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The influence of biologically active additives on the morpho-biochemical parameters of the blood of broiler chickens

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Abstract. The introduction of synbiotics and phytobiotics into the diet allows us to optimize metabolism, increase the level of use of feed components, and activate immune processes in the body. **The purpose of the work** is to determine the effect of "GerbaStor" and "ProStor" feed additives, including probiotic, prebiotic and phytobiotic components, on the morphological and biochemical parameters of the blood of broiler chickens of the "Ross-308" cross. **Methods.** In the course of a scientific and economic experiment at the age of 29 days, blood was taken from 5 medium-sized broilers by decapitation to determine morphological and biochemical parameters. The blood test was carried out at the Clinical and Diagnostic Center in Ekaterinburg. The morphological examination of blood was carried out in the hemostasis laboratory, on an automatic analyzer ADVIA 120, manufactured by BAYER, as well as by manual counting of formed elements in the Goryaev chamber. A biochemical blood test was performed in a clinical diagnostic laboratory using a Vitros 350 analyzer. **Results.** The inclusion of "GerbaStor" and "ProStor" in the diet did not adversely affect the metabolic processes of the bird. Morpho-biochemical blood parameters were within physiological values. In the experimental groups was observed an improvement in the assimilation of protein nitrogen, it is evidenced by a decrease in uric acid in the blood serum. In addition, the use of the studied feed was accompanied by the activation of lipid metabolism in the body of birds, which was characterized by a tendency to reduce the amount of triglycerides in the blood serum in individuals of the 1st and 2nd experimental groups. The results of the study showed that when "ProStor" is introduced into the feed concentrate, there is a tendency of decreasing in the activity of transaminases in the body of broilers, which demonstrates a higher resistance of their internal organs, in particular the liver and heart, to unfavorable exogenous and endogenous influences. **The academic novelty** lies in the fact that new data were obtained on the influence of the studied feed factors, including probiotic, prebiotic and phytobiotic components, on the morpho-biochemical parameters of the blood of broiler chickens.

Keywords: morphological and biochemical parameters of blood, broiler chickens, synbiotics, phytobiotics.

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Introduction

It is a well-known fact that blood is an important medium for all cells and tissues of animals and birds. Thanks to the blood, there are transported oxygen and nutrients and there are removed metabolic products and carbon dioxide in the body [3]. Also, blood is called the liquid tissue of the body. Circulating through the vessels, it ensures the regulation of the hormonal system and creative connections, is the carrier of immunity factors.

According to V. A. Gudin, V. F. Lysov, V. I. Maksimov, the amount of blood in chickens is from 180 to 315 ml or from 8.5 to 13 % of body weight. 65–70 % of the blood takes part in the circulation in the general blood flow, the rest is deposited and is used only when necessary [7].

Poultry blood is characterized by a relatively constant composition, and the resulting quantitative changes in its components reflect the features of redox and metabolic processes in the body [11]. The conditions for feeding birds have a significant effect on the morphological and biochemical parameters of blood.

Today, the use of synbiotic and phytobiotic additives in poultry feeding with meat productivity is promising [4], [16], [17].

Synbiotics and phytobiotics are new generation additives that have a stimulating effect on the growth and development of poultry [2], [10], [12]. Their introduction into the diet allows to optimize metabolism, increase the level of use of feed components, and activate immune processes in the body [13], [14].

Synbiotics are commonly understood as feed additives, which include probiotics and prebiotics. The main purpose of using synbiotics in animal husbandry is to restore and maintain a healthy intestinal microflora.

Phytobiotics are natural composition additives, based on raw materials of plant origin. This type of supplement is effective as a means to maintain an optimal and stable state of the gastrointestinal tract [1], [5], [10], [15].

The above drugs include biologically active additives "GerbaStor" and "ProStor".

"GerbaStor" contains live spore-forming bacteria of the genus Bacillus and lactic acid microorganisms, products of their metabolism (enzymes, organic acids, vitamins), fermented beet pulp, yeast autolysates, mineral salts, carbohydrates, herbal supplements (oregano herb, plantain leaf, chamomile flowers, herba hyperici).

