

A FEED ADDITIVE BASED ON LACTOBACILLI WITH ACTIVITY AGAINST CAMPYLOBACTER FOR MEAT-BREEDING CHICKENS PARENT FLOCK

Anna Balykina^{1*}, Elena Kapitonova², Ilya Nikonov³,
Yuri Kuznetsov⁴, Sergey Shlykov^{5*}

¹ Department of Biochemistry and Physiology, St. Petersburg State University of Veterinary Medicine, RUSSIA.

² Department of Private Livestock, Vitebsk State Awarded the «Badge of Honour» Order Veterinary Medicine Academy, BELARUS.

³ Research part "Agrotechnopolis", Perm State Agro-Technological University named after Academician D.N. Pryanishnikov, RUSSIA.

⁴ Department of Parasitology, St. Petersburg State University of Veterinary Medicine, RUSSIA

⁵ Department of Technology of Production and Processing of Agricultural Products, Stavropol State Agrarian University, RUSSIA.

ARTICLE INFO

Article history:

Received 04 June 2020
Received in revised form 07
September 2020
Accepted 18 September 2020
Available online 24 September
2020

Keywords:

Heterologous lactobacilli;
Parent stock of chickens;
Poultry feeding;
Probiotics; Parent hens;
Campylobacter;
Lactobacillus Plantarum.

ABSTRACT

This article presents the results of feeding an experimental group of laying hens for 1-14 days with a complete feed containing heterologous lactobacilli as a feed additive. Data were obtained on the sowing rate of target lactobacilli from poultry manure and eggs. The eggs were collected for the production of broiler chickens. Using a feed additive based on heterologous lactobacilli contributed to increasing egg production per average hen by 7.8-10.0 %, and egg-laying intensity by 5.7-7.3%. The addition of the additive had a zootechnical effect even after a single application. At the same time, the probiotic supplement showed the antagonistic activity of Lactobacillus Plantarum to Campylobacter jejuni. Thus, the introduction of a feed additive based on lactobacilli, in general, had a positive effect on the productivity of the parent hens.

Disciplinary: Agricultural and Animal Sciences, Bioscience, Biology.

©2020 INT TRANS J ENG MANAG SCI TECH.

1 INTRODUCTION

The fundamental and applied foundations of the creation and use of probiotics, metabiotics, and probiotic feed additives for veterinary medicine were laid by Russian scientists' works. Campylobacter belongs to the Proteobacteria phylum, whose representatives can cause necrotizing enterocolitis in both a newborn baby and a hatched chick.

The study of the microbial community's ecology of the gastrointestinal tract of agricultural poultry has now acquired enormous fundamental importance [1, 2, 3]. According to recently obtained data, the gastrointestinal tract's microbiota participates in stimulating the digestive system's postnatal maturation, the formation of innate immunity, and colonization resistance of the digestive system. It takes part in embryogenesis [4, 5].

It has been established that the microflora of the laying hen plays a crucial role in the formation of the pathogenic microbiota of the gastrointestinal tract of the egg embryo [5, 7]. The spectrum of microorganisms capable of causing the death of embryos is diverse. The main ones are *E. coli*, *Pseudomonas* sp., *Staphylococcus* sp., *Klebsiella* sp. [4, 5, 6]. Microorganisms present in the embryo's digestive tract are the basis that determines the formation of the initial intestinal biocenosis of hatched chickens [6].

The study aimed to feed the parent herd of meat-breed chickens for 1-14 days with a full-fledged compound feed, containing heterologous lactobacilli with anti-*C. jejuni* activity [11] as a feed additive. The tasks were to analyze sowing target lactobacilli from poultry droppings and eggs.

2 MATERIALS AND METHODS

For solving this problem, the experiment was in a vivarium on layers of a parent flock of the "Cobb 500" cross hens. For the study, laying hens were selected according to analogs' principle by age (205-207 days) [8]. The experimental design is in Table 1.

Table 1: Scheme of the experiment on laying hens of the parent flock

Group no.	Number of heads	Poultry feeding features
G1 - control	40	Basic ration (BR)
G2 - experiment	40	BR + filler from feed additive (feeding for 14 days)
G3- experiment	40	BR + feed additive (1 day)
G4 - experiment	40	BR + feed additive (feeding for 14 days)

Experiments on laying hens were in the vivarium of the International Laboratory of Molecular Genetics and Poultry Genomics (Federal State Budgetary Educational Institution of Higher Education "Moscow State Academy of Veterinary Medicine and Biotechnology - MBA named after K. I. Skryabin"). The diet of the parent flock hens was according to the recommendations of VNITIP [7].

