

Application of a complex probiotic preparation based on *B. subtilis* and *B. licheniformis* in the technology of edible eggs' production

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Abstract. This paper highlights the study results on the usage effectiveness of the domestic complex probiotic preparation Immunoflor developed by PC KROS Farm LLC on Dekalb White egg cross chickens in the production of edible eggs. The work was carried out in the conditions of the agricultural production cooperative "Gornomariyskaya Poultry Farm" of the Republic of Mari El. During the experiment, it was found that eggs from laying hens with the highest weight and more close to the ideal shape were obtained in the 1st and 2nd experimental groups. Indicators of weight, elastic shell strain, albumen index were higher in the experimental groups relative to the control. By the end of the productive period, the yolk index was higher in the 1st and 2nd experimental groups than in the control by 0.12 and 0.7%, respectively. An increase in the albumen height and the indicator of Haugh units was noted in the eggs of the 1st and 2nd experimental groups relative to the control. It was found that egg mass loss decreased when they were stored for 14 days. Thus, the weight of eggs increased and their morphological indicators improved against the background of using a complex probiotic preparation in young poultry.

1 Introduction

Industrial poultry farming using the global gene pool of modern crosses is one of the knowledge-intensive and dynamically developing branches of the agro-industrial complex aimed at providing the population with food of its own production, in particular high-quality poultry meat and edible eggs [1, 2].

To date, the methods used in many poultry enterprises for the poultry raising do not quite correspond to the parameters and physiological needs, which, negatively affect the general condition of the body in the end. A high level of poultry population with year-round keeping in confined areas in enclosed spaces is a factor contributing to the airshed disruption in poultry facilities, which leads to a decrease in immunity and results in increased morbidity; it also reduces preservation and deteriorates productive and reproductive qualities [3].

According to scientists, the functional state of the intestine significantly affects the health of the poultry. Due to the prohibition of antimicrobial growth stimulants in many

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countries of the world, alternative solutions are urgently needed for the preventive use of feed substances that promote intestinal health. It is difficult to find an effective alternative to antibiotics, because potential remedies should be effective not only in maintaining the functional state of the intestine, but also in maintaining good indicators in poultry. Another problem is that alternative agents should be easy to use by farmers at the commercial level and should not cause the potential development of bacterial resistance [4, 5].

Probiotic preparations are a suitable alternative. Currently, more and more scientific research is focused on the search for new probiotics. Previously, it was believed that probiotics suppress pathogenic flora in the intestine due to the production of antimicrobial substances, which is the main mechanism of their action. Other probiotics' effects are directed at the immune and nervous systems and even on the DNA stability. They have a high potential to improve the health of poultry; unlike antibiotics, they have not been reported to be responsible for the development of resistance mechanisms in bacteria living in the intestine. This is due to that probiotics have several ways of action that actually affect the intestinal microbiocenosis both indirectly and directly by producing antimicrobial compounds; their advantage is that they use a competitive exclusion that prevents or reduces pathogens' colonization of the intestine [6, 7].

At the moment, the main criteria for considering a strain as a probiotic are those properties that help it survive in the gastrointestinal tract. Namely: tolerance to low pH values, tolerance to bile, assessment of the cell surface hydrophobicity. The factor that contributes to the benefits of using probiotics is that they meet the criterion of the product's practicality. The addition of probiotic preparations to poultry feed can be fully integrated into industrial practice since probiotic strains can withstand environmental stress including feed granulation, and they are able to germinate in the gastrointestinal tract of chickens when ingested [8, 9, 10].

The purpose of this work is to carry out a morphological analysis of egg quality when using a probiotic preparation based on *B. subtilis* and *B. licheniformis* strains for chickens of a commercial herd.

2 Materials and methods

The scientific and economic experiment was conducted on the basis of the agricultural production cooperative "Gornomariyskaya Poultry farm". Experimental results' processing was carried out in the conditions of the Department of Morphology, Obstetrics and Therapy and in the Laboratory of Clinical and Hematological Studies of the FSBEI HE Chuvash SAU.

To establish the feasibility of using the preparation under testing, it was introduced for the chickens of the Dekalb White egg cross. For this purpose, three groups of one-day chickens (50 heads each) were selected according to the principle of analogues. In the 1st experimental group, the preparation was given to the birds with water at a dose of 15 g/t of water at the age from the 1st to the 21st day. Similarly to the 1st experimental group, the birds of the 2nd experimental group received the preparation under testing at the rate of 15 g/t of feed as part of the main diet. No probiotic preparation was received in the control group.

Immunoflor is a complex probiotic preparation developed by PC KROS Pharm LLC (Mytishchi, Russia), which is used to improve digestion and balance the diets of animals and poultry.

The composition of this preparation includes the following components: probiotic strains – *B. subtilis*, *B. licheniformis*, *Bifidobacterium globosum*, *Enterococcus faecium*, *Saccharomyces cerevisiae* and prebiotic substances – chitosan and lactose.

Eggs' selection and storage for the study of their weight and the dynamics of its changes

were carried out under the same conditions. The selected material was stored in a refrigerator at +4 °C.