The composition of the feed additive "ProStor" includes spore-forming microorganisms Bacillus subtilis auxiliary substances – fermented beet pulp, yeast autolysates, mineral salts, carbohydrates, phyto-additives (Echinacea purpurea herb, milk thistle fruits). The drugs are produced by LLC STC "BIO" by the method of microbiological synthesis.

Methods

The aim of the study was to determine the effect of feed additives "GerbaStor" and "ProStor" on the morphological and biochemical parameters of the blood of broiler chickens of the "Ross-308" cross.

The experimental part was carried out on the territory of the poultry house of the educational and experimental farm of the Ural State Agrarian University according to the scheme presented in table 1.

For the study there were formed three groups of poultry, 44 heads in each: control and two experimental. The average live weight of one chick in each group at the time of their formation was 42 g. From the 1st to the 5th day of life, all experimental birds received pre-starter compound feed. Starting from the 5th day, the diet of the 1st experimental group included the drug "GerbaStor" in the amount of 0.5 g per 1 kg of compound feed; the feed additive "ProStor" was introduced into the diet of the 2nd experimental group in the same dosage. For raising poultry, the same conditions were maintained throughout the experiment. At the age of 29 days, blood was taken from 5 average broilers in the group by decapitation to determine morphological and biochemical parameters. The blood test was carried out at the Clinical and Diagnostic Center of Ekaterinburg. The morphological examination of blood was carried out in the hemostasis laboratory, on an automatic analyzer ADVIA 120, manufactured by BAYER, as well as by manual counting of formed elements in the Goryaev chamber. A biochemical blood test was performed in a clinical diagnostic laboratory using a Vitros 350 analyzer (Ortho-Clinical Diagnostic, USA).

Results

The blood contains such shaped elements as: erythrocytes, leukocytes and platelets. The content of these elements is relatively constant.

Erythrocytes are red blood cells, have the shape of an ellipsoid, contain a nucleus, their formation in adult birds occurs in the red bone marrow from hemocytoblasts. The main function of erythrocytes is gas exchange. They carry out the transfer of O_2 from the lungs to the tissues of the body and the transport of CO_2 back [7]. В таблице 2 представлены морфологические показатели крови цыплят-бройлеров.

The inclusion of experimental feed additives in the diet contributed to an increase in the number of erythrocytes in the poultry blood: the superiority in 1st and 2nd experimental groups in comparison with the control was 4.7 and 3.9 %, respectively. The ratio of erythrocytes to total blood volume, or hematocrit, in 1st and 2nd experimental groups was higher than in the control by 1.69 and 1.81 %, respectively.

When assessing the amount of hemoglobin in the blood of broilers, there was noted a tendency of its increase in experimental individuals. This indicates that the introduction of feed additives "GerbaStor" and "ProStor" influenced on the activation of redox processes in the body. The highest hemoglobin content was in chickens that received "ProStor": the difference with the control group was 5.53 %. In the broilers of the 1st experimental group, who received "GerbaStor" with the main diet, the amount of hemoglobin in the blood was 4.3 % higher than in the control.

It is possible to determine the concentration of iron-containing protein in a red blood cell by the indicator - the average content of hemoglobin in one erythrocyte. This indicator in the 2nd experimental group was higher than in the control by 1.41 %, while in the 1st experimental group, on the contrary, it was lower in comparison with the control by 0.36 %. A similar situation was observed in the experimental groups with an average concentration of hemoglobin in the erythrocytes of the blood of chickens. In 1st experimental group, this indicator was lower than in the control group by 0.29 %, and in 2nd experimental group, on the contrary, it was higher than in the control group by 0.4 %.

Leukopoiesis is the formation of white blood cells. Leukocytes are called white blood cells that contain a nucleus and are characterized by an inconsistent shape. Their concentration can vary over a wide range. An increase can be observed even after ingestion of feed. After analyzing the morphological parameters of the blood of the experimental bird, it was found that leukopoiesis in the broilers of the experimental groups was enhanced. This is evidenced by the increase in the content of leukocytes in the 1st and 2nd experimental groups, in relation to the control by 8.03 and 14.75 %, respectively.

Monocytes are characterized by bactericidal and phagocytic abilities. An increase in their number occurs during inflammatory processes in the body. The function of lymphocytes is the formation of specific immunity, as well as the implementation of immune surveillance. Comparison of the percentage of different types of leukocytes in the blood of broilers showed that in all experimental groups the number of lymphocytes and monocytes was less than the control level: in 1st experimental group – by 5.2 and 1.0 %, in 2nd experimental group – by 11.8 and 0.6 %, respectively.