A feed additive containing a composition of heterologous lactobacilli was introducing into the chickens' diet for one day and 14 days. Experimental group 3 was introduced into the diet with a filler from an experimental feed additive [9, 10].

During the experiment, the studied indicators include

1. Egg production for the initial, middle, and final hens;
2. Safety of laying hens;
3. Intensity of egg-laying;
4. Weight of laid eggs;
5. Number of eggs with a detected defect (broken and notched);
6. Feed consumption for one head.

Sowing lactic acid bacteria to assess the number was carried out on agar nutrient media MRS at 37°C for 24 hours. Sterile skim milk was used to maintain the working cultures of lactobacilli. Cultivation of lactic acid bacteria was at 37°C under microaerophilic conditions using a desiccator

and a gas generating bag system.

3 RESULT AND DISCUSSION

Table 2 gives the main results of zootechnical accounting when growing laying hens.

Table 2: The indicators of the laying hens' parent flock productivity during the experiment period

Indicators	Groups			
	G1	G2	G3	G4
Safety, %	100	100	100	100
Egg production per initial hen, pcs/head	20.3	20.5	20.4	20.5
Egg production per final hen, pcs/head	22.1	23.7	24.1	24.2

As can be seen from the presented indicators in Table 2, the egg production per initial hen was practically the same in all experimental poultry houses. It varied within 20.3-20.5 pieces/bird, which was less than 1.0%.

All individual cages were with appropriate conditions for growing poultry. Into them maintained the normative parameters of the microclimate (temperature, humidity, air velocity, and microbial contamination) and technological requirements for ensuring the keeping of the livestock (feeding time, drinking time, light regime). Nevertheless, by the end of the experiment period, the bird showed different productivity results.

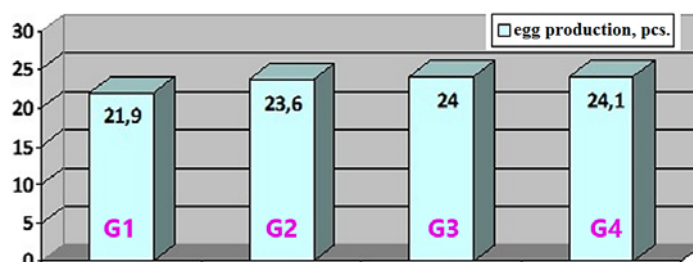


Figure 1: Egg production for an average hen, (number of eggs).

From the calculated indicators presented in Figure 1, it can see that during scientific research, egg production per average hen in the 1st group was 21.9 pcs. In the G2 group, the productivity of layers increased by 1.7 pieces. (+7.8%), compared to control. In the G3 group, egg production increased on average by 2.1 eggs/head. (+9.6%), compared to the productivity of G1 chickens. In the G4 group, egg production per average hen was 24.1 pieces, which was higher than the control achievements by 2.2 pieces. (+10.0%).

Analyzing edible eggs' production by experimental laying hens, we note once again that there are no significant differences in the productivity of hens of the 3rd and 4th groups.

Feed costs for growing layers of parent stock are in Table 3.

Table 3: Calculation of feed costs.

Indicator	Groups			
	G1	G2	G3	G4
One head, g/day	109	110	109	110

As shown in Table 3, the daily feed consumption per head in the experimental laying hens was

practically the same, with a slight balancing of 1 g. Figure 2 shows the egg-laying intensity of the test of laying hens.

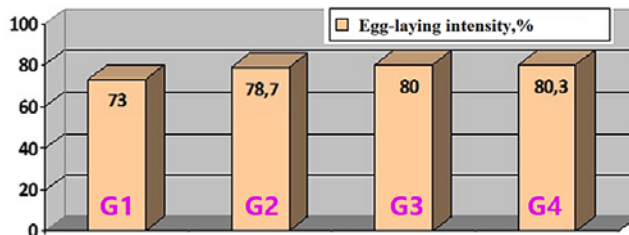


Figure 2: Intensity of egg-laying experimental laying hens (%).

Figure 2, the intensity of egg-laying in the control group was 73.0%. In the G2, the egg-laying intensity increased by 5.7 pp (percentage point), compared with the control and amounted to 78.7%. In the G3 experimental group, the egg-laying intensity indicator increased by 17.0 pp. In the G4 group, the egg-laying intensity reached its leading hands 80.3%, which was 7.3 pp higher than the control poultry house's achievements. However, the difference between the G3 and G4 groups was not significant. Thus, using a feed additive based on heterologous lactobacilli contributed to an increase in egg production per average hen by 7.8-10.0%, and egg-laying intensity 5.7-7.3 pp.

