# IMPACT OF POLYPROBIOTICS ON THE PRODUCTIVITY OF BROILER CHICKENS

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Probiotics are based on the normal microflora in the digestive tract of animals. They are ecological products. Their mechanism of action is the competition for nutrients and location in the digestive tract. The essence of the efficiency of probiotics lies in the stimulation of positive metabolic changes in the digestive tract of animals, improvement of absorption of nutrients, enhancement of the organism's resistance and antagonistic effect on harmful microflora. The use of probiotics in the digestive tract improves metabolic processes, increases vitality and resistance of individuals, improves the efficiency of digestion and absorption of nutrients, and intensifies various vital processes in a microorganism **(Некрасов и др.,** 2010; **Панин и Малик,** 2006**; Sengaut and Januskevicius,** 2010**)**.

Today probiotics are widely used in nutrition of people, livestock and poultry. The probiotic preparations used around the world consist of 6 to 8 or more strains of microorganisms. However, some think that the preparations and products consisting of a single strain of bacteria are better that the multicomponent ones. When being stored in a mixture a strain may begin to dominate, while others are inactivated and the number of their live cells significantly decreases. For this reason, it was suggested to use mixtures containing no more than 2–3 components. However, today's biotechnology advances allow successfully using the probiotic preparations consisting of a number of strains of microorganisms. That is why the terms "multiprobiotics" and "polyprobiotics" have appeared in the literature. Each probiotic strain discovers the most suitable conditions in the intestines and takes the microecologic niche that suits them the best, the so called biotope. Therefore, when creating probiotics the strains must be chosen after they are tested according to their symbiosis and selected by the ability to survive in adverse conditions. Isolation of strains from other taxonomic groups, which conclude a part of the normal microflora, allows the development of new probiotic complex products that complement each other by their effects, and the efficiency whereof can be further adjusted **(Ильясов и Чепуштанова,** 2010; **Коршунов,** 1995**; Крапивина и др.,**2012; **Лукашенко,** 2011; **Grajek et. al., 2005)**.

The purpose of this work is to determine the impact of the polyprobiotic on the growth and physiological condition of broiler chickens.

## MATERIAL AND METHODS

The polyprobiotic is produced in the USA according to the patented technology. The product consists of the following strains of the representatives of the natural macroorganisms of the digestive tract: *Bacillus subtilis, Bifidobacterium,*

*Enterococcus, Lactobacillus, Saccharomyces cerevisiae*. Experiments were carried out with the broiler chickens of 1 to 41 days age. The research was carried out at the poultry farm UAB "Zelve" in Lithuania, having formed two parallel groups of chickens. Both groups of birds were fed and kept under the same conditions. The only difference was that polyprobiotic preparation had been mixed into the water intended for the experimental group of broiler chickens. The experiment was conducted according to the scheme presented in the Table 1.

We calculated the daily gain based on the data of the control weights. At the end of the experiment 100 chickens from each group best corresponding to the average mass were selected, their blood was collected for the haematological test, and the control slaughter was carried out. During slaughter the samples of contents of the glandular stomach and the caecum were taken for the purpose of microbiological tests. In peripheral blood the content of haemoglobin, the amount of erythrocytes, leucocytes and hematology was measured on the automatic haematological analyser "MEDONIC CA 620" (Boule Medical AB, Sweden).

Table 1. **Experiments scheme**

| |
|---|
| Broiler chickens cross *ROSS-308* |
| Normal Ration (NR) |
| NR+polyprobiotic added into the drinking water of chickens with the ratio of 1:5000 on the 1–21 day of age |
| NR+polyprobiotic with the ratio of 1:3000 on the 22–41 day of age |

Biochemical analyzes were performed on biohromotografe POINTE-180 (Sweden).

In the serum the following parameters were measured: the content of protein and its fraction, calcium, inorganic phosphorus on the bio chromatograph POINTE-180 and spectrometer "Fluorat-02-2M" (JSC "Lumex", Russia) according to the accompanying methods.

