The use of biologically active supplements as an alternative for application of antibiotics in production of pork

A S Miroshnik, M I Slozhenkina, I F Gorlov, M V Frolova, D V Nikolaev1 and N I Mosolova
Volga Region Research Institute of Manufacture and Processing of Meat-And-Milk Production, Volgograd, Russia

1E-mail: dmitriynikolaev1978@yandex.ru

Abstract. The article presents materials of scientific research of possibility of using biologically active substances as an alternative for antibiotics in production of pork. The aim of this work is to study the effect of drug for veterinary use, that contain the main functional component – NONON, in comparison with biologically active additives «Lactusil» and «Sporothermin» in feeding young pigs on their growth and development. The studies were carried out on the basis of Lenin PZK of Surovikinsky district of Volgograd Oblast from January to June 2019. For this, 4 groups of large white piglets were formed, 30 heads each at the age of 2 months. In this case, the control group received a general economic diet (GED), animals of the I group received GED in common with VD in an amount of 0.5 g (5 mg of active ingredient) per day per 1.0 kg of feed for 14 days; II groups - GED + dietary supplement «Sporothermin» in amount of 1.0 kg per ton of compound feed; Group III - GED + dietary supplement «Lactusil» - at a dose of 0.2 ml / kg of live weight. The use of probiotics in the diets in comparison with VD has shown their high efficiency on growth, development and meat productivity of gilts. It was found that animals of groups II and III in biochemical, immuno-biological indicators and meat productivity were superior to analogs from control group and I group (treated with VD).

1. Introduction
Providing population with high-quality food products of animal origin remains the main task of Russian agricultural sector, which is especially important in view of the implementation of the program of Russian government to ensure food import substitution. One of the most dynamically developing branches of animal husbandry in our country is pig breeding due to modernization of existing livestock complexes and construction of new ones, as well as using of technologies that provide of improving quality indicators of animals by enhancement selection and breeding work when growing the best domestic breeds using foreign selection. At large pig breeding enterprises, antibiotics, pro, pre-, synbiotics and various growth-stimulating drugs are used to increase productivity, natural resistance, and realize a high genetic potential of farm animals [1-15].

The primary issue of intensifying pork production today is conservation of the livestock of young animals in initial postnatal period, which directly depends on the correct formation and development of internal organs. Pathologies of digestive system of pigs in large agricultural holdings and breeding farms are widespread and occur in more than 50% of cases. It is believed that causes of the pathologies are associated with increased activity of pathogenic intestinal microflora caused by feeding technologies impaired (unbalanced diets, violation of the transition from one food to another, etc.) and conditions for
keeping animals. In this regard, pig farms with a high concentration of livestock are forced to use antibiotics for the prevention and treatment of diseases [16-18].

The most effective means for the prevention and treatment of diseases caused by this kind of pathologies is using of antibacterial agents, including bicyclic thiopeptides [23].

In our studies, we have used drug for veterinary use (VD), that contain the main functional component – NONON, belonging to the group of bicyclic thiopeptides, which are capable of producing fungus Streptomyces actuosus. This drug is used as a powerful bactericidal agent that affect on gram-positive and gram-negative bacteria, including Clostridium Perfringens, Staphylococcus aureus, Streptococcus pyogenes hemolyticus, Streptococcus viridans, Streptococcus faccalis, Diplococcus pneumonia. The mechanism of action of VD on body of an animal is in a sharp disruption of protein synthesis in cells of sensitive microorganisms, which promotes to their rapid death. It should be noted that the drug is very effective against gram-positive bacteria that are resistant to majority antibiotics and do not have cross-resistance.

Earlier studies was found that using of the NONON, does not contribute to development of bacteria resistance. It has no mutagenic or teratogenic effects and does not contain GMOs.

When administered orally VD is not absorbed in gastrointestinal tract, does not accumulate in organs and tissues, and excreted unchanged. Currently, there is a clear tendency in the world to stop or reduce using feed antibiotics in animal husbandry. There are a number of alternatives for feed antibiotics that are successfully used in livestock production, such as pro- and prebiotic dietary supplements.