The addition of the additive had a zootechnical effect even after a single application. Prolongation of administering the probiotic supplement up to 14 days did not lead to significant differences between G3 and G4. Simultaneously, discrepancies were between the groups receiving the feed supplement with bacteria and the group receiving the filler without bacteria.

Figure 3 shows the *Lactobacillus Plantarum* selected from litter samples growing on the surface of the MRS agar medium. Simultaneously, Figure 4 notes a high antagonistic activity of *Lactobacillus Plantarum* to *Campylobacter jejuni*. Table 4 shows the data on the injection of lactic acid bacteria by classical microbiology methods.

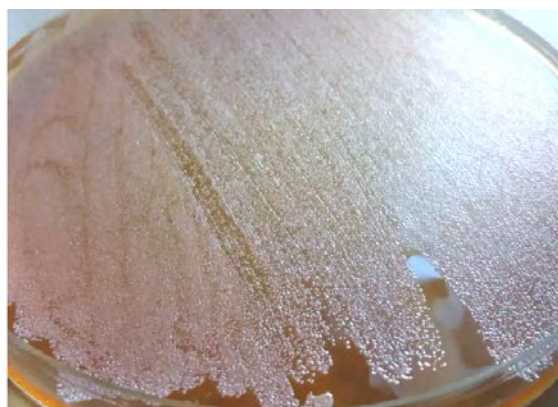


Figure 3: Sowing of *Lactobacillus Plantarum* on a nutrient medium.

Table 4: Assessment of seeding of lactobacilli from litter in layers of parent flock

Age	Groups (CFU/g)			
	G1 control group	G2 test group	G3 test group	G4 test group
1 day	$19.13 \cdot 10^6 +$ $0.40 \cdot 10^6$	$19.13 \cdot 10^6 +$ $0.40 \cdot 10^6$	$19.13 \cdot 10^6 +$ $0.40 \cdot 10^6$	$19.13 \cdot 10^6 +$ $0.40 \cdot 10^6$
7 days	$25.07 \cdot 10^7 +$ $0.60 \cdot 10^7$	$32.19 \cdot 10^8 +$ $0.50 \cdot 10^7$	$36.20 \cdot 10^7 +$ $0.70 \cdot 10^7$	$41.27 \cdot 10^9 +$ $0.30 \cdot 10^9$
14 days	$31.16 \cdot 10^8 +$ $0.80 \cdot 10^8$	$35.20 \cdot 10^8 +$ $0.60 \cdot 10^8$	$38.39 \cdot 10^8 +$ $0.60 \cdot 10^8$	$45.32 \cdot 10^9 +$ $0.50 \cdot 10^9$
22 days	$36.10 \cdot 10^5 +$ $0.80 \cdot 10^7$	$40.38 \cdot 10^8 +$ $0.70 \cdot 10^8$	$42.44 \cdot 10^9 +$ $0.60 \cdot 10^9$	$48.56 \cdot 10^{10} +$ $0.50 \cdot 10^{10}$



Figure 4: Growth of *Campylobacter jejuni* after 72 hours of culture

The introduction of a feed additive containing lactobacilli has a positive effect on the intestinal microbiocenosis of layers. It is stimulating the colonization of lactic acid bacteria from 7 days of age and colonization rate lactobacilli at an earlier date as well as reducing the content of opportunistic microflora.

4 CONCLUSION

Thus, the introduction of a feed additive containing lactobacilli has a positive effect on poultry's productivity. The use of heterologous lactic acid bacteria with antagonistic activity to *Campylobacter* normalizes the colon microbiota of hens, stimulating the colonization of lactic acid bacteria from 7 days of age, accelerates the rate of colonization by lactobacilli at an earlier date, and reduces the content of opportunistic microflora.

5 AVAILABILITY OF DATA AND MATERIAL

Data can be made available by contacting the corresponding author.

6 ACKNOWLEDGEMENT

The study was performed at the FGBOU VO "St. Petersburg State Academy of Veterinary Medicine» with the aid of the Russian Science Foundation Grant (Project No. 18-76-10017).