Microbiological research of the contents of different parts of the gastrointestinal tract. Samples were stored at 4° C (8 hours). Weighed samples (about 0.3 g) were homogenized in 0.9% saline as a 10-fold dilution and further diluted 10 times in the same environment. Parts (100 micrograms) of a corresponding dilution were placed in agar cup of meat-peptone broth and meat-peptone agar containing 50 micro $ml^{-1}$ tetracycline or 15 micrograms $ml^{-1}$ fusidic acid to make a selection of corresponding groups of microorganisms. Cups were incubated anaerobically for 40 hours at 37 ° C and colonies in the cups were counted in order to determine the amount of the micro-organisms in all samples. In addition, the colonies in all agar cups were identified morphologically.

The method is based on seeding the sample on selective environment, cultivation of inoculations, counting all visible colonies of yeast, typical by macro-and microscopic morphology. In inoculations the amount of yeast (confirmed by microscopy) is counted separately, using a magnifying glass with a magnification of 4-10x. For quantification is selected a cup on which has grown from 15 to 50 colonies of yeast. The colonies are counted in each of the parallel inoculations and the arithmetic mean value of the number of colonies is found (**Solonenko et al.,** 2000). During the control slaughter the carcass output, the output of chest, legs and other muscles of the carcass were assessed. The internal and digestive organs were evaluated during the control slaughter. Muscles of chest and legs were taken for the tests of meat quality. The chemical composition and physical and chemical properties of the muscles were assessed according to the commonly accepted methods.

All results are expressed as mean and standard error of the mean (SEM). The authenticity of the difference in variables between the groups was

measured by the Student's criterion (**Sakalauskas,** 1998). Statistical differences between groups for different parameter concentrations were determined using ANOVA general linear model, (GLM). P-values 0.05 and less were considered significant.

## RESULTS AND DISCUSSION

The growth dynamics of the chickens is presented in the Table 2. The data in this table show that the broiler chickens that received the polyprobiotic product during the experiment grew faster than the analogue animals of the control group. It was stated by other authors that chicks receiving probiotic, grew faster (**Pelícia et al.,** 2004). During the experiment the broiler chickens of the experimental group gained 145 g or 5.98 percent more than their analogues in the control group. Accordingly, the average daily gain was 3.86 g or 6.02 percent larger in the experimental group compared to the one of the control group. Feed consumption per 1 kg of weight gain of the experimental control group of birds was 20 g or 1.21 percent lower than the one of the birds from the control group.

The presented results of the control slaughter (Table 3) show that the pre-slaughter weight of the experimental group of broiler chickens was 13.56 percent higher than the one of the control group of chickens. Accordingly, carcass weight was 254.4 grams or 14.69 percent and slaughter output 0.75 percent higher than the one of the control group of chickens.

According to the presented results we can observe a tendency that the polyprobiotic preparation affects the formation of the muscle tissue of separate body parts. It was showed by other researchers to carry out studies (**Denli et al., 2003; Ranade and Rajmane., 1992**). The leg muscles of the birds who had received the product developed more slowly, but the chest muscles developed faster. The quantity of the chest muscles in the carcass of the experimental group of birds during the experiment was 1.62 percent more than in the carcass of the control group.

As the research results show, the polyprobiotic preparation activates the vital processes of the birds and stimulates the development of internal organs. That, probiotics stimulate the natural resistance of the organism and activates the vital processes of the birds and other scientists was determined (**Ghadban,** 2002). The heart and liver of the experimental group of chickens were bigger than the ones of the control group. However, the length of the intestine of the chickens who received the product was shorter. It can be explained by the fact that the microorganisms strains contained in the product ensure faster feed intake and digestibility, and this affects the shortening of the intestine.

In order to explore the influence of polyprobiotic on the muscle tissue of the broilers analytical studies, the results whereof are presented in the Table 4, were carried out.