One of these prebiotics is «Lactusil», which is recommended for use in cattle breeding as a biologically active feed additive (RU Specification TU 9197-157-10514645-08, developed by I F Gorlov et al., GNU NIIMMP). However, to date, there is no information on its use in pig breeding in the available information sources.

«Lactusil» is a mixture of biologically active substances obtained by combining lactulose and honey extract from sprouted milk thistle seeds. The supplement contains a balanced set of lactulose (18.0%), flavonoids (not less than 8.0 mg / 100 g), polyphenols (not less than 0.30 mg / 100 g), and traces of lactose, galactose and other carbohydrates. These active components can present in contents of colon of farm animals, as well as in mucous membranes and salivary secretions. Normally, these compounds perform a number of the following important functions in the body: stimulate the growth of lactic acid microorganisms and bifidobacterium, inhibit the growth of pathogenic microflora, play the role of energy metabolites for epithelial cells, and substrate for gluconeogenesis. The use of additive «Lactusil» in the diets activates digestion and restoration of damaged mucous membranes, increases microbiological resistance of intestinal mucosa, and helps prevent dysbacteriosis diarrhea.

The mechanism of therapeutic and preventive action of the developed dietary supplement is based on positive effect of lactulose, which is associated with the stimulation growth of bifidobacterium. It has been scientifically proven that when lactulose is taken orally, process of its absorption begins only in the lower part of intestine, because only there are enzymes necessary for its breakdown present.

In our studies, we also used another feed additive – «Sporothermin», which had been developed by specialists of production association «VetSelkhoz». It is a homogenous, highly dispersed powder with a slightly milky odor, white or creamy color. The studied feed additive contains sublimated bacterial spores B. Subtilis G-28 (VKPM 2159) and B. Licheniformis 94 (VKPM 2985) in a ratio of 50/50 and a filler lactose. One gram of feed additive «Sporothermin» contains at least 3∙109 CFU live spores.

The use of the feed additive in animal feeding allows to increase non-specific immunity, improve functioning of gastrointestinal tract and enzymatic activity, as well as increase live weight gain.

Studies on the influence of a VD, probiotic and prebiotic in comparative aspect in productive capacity young pigs were not carried out to date.

The purpose of this work is to study the effect of VD in comparison with biologically active additives «Lactusil» and «Sporothermin» in feeding young pigs on their growth and development.
2. Materials and methods
The studies were carried out on the basis of a farm-breeding plant named after Lenin of Surovikinsky District of Volgograd Region from January 2019 to June 2019. For this, 4 groups of large white piglets were formed, 30 heads each at the age of 2 months.

In this case, the control group received a general economic diet (GED), animals of group I received GED in common with VD in an amount of 0.5 g (5 mg of active ingredient) per day per 1.0 kg of feed for 14 days; group II – GED + dietary supplement «Sporothermin» in the amount of 1.0 kg per ton of compound feed; group III – GED + dietary supplement «Lactusil» – at a dose of 0.2 ml / kg of live weight (table 1).

Table 1. Dosage of tested drugs.

| Index                  | Group control | I                      | II                      | III                      |
|------------------------|---------------|------------------------|------------------------|------------------------|
| Animal weight, kg      | 14.92±0.72    | 15.25±0.60             | 14.96±0.50             | 15.0±0.86              |
| Dosage of test drugs   |               | VD Sporothermin        | Lactusil               |
| per 1.0 kg of feed     | 0.33±0.02 g / day | 1.0 kg / ton of       |
|                        |               | compound feed          | weight                 |

The composition of 1 kg of GED for young pigs contained: exchange energy 12.34 MJ; dry matter 88.24%; crude protein 16.22%; crude fiber 5.86%; lysine 0.62%; methionine with cystine 0.47%; threonine 0.54%; calcium 0.66%; phosphorus 0.52%, as well as vitamins (A, D3, E, K3, B1, B2, B3, B4, B5, B6, B12) and trace elements (iron, copper, zinc, manganese, cobalt, iodine and selenium). Experimental animals received food twice – in the morning and in the evening with free access to water.