7 REFERENCES

- [1] Park, S. H., Lee, S. I., & Ricke, S. C. (2016). Microbial populations in naked neck chicken ceca raised on pasture flock fed with commercial yeast cell wall prebiotics via an Illumina MiSeq platform. *PLoS One*, 11(3), e0151944.
- [2] Saxena, S., Saxena, V. K., Tomar, S., Sapkota, D., & Gonmei, G. (2016). Characterisation of caecum and crop microbiota of Indian indigenous chicken targeting multiple hypervariable regions within 16S rRNA gene. *British poultry science*, 57(3), 381-389.
- [3] Oakley B.B. and Kogut M.H. (2016). Spatial and Temporal Changes in the Broiler Chicken Cecal and Fecal Microbiome and Correlations of Bacterial Taxa with Cytokine Gene Expression. *Frontiers in Veterinary Science*, 3(11).

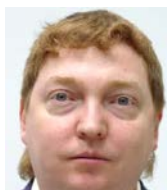
- [4] Babaca Z. (2014). Isolation of Bacterial Pathogens from Dead in Shell Chicken Embryos from Local Hatcheries. *Journal of Veterinary Science and Technology*, 5(2), 170–171.
- [5] Rossi D.A., Fonseca B.B., de Melo R.T., da Silva Felipe G., da Silva P.L., Mendon E.P., Filgueiras A.L., Beletti M.E. (2012). Transmission of *Campylobacter Coliin* Chicken Embryos. *Brazilian Journal of Microbiology*, 43(2), 535–543.
- [6] Ilina L.A., Yildirim E.A., Nikonov I.N., Filippova V.A., Laptev G.Y., Novikova N.I., Grozina A.A., Lenkova T.N., Manukyan V.A., Egorov I.A., Fisinin V.I. (2016). Metagenomic Bacterial Community Profiles of Chicken Embryo Gastrointestinal Tract by Using T_RFLP Analysis. *Doklady Akademii Nauk*, 466(4), 482–486.
- [7] Fisinin V.I. (2000). *Feeding poultry*. Sergiev Posad, 375p.
- [8] Egorov I.A., Manukyan V.A., Okolelova T.M., Lenkova T.N., Andrianova E.A. (2019). *Guidelines for feeding poultry*. Sergiev Posad: VNITIP, 226p.
- [9] Ponomarenko Yu.A., Fisinin V.I., Egorov I.A. (2012). Safety of Feed, Feed Additives, and Food. *Minsk. Ecological perspective*, 864.
- [10] RFC-HER. (2010). *Guidelines for the sanitary and epidemiological assessment of the safety and functional potential of probiotic microorganisms used for food production*. Federal Center for Hygiene and Epidemiology of Rospotrebnadzor, MU 2.3.2.2789-10, 93p.
- [11] Kuznetsov, Y., Nikonov, I., Kapitonova, E., Kuznetsova, N., Omarov, R. (2020). Evaluation Lactic Acid Bacteria Autostrains with Anti-*Campylobacter Jejuni* Activity on Broiler Chickens Productivity. *International Transaction Journal of Engineering, Management, & Applied Sciences & Technologies*, 11(15), 11A15S, 1-5. <http://doi.org/10.14456/ITJEMAST.2020.307>



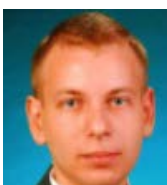
Anna Balykina is an Associate Professor at the Department of Biochemistry and Physiology, the St. Petersburg State University of Veterinary Medicine. She is a Candidate of Veterinary Sciences, Her research interests pertain to the Development of Biologically Active Feed Additives, Feeding Farm Animals (Birds).



Elena Kapitonova is an Associate Professor at the Department of Private Livestock, Vitebsk Order of the Badge of Honor State Academy of Veterinary Medicine. She is a Candidate of Agricultural Sciences Her research interests are the Development of Feed Additives, the Technology of Raising Animals (Birds), Feeding Farm Birds.



Ilya Nikonov is Deputy Director for Science and Development, BIOTROF LLC. Senior Researcher of the Scientific and Technical Information Department of the All-Russian Scientific Research Veterinary Institute of Poultry (VNIVIP), St. Petersburg. His research deals with the study of the intestinal microflora of poultry, Feed, Habitat. He is a developer of the composition and methods for assessing the Effect of Antimicrobial Feed Additives (Probiotics, Prebiotics, Phytobiotics, etc.) and Mycotoxin Sorbents.



Yuri Kuznetsov is an Associate Professor. He is a Candidate of Veterinary Sciences. His research includes Veterinary Immunology and Stress Pharmacology, Increasing the Effectiveness of Drugs, and studying the Side Effects of Drugs and Reducing their Negative Impact on the Body.



Professor Dr. Sergei Shlykov works at the Department of Technology of Production and Processing of Agricultural Products, Stavropol State Agrarian University, Russia. He holds a Doctor of Biological Science. His research focuses on Meat, Beef, Animal Products and Meat Products.