Table 4 shows that the polyprobiotic preparation improves the qualitative indicators of the muscle tissue of the broiler chickens. Less moisture is stored in the muscles of chest and legs of the chickens that received the probiotic; accordingly, there is more dry matter in their muscles. In the leg muscles of the experimental group of birds there was 0.83 percent of proteins and 29.17 percent of calcium more than in the leg muscles of the control group of birds. The fat content in the chest and leg muscles of the experimental group of chickens was higher than in the control group of chickens, respectively, 0.82 percent and 1.43 percent.

The benign microflora in the digestive tract of chickens consists of lactobacteria, bifidobacteria, enterecocci, staphylococci, escherichia and yeast. The microflora releases antimicrobial substances and prevents the growth of pathogenic microorganisms in the digestive tract, enhances the natural resistance of the digestive tract, stimulates the regeneration of the intestinal mucous after various injuries and infections, positively effects the immune system, and improves the digestibility and absorption of nutrient. The balance of normal intestinal microflora can be upset by stress, antibiot-

Table 2. **Growth dynamics of the broiler chickens**

| Age of chickens, d | Groups | | | |
|---|---|---|---|---|
| | Control, *n*=26 800 | | Experimental, *n*=26 690 | |
| | Live weight, g | Daily gain, g | Live weight, g | Daily gain, g |
| 0 | 35 ±0.01 | | 35 ±0.01 | |
| 7 | 180 ±9.5 | 20.72 ±1.22 | 198 ±12.3 | 23.29 ±1.18 |
| 14 | 431 ±35.5 | 35.86 ±2.91 | 502 ±40.2 | 43.43 ±2.24 |
| 21 | 851 ±45.8 | 60.0 ±3.12 | 943 ±53.6 | 63.0 ±3.33 |
| 28 | 1470 ±65.3 | 88.42 ±3.54 | 1580 ±49.9 | 91.0 ±4.56 |
| 35 | 2057 ±110.5 | 83.86 ±4.47 | 2199 ±120.3 | 88.43 ±4.11 |
| 41 | 2460 ±115.8 | 67.17 ±3.38 | 2605 ±130.0 | 67.67 ±3.65 |
| Average during the experiment | 2425 ±113.6 | 59.12 ±3.22 | 2570 ±124.8 | 62.68 ±3.24 |
| Feed consumption for the gain of 1 kg | 1.65 | | 1.63 | |
| Retention, % | 97.39 | | 96.99 | |

Table 3. **Results of control slaughter of broilers chickens**

| Indicators | Group | |
|---|---|---|
| | Control | Experimental |
| Carcass mass, g | 1857.8 ±33.78 | 1986.7 ±51.67 |
| Carcass output, % | 75.52 ±1.65 | 76.27 ±1.32 |
| Muscles, % (from carcass weight): | | |
| legs | 22.78 ±1.12 | 22.07 ±0.98 |
| chest | 24.82 ±1.07 | 26.44 ±0.64 |
| other | 13.30 ±1.25 | 11.63 ±1.32 |
| all muscles | 60.90 ±1.85 | 60.14 ±2.01 |
| Development of internal organs, % (from live weight): | | |
| heart | 0.56 ±0.02 | 0.63 ±0.04 |
| liver | 1.66 ±0.08 | 1.92 ±0.12 |
| glandular stomach | 0.40 ±0.02 | 0.42 ±0.02 |
| muscular stomach without content and cuticle | 1.82 ±0.12 | 1.74 ±0.95 |
| intestine with the contents | 4.82 ±0.31 | 4.89 ±0.29 |
| gall-bladder | 0.09 ±0.005 | 0.08 ±0.006 |
| Length of the intestine, cm | 180.5 ±15.7 | 163.0 ±13.0 |