Zootechnical, biochemical and hematological studies were carried out by conventional methods [2].

The quantitative determination of the level of immunoglobulins of certain classes (isotypes) was performed in duplicate by the method of radial immunodiffusion according to the Mancini method using monospecific antisera and monoclonal antibodies to certain classes of immunoglobulins and a reference standard blood serum of pigs with a known content of immunoglobulins of certain classes [22].

To determine the dynamics of live weight of experimental animals, weighing was carried out monthly up to 180 days of age.

3. Results and discussion
The results of studying biochemical parameters of experimental animals blood are shown in figure 1. The data in the figure indicate that inclusion of the tested agents in the diets of young pigs in the experimental groups contributed to the optimization of the biochemical status of the organism. Gilts of groups I, II and III exceeded their counterparts in the control group in terms of total protein content in blood by 0.06 g / l, or 0.07%; 3.9 g / l, or 4.82% (P≤0.05) and 3.0 g / l, or 3.71% (P≤0.05), albumin - by 1.46 g / l, or 3.67% (P≤0.05); 0.07 g / l, or 0.18% and 0.09 g / l, or 0.23%; globulins – by 0.37 g / l, or 0.93%; 1.95 g / l, or 4.88% (P≤0.05) and 1.63 g / l, or 4.08% (P≤0.05), respectively. The blood of animals of groups I, II and III contained more β and γ-globulins, calcium and inorganic phosphorus in comparison with control analogs, but the difference was insignificant and unreliable.

Introduction of test drugs into the diets of young pigs in experimental groups helped to increase immunobiological defense of organism. Increase of content of protein, globulins, β-globulins in blood serum of pigs treated with VD, pro- and prebiotics, indicates improvement of digestibility and an acceleration of biosynthesis process of proteins. According to biological role in the body, blood serum globulins are cells of immune system. Our experimental data are consistent with the results obtained in the work of Dezhatkin S, Mukhitov A and Dozorov A et al., 2013.
Figure 1. Biochemical parameters of experimental animals blood (n=4).

In general, it should be noted that in the blood serum of animals of all studied groups, a relatively high content of calcium and phosphorus was found.

Table 2 shows the results of morphological studies of blood obtained from animals of the compared groups.

| Index                          | Unit          | Group control | I          | II         | III         |
|--------------------------------|---------------|---------------|------------|------------|-------------|
| Erythrocytes                   | $10^{12}$/l   | 6.36±0.03     | 6.35±0.02  | 6.36±0.03  | 6.37±0.02   |
| Leukocytes                     | $10^9$/l      | 8.29±0.04     | 8.26±0.02  | 8.32±0.03  | 8.28±0.02   |
| Hemoglobin                     | g /l          | 104.69±1.36   | 105.32±0.80 | 105.68±1.37 | 104.72±1.33 |
| Sedimentation rate of erythrocytes (ESR) | mm /h     | 2.84±0.17     | 3.12±0.14  | 3.38±0.73  | 3.41±0.15   |

The study of morphological composition of blood obtained from animals of all studied groups showed that there were no significant differences in content of erythrocytes and leukocytes. However, the hemoglobin content in the blood of young pigs of groups I, II and III was higher in comparison with the analogs of control group by 0.63 g / l, or 0.60%; 0.99 g / l, or 0.95% and 0.03 g / l, or 0.03%; ESR - by 0.28 mm / h, or 9.86%; 0.54 mm / h, or 19.01% and 0.57 mm / h, or 20.07%, respectively.