The average weight of laid eggs was determined at the end of each egg-laying month for 5 consecutive days by weighing on laboratory analytical scales Shinko AJH-620 CE with an accuracy of 0.1 g; egg shape index, % – by calculating the ratio of longitudinal and transverse diameters; albumen index, % – the ratio of the height of the dense albumen layer to its average diameter; yolk index, % – the ratio of the yolk height to its average diameter; the shell thickness – using a micrometer with an accuracy of 0.01 mm at the blunt and pointy ends and the equatorial part of the egg; the relative weight of albumen, yolk, and shell, % – by weighing on laboratory analytical scales Shinko AJH-620 CE; the height of the air cell using a template on a millimeter scale placing the "zero" of the template at the central point of the cell; the Haugh units were determined by the formula:

$$Ex = 100 \times \log(h - 1,7 \times 0,37 + 7,60)$$

where h is the height of the dense albumen, mm (measured at the highest point of the dense albumen with an altimeter with an accuracy of 0.01 mm);

M – egg weight, g;

1,7; 0,37; 7,6 – constant coefficients.

3 Results and discussion

The obtained results' analysis of morphological analysis of eggs of laying hens is presented below (Tables 1-3).

Table 1. Morphological analysis of eggs of industrial herd's chickens (19-30 weeks).

Indicator	Group		
	control	1st experimental	2nd experimental
Egg weight, g	50,6 ± 1,17	54,71 ± 0,97*	53,77 ± 0,52*
Egg shape Index, %	76,8 ± 0,59	76,4 ± 0,61	76,6 ± 0,67
Shell weight, g	6,35 ± 0,12	6,46 ± 0,13	6,41 ± 0,17
Height of air cell, mm	0,5 ± 0,16	0,4 ± 0,11	0,4 ± 0,13
Elastic shell strain, microns	19,4 ± 0,68	21,7 ± 0,39*	21,2 ± 0,31*
Albumen index, %	7,4 ± 0,17	7,9 ± 0,09*	7,7 ± 0,11
Yolk index, %	46,2 ± 0,16	47,9 ± 0,11***	47,6 ± 0,12***
Albumen height, mm	5,5 ± 0,24	5,7 ± 0,21	5,7 ± 0,23
Ratio, %:			
albumen	56,7 ± 0,93	57,1 ± 0,72	57,2 ± 0,58
yolk	30,8 ± 1,35	31,1 ± 0,97	31,1 ± 0,72
shell	12,5 ± 0,24	11,8 ± 0,23	11,7 ± 0,31
Haugh Units	76,40 ± 2,24	76,92 ± 2,97	76,86 ± 2,43

* P<0,05, ***P<0,001

It was found that the average egg weight index increased throughout the entire period of laying hens' productivity in the control, 1st and 2nd experimental groups.

The egg shape index was in the range of 76.4-76.8% at the age of 19-30 weeks, 76.1–76.7% at the age of 31-60 weeks and 76.9-77.4% at the age of 61-90 weeks. Based on the data obtained, it was found that the eggs had a shape index close to "ideal". It should be noted that the closest egg shape was observed in the 1st and 2nd experimental groups, unlike control group's birds aged 31-60 weeks – 76.1 ± 0.25 and 76.4 ± 0.34%, respectively.

Table 2. Morphological analysis of eggs of industrial herd's chickens (31-60 weeks).

Indicator	Group		
	control	1st experimental	2nd experimental
Egg weight, g	62,53 ± 0,57	64,88 ± 0,49*	64,17 ± 0,41*
Egg shape Index, %	76,7 ± 0,32	76,1 ± 0,25	76,4 ± 0,34
Shell weight, g	7,18 ± 0,11	7,54 ± 0,07*	7,35 ± 0,06*
Height of air cell, mm	0,6 ± 0,09	0,5 ± 0,09	0,6 ± 0,08
Elastic shell strain, microns	19,1 ± 0,41	20,9 ± 0,43*	20,4 ± 0,34*
Albumen index, %	7,8 ± 0,14	8,2 ± 0,11	8,1 ± 0,07
Yolk index, %	47,4 ± 0,11	48,7 ± 0,16***	48,3 ± 0,11***
Albumen height, mm	6,1 ± 0,17	6,3 ± 0,13	6,2 ± 0,21
Ratio, %:			
albumen	55,57 ± 0,64	56,29 ± 0,72	56,05 ± 0,58
yolk	32,95 ± 0,32	32,09 ± 0,41	32,50 ± 0,39
shell	11,48 ± 0,18	11,62 ± 0,11	11,45 ± 0,09
Haugh Units	76,97 ± 1,74	77,85 ± 1,43	77,21 ± 1,37

* P<0,05, ***P<0,001

It was found that with the bird's age, the shell weight increased in the control, 1st and 2nd experimental groups from 6.35 ±0.12 to 7.24±0.07 g, from 6.46±0.13 to 7.61±0.09 and from 6.41±0.17 to 7.48±0.07 g, respectively. It was noted that the shell weight index in the 1st and 2nd experimental groups was higher relative to the control during the chickens' productive period.