Table 1

Scheme of scientific and economic experience

Group	*Number of heads*	*Feeding conditions*
Control	♂ 22 ♀ 22	*The basic diet (BD) is a compound feed with a nutritional value corresponding to the recommendations for cross-breeding.*
1st experienced	♂ 22 ♀ 22	*BD + "GerbaStor" in the amount of 0.5 g/kg of compound feed, from the 5th day of growing until the end of the feeding period.*
2nd experienced	♂ 22 ♀ 22	*BD + "ProStor" in the amount of 0.5 g/kg of compound feed, from the 5th day of growing until the end of the feeding period.*

Table 2

Morphological parameters of blood of broiler chickens at the age of 29 days, M ± m (n = 5)

Indicator	Group		
	Control	1st experienced	2nd experienced
Red blood cells, 10^{12}/l	3.38 ± 0.20	3.54 ± 0.13	3.51 ± 0.13
Average red blood cell volume, femtoliter (fl), or mm^3	121.84 ± 0.80	121.78 ± 1.2	138.94 ± 17.05
Distribution of red blood cells by volume, %	8.10 ± 0.07	8.04 ± 0.13	6.56 ± 1.78
Hematocrit (the ratio of red blood cells to total blood volume), %	36.31 ± 1.97	38 ± 1.24	38.12 ± 1.17
Hemoglobin, g/l	97.80 ± 5.44	102 ± 2.83	103.20 ± 3.07
Average hemoglobin content in one red blood cell, picogram (pg)	32.82 ± 0.15	32.70 ± 0.66	33.28 ± 0.35
White blood cells, 10^9/l	269.40 ± 1.57	268.60 ± 3.31	270.40 ± 0.57
Lymphocytes, %	19.80 ± 1.87	21.39 ± 1.33	22.71 ± 1.96
Monocytes, %	66.80 ± 1.71	61.60 ± 2.71	55 ± 1.87**
Pseudoeosinophils, eosinophils, %	10.40 ± 1.89	9.40 ± 0.91	9.80 ± 0.55
Basophils, %	21.00 ± 0.87	26.60 ± 1.48*	33.40 ± 1.52***
ESR, mm/hour	1.80 ± 0.42	2.40 ± 0.57	1.80 ± 0.42
ESR, mm/hour	2.6 ± 0.45	2.4 ± 0.27	2.6 ± 0.45

The content of pseudo-eosinophils and eosinophils (in total) in the control group was at the level of 21.00 %. The use of the tested feed additives in the diet was accompanied by an increase in these groups of leukocytes in the blood of chickens: in the 1st experimental group – by 5.6 %, and in the 2nd experimental group – by 12.4 % ($P \leq 0.001$). These changes were within the standard values.

The function of basophils is the production of histamine and heparin, which in turn prevent blood coagulation, and also promote relaxation of capillaries, blood flow and resorption of inflammatory foci. The level of basophils in the blood of broilers of all groups corresponded to the physiological requirements for birds of this age and ranged from 1.8 to 2.4 %.

The erythrocyte sedimentation rate in the chickens involved in the experiment corresponded to the standard level and ranged from 2.4 to 2.6 mm/h.

Table 3 shows the data of biochemical parameters of the blood of broiler chickens at the age of 29 days.

Analyzing the content of total protein in the blood serum of meat poultry, it can be seen that the inclusion of the feed additive "GerbaStor" in the diet of the 1st experimental group contributed to its slight decrease in comparison with the control group – by 0.5 %. In the 2nd experimental group, the birds of which received a complex additive "ProStor", including synbiotics and phytobiotics, in the composition of the feed, the total protein value tended to increase by 4.32 %. Indicators in the experimental groups corresponded to physiological norms.

The amount of albumin in the blood serum of the 1st experimental group was 3.11 % lower than the level of the control group. In 2nd experimental group, this indicator was the same as in the control group and amounted to 10.28 g/l.