One of the most important indicators studied in blood serum are immunoglobulins, since they show the level of evolution of the immune response of animal body to effects of pathogens, including gram-positive bacteria.
It was experimentally found that in blood serum of the studied pigs, the content of IgG immunoglobulins was higher in groups I, II and III in comparison with the control by 0.11 mg / ml, or 0.92%; 0.51 mg / ml, or 4.29% (P≤0.001) and 0.71 mg / ml, or 5.97% (P≤0.001); IgA – by 0.10 mg / ml, or 5.15%; 0.31 mg / ml, or 15.98% (P≤0.001) and 0.37 mg / ml, or 19.07% (P≤0.001); IgM – by 0.02 mg / ml, or 3.66%; 0.07 mg / ml, or 12.73% (P≤0.001) and 0.12 mg / ml, or 21.82% (P≤0.001), respectively (figure 2).

The level of lactic acid bacteria in gastrointestinal tract has a direct effect on content of immunoglobulins IgG and IgA in blood serum in such a way that an increase in some leads to an increase in others. From the analysis of literature data, it is known that in the body of animals to suppress growth of pathogenic or opportunistic pathogenic microorganisms, IgM immunoglobulins are produced. In our studies, the highest amount of this blood cells was found in the blood serum of animals of groups I, II and III in comparison with control analogues. Gilts all studied groups were healthy. Research data analysis showed superiority of indicators of physiological status of studied animals I, II and III groups in comparison with analogs of the control group. This circumstance indicates a better metabolism in their body. The results of calculating absolute live weight gain are presented in table 3.

### Table 3. Absolute live weight gain rates of experimental animals (n=30).

| Age, days | Absolute live weight gain, kg |          |          |          |
|-----------|-----------------------------|----------|----------|----------|
|           | control                     | I        | II       | III      |
| 60-90     | 21.8±0.09                   | 21.6±0.10| 22.0±0.08| 21.8±0.07|
| 90-120    | 23.0±0.12                   | 23.4±0.13| 22.7±0.16| 23.3±0.14|
| 120-150   | 25.3±0.18                   | 25.4±0.15| 26.2±0.17**| 26.7±0.20**|
| 150-180   | 27.6±0.19                   | 27.7±0.14| 28.4±0.17*| 28.9±0.21**|
| 60-180    | 101.4±0.23                  | 102.6±0.19*| 103.4±0.27*| 104.4±0.25**|

The data presented in table 3 indicate that animals of the II and III experimental groups, from 120 to 150 days, surpassed their counterparts in absolute live weight gain by 0.90 kg, or 3.56% (P≤0.01) and 1.40 kg, or 5.53% (P≤0.01); from 150 to 180 days – by 0.8 kg, or 2.90% (P≤0.05) and 1.30 kg, or 4.71% (P≤0.01), respectively. Animals of control group were inferior to their counterparts from group I in absolute live weight gain by 1.20 kg, or 1.18% (P≤0.01), II – by 1.0 kg, or 0.99% (P≤0.05), III – by 2.0
kg, or 1.97% (P≤0.01). During the whole period of experiment farm animals of the I, II and III groups reached a live weight of 117.85 kg, 117.36 and 118.40 kg, while the control group analogs weighed 116.32 kg.

In studies Liu J et al., 2003; Sukonina V et al., 2006; Sonnenburg E D et al, 2010 and Fu R et al., 2019, relationship between growth of microflora of the gastrointestinal tract and absorption of nutrients was found.

Thus, the suppression of opportunistic microorganisms can promote the development of useful biota and stimulate metabolic processes in young pigs. This, we believe, activates protein synthesis and favors body weight gain.

4. Conclusion
During experimental studies, a positive effect on physiological parameters of experimental young pigs of introduction of VD, the prebiotic «Lactusil» and probiotic «Sporothermin» has been found.

The highest results in terms of meat productivity were obtained when the dietary supplement «Lactusil» was introduced into the diets of experimental young animals. In our opinion, this is associated with a higher growth of lactic acid bacteria in gastrointestinal tract of the animals of group III compared with other groups.

The increase in the serum levels of immunoglobulins IgG, IgA and IgM in animals of groups I, II and III in comparison with analogs of the control group allows us to judge the potential growth of lactic acid bacteria in gastrointestinal tract, which had an effect on the increase in live weight. The use of probiotics in the diets in comparison with VD has shown their high efficiency on growth, development and meat productivity of gilts.

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