Table 4. **Test results of the muscle tissue of the broiler chickens**

| Indicators | Group | |
|---|---|---|
| | Control | Experimental |
| Moisture, %: | | |
| in the chest muscles | 73.65 ±1.15 | 72.85 ±0.65 |
| in the leg muscles | 74.36 ±0.78 | 72.17 ±0.85 |
| Proteins, %: | | |
| in the chest muscles | 23.46 ±0.22 | 23.20 ±0.32 |
| in the leg muscles | 18.23 ±0.25 | 19.06 ±0.33 |
| Fat, %: | | |
| in the chest muscles | 2.11 ±0.28 | 2.93 ±0.41 |
| in the leg muscles | 6.42 ±1.21 | 7.85 ±0.95 |
| Ashes, %: | | |
| in the chest muscles | 0.94 ±0.01 | 1.01 ±0.01 |
| in the leg muscles | 1.03 ±0.02 | 1.02 ±0.01 |
| Calcium, g/kg: | | |
| in the chest muscles | 0.099 ±0.006 | 0.096 ±0.005 |
| in the leg muscles | 0.096 ±0.001 | 0.124 ±0.01 |
| Phosphorus, g/kg: | | |
| in the chest muscles | 2.42 ±0.04 | 2.46 ±0.05 |
| in the leg muscles | 2.38 ±0.03 | 2.35 ±0.04 |

Table 5. **Microflora composition of the digestive tract of chickens, CFU/g**

| Indicators | Group | |
|---|---|---|
| | Control | Experimental |
| | In glandular stomach | |
| *Lactobacteria* | $1.8 \times 10^8 \pm 0.21 \times 10^8$ | $8.0 \times 10^8 \pm 0.93 \times 10^8$ |
| *Bifidobacteria* | $10^5 \pm 10^3$ | $10^7 \pm 10^4$ |
| *Staphylococci* | $1.5 \times 10^4 \pm 0.08 \times 10^4$ | $1.7 \times 10^4 \pm 0.1 \times 10^4$ |
| *Enterococci* | $7.2 \times 10^4 \pm 0.42 \times 10^4$ | $7.1 \times 10^4 \pm 0.46 \times 10^4$ |
| Yeast | $9.4 \times 10^4 \pm 0.51 \times 10^4$ | $6.5 \times 10^5 \pm 0.69 \times 10^4$ |
| | In caecum | |
| *Lactobacteria* | $4{,}2 \times 10^9 \pm 0{,}3 \times 10^8$ | $1.4 \times 10^{10} \pm 0.8 \times 10^9$ |
| *Bifidobacteria* | $10^8 \pm 10^7$ | $10^9 \pm 10^7$ |
| *Staphylococci* | $3.1 \times 10^8 \pm 1.8 \times 10^7$ | $2.8 \times 10^8 \pm 1.7 \times 10^7$ |
| *Enterococci* | $1.8 \times 10^7 \pm x\ 10^6$ | $2.0 \times 10^7 \pm x\ 10^6$ |
| Yeast | $1.6 \times 10^9 \pm 0.9x\ 10^8$ | $1.7 \times 10^9 \pm 08 \times 10^8$ |
| Microorganisms fermenting lactates | $9.5 \times 10^7 \pm 0.7 \times 10^7$ | $3.0 \times 10^8 \pm 0.2 \times 10^9$ |
| Cellulolytic microorganisms | $2.2 \times 10^7 \pm 0.3 \times 10^7$ | $8.8 \times 10^7 \pm 1{,}5 \times 10^7$ |

Table 6. **Results of blood morphological and biochemical tests**

| Indicators | Group | |
|---|---|---|
| | Control | Experimental |
| Erythrocytes, $10^{12}$ L | 3.55 ±0.58 | 3.96 ±0.46 |
| Leucocytes, $10^{9}$ L | 25.0 ±2.50 | 26.50 ±3.36 |
| Haemoglobin, g/L | 8.95 ±2.01 | 9.72 ±1.13 |
| Protein, g/L | 46.5 ±0.33 | 46.6 ±0.51 |
| Albumins, % | 33.75 ±3.33 | 38.53 ±2.96 |
| Globulins, % | 54.61 ±4.52 | 58.74 ±5.24 |
| α-globulins, % | 16.75 ±0.58 | 17.78 ±0.36 |
| β-globulins, % | 7.89 ±0.66 | 7.91 ±0.74 |
| γ-globulins, % | 29.82 ±2.35 | 34.78 ±1.48 |
| Calcium, mg% | 11.60 ±0.23 | 12.34 ±0.34 |
| Phosphorus, mg% | 4.61 ±0.31 | 4.82 ±0.43 |

ic preparations, preservatives and other substances (**Wenk,** 2003).