The maximum amount of globulins was noted in the blood serum of broilers of the 2nd experimental group, which exceeded the control by 6.93 %. The level of globulins in chickens of the 1st experimental group slightly outstripped the control – by 1.06 %. The ratio of albumins to globulins – A/G index, was 0.60 in the control group; in 1st experimental group 0.58; in the 2nd experimental group – 0.57 units.

With protein metabolism in the body of the bird there is formed the final product – urea. It is represented by diamide of carbonic acid, which is formed in the liver in the process of neutralizing ammonia. The synthesis of urea is carried out by means of a special group of enzymes. In the blood and internal organs, it does not perform any functions. By the content of urea in the blood, the balance between the rate of synthesis in the liver and the rate of excretion by the kidneys in urine is assessed. Thanks to this compound, the safe excretion of nitrogen from the body is ensured. In the course of the study, it was found that the introduction of "GerbaStora" into the mixed feed of chickens from the 1st experimental group did not have any effect on this indicator, it was on the same level with the control and amounted to 0.26 mmol/l. At the same time, in the 2nd experimental group, there was a significant decrease in this metabolite in the blood serum of broilers in comparison with the control: the difference was 15.38 %. This allows us to say that in the body of the birds of the 2nd experimental group there was a decrease in catabolic processes and an increase in the degree of assimilation of nitrogen of free amino acids. All these factors have a positive effect on the productivity of broilers.

One of the chemical compounds in the body that is formed after the breakdown of proteins is creatinine. Thanks to this compound, bioenergetics is carried out at the level of mitochondria. The body ultimately does not use creatinine in any way, so it is excreted from it. With a high accumulation of it in the body, it begins to have a toxic effect.

Based on the data of scientific and economic experience, it can be seen that in birds of the 1st and 2nd experimental groups, the amount of creatinine was at the level of the control value, being in the range of 23.06–23.34 μmol/l, which corresponds to physiological norms.

The results of a biochemical study of blood serum showed that in broilers of all experimental groups, the amount of uric acid (the main end product of protein metabolism in birds) did not exceed the optimal level. At the same time, the lowest value of this metabolite was noted in the 2nd experimental group – 131.13 μmol/l, which is 33.36 % lower than the control. In 1st

experimental group, there was also a tendency to decrease this metabolite in the blood serum: the difference with the control was 26.45 %. The established changes in the content of uric acid in the blood serum of chickens from the experimental groups indicate that the bird began to better assimilate protein nitrogen. This is confirmed by a higher average daily gain in live weight of birds that received "GerbaStor" and "ProStor", compared to control analogs by 2.1 and 1.8 % on average during the feeding period and a decrease in feed consumption per 1 kg of live weight gain by 1.14 and 2,8 % respectively. The level of carbohydrate metabolism was demonstrated by the serum glucose content. The role of this metabolite is energy and plastic functions in the body. A higher content of glucose in blood serum was noted in chickens of the 2nd experimental group – 14.29 mmol/l, which was 7.37 % higher than in the control. In the broilers of the 1st experimental group, the difference in this indicator with the control was insignificant – lower by 0.3 %.

Lipid metabolism was assessed by the concentration of such components as cholesterol and triglycerides in the blood serum of chickens. Cholesterol is found in all cells of living organisms, being necessary from the very beginning of development. Through homeostasis of the body, the level of this component is approximately at the same level under any exogenous influences [6]. In our studies, the level of cholesterol in birds of the control group was 2.94 mmol/l, in chickens of the 1st experimental group, its decrease was observed in relation to the control level by 6.46 %, and in the 2nd experimental group, the increase was by 12.59 %.

With regard to the content of triglycerides, a tendency of their decrease was established in chickens of the 1st and 2nd experimental groups, in comparison with the control, by 32.1 and 21.43 %, respectively, which indicates the activation of lipid metabolism in the body of the bird and is consistent with the studies of other authors [8], [9].

The enzymes ALAT and ACAT transport amino acids from one molecule to another [6]. Amino acids are needed to build proteins.

The obtained results indicate that the level of these enzymes did not differ significantly between the groups. There was a decrease in the level of ALAT in broiler chickens of the 2nd experimental group in comparison with the control by 8.55 % and an increase in chickens of the 1st experimental group by 19.7 %. Since this enzyme is synthesized in predominant amounts in liver cells, it can be assumed that there is less cellular damage in this organ in birds receiving "ProStor".

