (granulocytes) of blood in vitro). It characterizes the degree of activation of the intracellular NADPH oxidase antibacterial system. The NBT test results increase in the initial period of acute bacterial infections, whereas in the subacute and chronic course of the infectious process they decrease. Sanitation of the organism from the pathogen is accompanied by the normalization of the indicator.

One of the most important indicators of the physiological and immunological homeostasis of macroorganism is the state of the intestinal microbiocenosis. Our research of the relationship between the animal's organism and its intestinal microflora has shown that the latter plays an extremely important role in the formation and functioning of various organs and systems due to the production of various metabolites, enzymes, vitamins, biologically active substances, antigens and other compounds that are formed in the process of microbiological transformation from products of exogenous and endogenous origin.

The microflora of the digestive tract is characterized by relative stability, which is an essential factor supporting the permanence of the internal environment. It participates in many physiological reactions of the body, including the processes of digestion, metabolism, immune reactions, etc.

Due to its biochemical activity, the intestinal microflora is involved in the detoxication of xenobiotics. Moreover, the detoxifying ability of endogenous intestinal microflora is comparable with the detoxifying ability of parenchymal organs.

The conducted studies of intestinal microbiocenosis in animals showed that the use of LAL did not have a significant effect on the content of opportunistic microorganisms, however, the number of useful microflora (lactobacillus and lactococcus) increased by an order of magnitude. In addition, there were no storage molds and the amount of microscopic fungi of the genus *Candida* was significantly reduced (Table 4).

Table 4: Results of the study of intestinal microbiocenosis (lg ufc / g) in pigs (n=35)

Microflora	Group	
	1 – control (SR contamination with micromycetes 1×10^4 ufc / g)	2 – experimental (SR+LAL)
E. coli	6,3	6,0
Hemolytic E. coli	1,4	1,3
Enterococcus	9,3	9,0
Staphylococcus	5,0	4,5
Lactobacilli	9,0	10,0
Lactococcus	8,0	9,0
Bifidobacteria	6,0	7,3
Clostridia	4,3	4,0
Fungi <i>Candida</i> (spores)	2,0	1,0
Mold fungi (spores)	2,0	–

It should be taken into account that meat raw materials obtained from pigs grown in Breeding Plant "Leninsky Put" in the Novokubansky District of the Krasnodar Territory are used for the production of child nutrition based on meat. Therefore, its quality and safety must comply with state standards that ensure the production of organic pork [11].

To study the quality of the meat of pigs at slaughter, the longest muscle of the back and the general test of minced carcass were taken. In terms of pH and moisture capacity of meat there is no difference between the control group and the experimental group. However, in animals receiving LAL, was noted a tendency to decrease in fat content. On the other indicators of nutritional value (protein content, macro- and microelements), differences are not determined. Safety indicators of meat raw materials did not exceed the permissible levels.

Mycological examination of the internal organs of animals showed that the control group had the largest number of spores of microscopic fungi *Penicillium* and *Aspergillus*. In the animals of the experimental group spores of these fungi were absent and only in one animal they were found in the lungs. Due to this, presumably, the lactic acid leaven had a positive effect on the growth of pigs and the state of their health (Table 5).

Table 5: The content of spores of fungi *Penicillium* and *Aspergillus* in the internal organs of pigs (n=9)

Group	Amount of spores of fungi, ufc / g			
	lungs	liver	blood	heart
1 – control	83±9,9	250±23,7	93,0±11,0	30,0±3,6
2 – experimental	6,4 (in one animal from nine)	–	–	–

It should be noted that the addition of LAL to poorly contaminated with molds mixed fodders contributed to the almost complete absence of fungal spores in the organs of animals (liver, heart), which have the greatest nutritional value.

CONCLUSION

Probiotic lactic acid leaven based on lactic acid microorganisms *Streptococcus lactis* and *Lactobacillus acidophilus* (LAL) had a positive effect on the body of pigs. Its addition to mixed fodders increases the body weight gain of pigs by 9,9%, removes the negative effect of poorly contaminated with molds feed on the immune system, improves the parameters of intestinal microbiocenosis. The number of lactobacilli, lactococci and bifidobacteria in the intestine increased in more than 100 times in comparison with the control. In the organs of animals (liver, heart) used for the preparation of child nutrition there were practically no spores of fungi.

The presented data indicate that to reduce the negative effect of microscopic fungi in poorly contaminated with molds fodders on growth rates, the physiological state of animals and the quality of meat products, the use of a liquid lactic acid leaven LAL, containing 1×10^{10} - 4×10^{12} ufc / ml of *Streptococcus lactis* and *Lactobacillus acidophilus*, in rations of pigs during the periods of growing and fattening to obtain organic pork used for the production of child nutrition on a meat basis.

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