The data of the Table 5 shows that the polyprobiotic preparation had a positive impact on the formation of benign microflora in the digestive tract of the broiler chickens. Our study showed that the preparation had the greatest impact on the amount of lacto- and bifidobacteria. In the glandular stomach of the experimental group of chickens there was 6.2 x$10^{8}$ or 4.45 percent of lactobacteria more than in the glandular stomach of the control group of chickens. Accordingly, there were 100 times more of bifidobacteria and 6.9 times more of yeast. In the caecum of the experimental group of chickens there was 3 times of lactobacteria and 10 times of bifidobacteria more than in the caecum of the control group of chickens. In the caecum of the experimental group of chickens there were 3.0 x $10^{8}$ microorganisms fermenting lactates, while in the control group – 9.5 x $10^{7}$ or 3.16 times less, and respectively, there were 4 times more of cellulolytic microorganisms in the caecum of the experimental group of chickens than of the control group of chickens.

The results of the haematological tests are presented in the Table 6. They show that the polyprobiotic used in the experiment had a positive effect on the birds' organism and confirmed the potential positive effect of the preparation on the previous biological indicators. The levels of erythrocytes, leucocytes and haemoglobin in the blood of tested chickens were within the normal physiological range, but in the experimental group they were higher than in the control group of chickens. The albumins in the organism are involved in the transportation of fatty acids, and regulate the concentration of cations. They help to maintain a constant pH, and regulate the level of free water in the body **(Лукашенко и др.,** 2011**)**. The albumin level was 4.78 percent higher in the experimental group of birds than the control group. γ-globulins are involved in the organism's protective function **(Dhanalakshmi at al.,**2002**)**. Our results show that there were 4.96 percent more globulins of the γ-globulin fraction in the blood of the experimental group of birds than of the control group. This shows that the preparation used in the experiment strengthens the immune system of birds and stimulates their protective response to harmful environmental effects. α and β globulins are involved in the transportation of phospholipids, hormones and vitamins in the body, and in blood clotting **(Skiba,** 2002**)**. The levels of α and β globulins in the blood of the experimental group of chickens are higher than the ones of the control group of chickens. The total levels of protein, phosphorus and calcium in the blood serum of the broiler chickens were within the normal physiological limits.

## CONCLUSIONS

Based on the results of the experiments it can be concluded that the polyprobiotic preparation activates the growth rate of boiler chickens, has positive effect on the microflora of the digestive tract and enhances the immune system. The daily gain of the broiler chickens who had received the polyprobiotic 6.02 percent, pre-slaughter weight 13.56 percent, the carcass weight 14.69 percent, the carcass output were 0.75 percent higher in comparison with the chickens who had not received the preparation.

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SUMMARY

The purpose of this work is to determine the impact of the polyprobiotic on the growth and physiological condition of broiler chickens. Experiments were carried out with the broiler chickens of 1 to 41 days age. The research was carried out at the X poultry farm in Lithuania, having formed two parallel groups of chickens. Both groups of birds were fed and kept under the same conditions. The only difference was that polyprobiotic preparation had been mixed into the water intended for the experimental group of broiler chickens. During the control slaughter the carcass output, the output of chest, legs and other muscles of the carcass were assessed. The internal and digestive organs were evaluated during the control slaughter. Muscles of chest and legs were taken for the tests of meat quality. The chemical composition and physical and chemical properties of the muscles were assessed according to the commonly accepted methods. The results obtained of the experiments showed that the polyprobiotic preparation activates the growth rate of boiler chickens, has positive effect on the microflora of the digestive tract and enhances the immune system. The daily gain of the broiler chickens who had received the polyprobiotic was 6.02 percent, pre-slaughter weight was 13.56 percent, the carcass weight was 14.69 percent, the carcass output was 0.75 percent higher in comparison with the chickens who had not received the preparation.

**Key words**: *polyprobiotics; nutrition; broiler chickens; digestive tract; growth dynamics*