Similar to the changes in ALAT, the activity of the ACAT enzyme in the blood serum of the chickens from the experimental groups changed. Its amount in the blood of broilers of the 2nd experimental group was less, compared with the control, by 6.73 %, and in broilers of the 1st experimental group, it exceeded the control value by 11.1 %.

During the analysis of the mineral composition of the blood serum of the experimental broilers, it was found that in all the birds participating in the experiment, the calcium level corresponded to the physiological norm, while in the experimental groups the content of this macroelement was lower than the control by 1.92–3.07 %. The amount of phosphorus in the blood of birds of the 1st and 2nd experimental groups exceeded the control value by 13.75 and 5.42 %. The content of potassium in the blood of the experimental birds ranged from 4.85 to 5.65 mmol/l, sodium in the range from 150.4 to 151.2 mmol/l and chlorine from 112.2 to 115.6 mmol/l. All indicators corresponded to physiological norms.

Discussion and Conclusion

The inclusion of feed additives "GerbaStor" and "ProStor" in the diet did not have a negative effect on metabolic processes in the body of birds: the analyzed morphological and biochemical blood parameters were within physiological norms. It was noted that against the background of the use of

Table 3

Biochemical composition of blood of broiler chickens at the age of 29 days, M ± m (n = 5)

Indicator	Group		
	Control	1st experienced	2nd experienced
Total protein, g/l	27.32 ± 1.52	27.18 ± 1.54	28.50 ± 1.62
Albumin, g/l	10.28 ± 0.58	9.96 ± 0.71	10.28 ± 0.36
Globulins, g/l	17.04 ± 0.96	17.22 ± 0.89	18.22 ± 1.33
A/G index	0.60 ± 0.01	0.58 ± 0.02	0.57 ± 0.03
Urea, mmol/l	0.26 ± 0.06	0.26 ± 0.06	0.22 ± 0.07
Creatinine, μmol/l	23.06 ± 0.90	23.06 ± 1.23	23.34 ± 1.27
Uric acid, mmol/l	196.80 ± 27.59	144.74 ± 35.97	131.13 ± 26.67
Glucose, mmol/l	13.31 ± 0.31	13.27 ± 0.26	14.29 ± 0.33
Cholesterol, mmol/l	2.94 ± 0.23	2.75 ± 0.24	3.31 ± 0.24
Triglycerides, mmol/l	0.56 ± 0.05	0.38 ± 0.06	0.44 ± 0.08
ALAT, IU/ml	2.34 ± 0.36	2.80 ± 0.44	2.14 ± 0.39
ASAT, IU/ml	583.82 ± 88.30	648.65 ± 82.43	544.52 ± 74.18
Calcium, mmol/l	2.60 ± 0.08	2.55 ± 0.05	2.52 ± 0.06
Phosphorus, mmol/l	2.40 ± 0.07	2.73 ± 0.19	2.53 ± 0.08
Potassium, mmol/l	5.65 ± 0.51	5.56 ± 0.31	4.85 ± 0.58
Sodium, mmol/l	150.60 ± 1.04	150.40 ± 1.79	151.20 ± 0.42
Chlorine, mmol/l	114.00 ± 0.94	112.2 ± 1.02	112.80 ± 0.74

supplements containing probiotics, prebiotics and phytobiotics, an improvement in the assimilation of protein nitrogen was observed in the experimental groups, as evidenced by a decrease in uric acid in the blood serum. In addition, the use of the studied feed was accompanied by the activation of lipid metabolism in the body of birds, which is characterized by a tendency to reduce the amount of triglycerides in the blood serum in individuals of the 1st and 2nd experimental groups. The results of the study showed that when "ProStor" is introduced into the feed concentrate, there is a tendency to a decrease in the activity of transaminases in the body of broilers, which demonstrates a higher resistance of their internal organs, in particular the liver and heart, to unfavorable exogenous and endogenous influences.

All of the above served as a physiological and biochemical basis for increasing the productivity of broilers that received experimental feed preparations: the experimental chickens grew better and used feed concentrate economically.

Based on the obtained results, we recommend to include "GerbaStor" and "ProStor" feed additives in the compound feed for broiler chickens from the 5th day of growing until the end of the feeding period in the amount of 0.5 g/kg of feed concentrate.

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