The height of the air cell had minor changes in all experimental groups. It was found that the air cell height was lower in the 1st and 2nd experimental groups compared to the control, but the established difference in the context of the compared groups turned out to be unreliable.

It was found that the elastic shell strain was higher in chickens of the 1st and 2nd experimental groups than in the control: at the age of 19-30 weeks - by 2.3 and 1.8 microns, 31-60 weeks - 1.8 and 1.3 microns, and 61-90 weeks – by 1.2 and 1.0 microns, respectively (P <0.05).

It was found that the albumen index was more than 7.0% in the control, 1st and 2nd experimental groups. The analyzed indicator gradually increased over the entire productive period in all experimental groups. By the end of the productive period, it was found that in the 1st and 2nd experimental groups, the albumen index exceeded that in the control by 0.4 (P<0.05) and 0.1%, respectively.

It was found that the yolk quality was high in the control, 1st and 2nd experimental groups. It was noted that by the end of the productive period, the yolk index was higher in the 1st and 2nd experimental groups than in the control by 0.12 (P<0.001) and 0.7 (P<0.05)%, respectively.

An increase in the albumen height in the productive herd's eggs of experimental groups' chickens was found for the entire experimental period with respect to control.

The indicator of Haugh units in the 1st and 2nd experimental groups was higher than in the control: by 0.52 and 0.46% – at the age of 19-30 weeks, by 0.88 and 0.24% – 31-60 weeks and by 0.56 and 0.37% – 61-90 weeks, respectively.

Table 3. Morphological analysis of eggs of industrial herd's chickens (61-90 weeks).

Indicator	Group		
	control	1st experimental	2nd experimental
Egg weight, g	64,57 ± 1,09	69,71 ± 1,11*	68,52 ± 1,03*
Egg shape Index, %	77,4 ± 0,41	76,9 ± 0,31	77,2 ± 0,28
Shell weight, g	7,24 ± 0,07	7,61 ± 0,09*	7,48 ± 0,07*
Height of air cell, mm	0,7 ± 0,13	0,6 ± 0,11	0,6 ± 0,11
Elastic shell strain, microns	18,9 ± 0,33	20,1 ± 0,34*	19,9 ± 0,27*
Albumen index, %	8,1 ± 0,11	8,5 ± 0,09*	8,2 ± 0,12
Yolk index, %	48,1 ± 0,18	49,3 ± 0,13***	48,8 ± 0,14*
Albumen height, mm	6,6 ± 0,14	6,8 ± 0,17	6,8 ± 0,11
Ratio, %:			
albumen	51,28 ± 0,84	47,88 ± 0,63	47,24 ± 0,71
yolk	37,51 ± 0,46	41,21 ± 0,38	41,84 ± 0,42
shell	11,21 ± 0,11	10,91 ± 0,13	10,92 ± 0,11
Haugh Units	79,37 ± 1,87	79,93 ± 1,12	79,74 ± 1,28

* P<0,05, ***P<0,001

The dynamics of egg mass loss during 7 and 14 days from the laying day was studied (Table 4).

Table 4. Dynamics of egg weight during storage, g

Group	Average egg weight,		
	Day 1	Day 7	Day 14
<i>chicken age 19-30 weeks</i>			
control	50,67±1,13	49,83±1,53	48,52±1,44
1st experimental	54,71±0,71*	53,89±0,82*	52,84±1,15*
2nd experimental	53,77±0,68*	52,93±1,04	51,68±1,09
<i>chicken age 31-60 weeks</i>			
control	62,53±0,57	60,34±0,47	58,76±0,72
1st experimental	64,88±0,49*	63,12±0,32**	61,44±0,61*
2nd experimental	64,17±0,41*	62,48±0,41**	60,59±0,54
<i>chicken age 61-90 weeks</i>			
control	64,57±1,09	62,16±0,38	59,98±0,41
1st experimental	69,71±1,11*	67,32±0,47***	65,91±0,78***
2nd experimental	68,52±1,03*	66,95±0,51***	64,07±0,72**

* P<0.05, ** P<0.01, *** P<0.001

It was found that the eggs' weight loss of the control, 1st and 2nd experimental groups during storage for 14 days laid by a bird aged 19-30 weeks was 2.15, 1.87 and 2.09 g, 31-60 weeks - 3.77, 3.44 and 3.58 g, 61-90 weeks – 4.59, 3.80 and 4.45 g, respectively. It should be noted that the weight loss of eggs in the 1st and 2nd experimental groups was lower than in the control.

4 Conclusion

Thus, the results of the conducted research work indicate the positive effect of the complex

probiotic preparation Immunoflor due to optimizing the digestive process and stimulating the development of a positive microbiota balance in the digestive tract. Therefore, against the background of using the preparation under testing, the weight of eggs increased, their morphological parameters improved, the loss of egg mass decreased when they were stored for 14 days, which determines the quality of food eggs; tested preparation's admission with water gave a greater positive effect than when it was introduced into the feed.

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