UDC 60:612.017.11/.12:636.5.034.087.7

INFLUENCE OF PHYTOADDITIVES AND SODIUM SELENITE ON INDEX OF NATURAL RESISTANCE OF LAYING HENS

Antonenko P. P. [1], Kovaleva I. V. [2], Pushkar T. D. [3], Timchenko O. V. [2]

[1] Dnipro State Agrarian and Economic University, Dnipro, Ukraine, e-mail: antonenko1946@i.ua
[2] Odessa Branch of the State Research Institute of Laboratory Diagnostics and Veterinary and Sanitary Expertise, Odessa, Ukraine, e-mail: kiv3kiv3@i.ua
[3] Odessa State Agrarian University, Odessa, Ukraine, e-mail: t_pushkar@ukr.net

Summary. The results of the complex influence of phytoadditives and sodium selenite on the parameters of the natural resistance of laying hens in the intensive fertility period are presented, as well as the determination of the total bacterial contamination and contamination of phytoadditives 'Phytopank' and 'Phytochol' and sodium selenite with molds. Increase of the total protein content in serum blood of laying hens by 13.7% and protein albumin fractions by 12.8% ($p < 0.01$), the α-globulin fraction by 11.3%, the γ-globulin fraction by 2.2% and the β-globulin fraction by 4.5%. Moreover, increase in the level of bactericidal activity of blood serum (BASK) by 16.62% ($p < 0.01$) and lysozyme activity of blood serum (LASK) by 16.7% ($p < 0.01$), and improvement of metabolism, liver function, feed consumption and assimilation of its nutrients. Determination of total bacterial contamination and contamination fungi showed that phytoadditives and sodium selenite do not exceed the overall contamination rate for one dose.

Keywords: blood, total protein, albumin, globulins, bacterial contamination, mold mushrooms, BASK, LASK, chicken bear, sodium selenite, Phytopank, Phytochol

Introduction. The intensive development of poultry farming in Ukraine, both on an industrial basis and in private farms, makes it possible to obtain high quality products in a short time. It is known, that the increase in the productivity of laying hens in turn is accompanied by an increase in physiological stresses on the body and an increase in requirements for environmental factors, in particular the conditions of housing and feeding. Violation of the optimal parameters of the microclimate causes decrease of resistance and productivity (Babina, 1998; Bashkirov, 2008).

Recently, one of the priority directions of the intensification of poultry farming was the search for highly effective ways to increase the productivity of poultry through the by using different biologically active substances (BAS), which have antimicrobial and growth stimulating qualities, but are not harmful to humans and animals (Bazylev, 2002; Draganov, Makartsev and Tyurkina, 2008; Bol'shakova, 2011).

In this direction at the modern level perspective is the use of biologically active substances, particular, preparations of herbal origin, which affect the organism in the form of a complex of minerals, vitamins, etc. When phytoadditives gets into the body, they penetrate the tissues, have a positive effect on the level of intracellular metabolism (Antonenko et al., 2014). Such influenced phytoadditives 'Phytopank' and 'Phytochol' (Kosenko and Malik, 2001).

In most countries of the world, research on the search for new sources of mineral-vitamin supplements, the improvement of their feeding technologies, and the refinement of the need for poultry in trace elements, which have not been taken into account earlier, are being actively carried out, but have proven to have a significant impact on the body. To such microelements of interest to scientists, include selenium (Sobolev and Povoznikov, 2015).

One of the promising directions stimulation of natural resistance, ensuring the normal functioning of the immune system, improving the metabolism of the organism laying hens, and increasing the biological value of fodder in intensive poultry farming is the use of various feed additives including of plant origin and trace elements (Okolelova, 2006; Glaskovich, Shul'ga and Sadomov, 2010).

The purpose of the research was to determine the effect of 'Phytopank', 'Phytochol', and sodium selenite on the indices of the natural resistance in laying hens during the period intensive fertility and to determine their total bacterial contamination and contamination with mold fungi.

Materials and methods. The research was carried out on the laying hens of the Adler Silver breed under the conditions of private farming LLC 'TAGR' (Bilyayivka Raion, Odessa Oblast). Based on the principle of par-analogue was formed three experimental groups and control group (60 in each). The conditions of feeding and breeding of laying hens of all groups met all veterinary and sanitary norms. The poultry in the control and experimental groups were kept in two-tiered cell batteries, equipped with feeders and jets, density of planting in accordance with the normative instructions. Feeding was carried out with a complete feed.

Laying hens of the 1st experimental group in addition to the main ration received a trace element of selenium in a dose of 0.2 mg/kg of dry matter of mixed fodder. As a

source of selenium, selenite sodium (sodium selenite TU 6-09-1315-76) was used. The inorganic mixture of selenium in the form of a white powder was thoroughly mixed with the feed.

The laying hens of the 2nd experimental group was given complete feed and phytoadditives 'Phytopank' and 'Phytochol' as follows: daily doses of water were injected everyday of 6 ml of each preparation (2 drops per chicken). 'Phytopank' is a complex composition of seven infusions with 40% ethyl alcohol of individual medicinal plants in a ratio (rhubarb root, rooster cockroach root, decoction root, trichophytic foxtail leaves, fragrant fennel, leaves of medicinal sage and fruits of the *Conium maculatum*). The 'Phytochol' is an aqueous extract of medicinal plants, which contains grass of shepherd's purse, flowers of sand caraway, flowers of common tansy, magnesium sulfate, sodium salicylate, hexamethylenetetramine, tincture of peppermint leaves, tincture of valerian root, belladonna and glycerin (in doses that do not exceed those generally accepted for use in homeopathy and allowed pharmacopoeia) (Antonenko et al., 2014). Phytomedications were added to the water.

The 3rd experimental group, along with the main diet and conditions of retention, received phytomedications ('Phytopank' and 'Phytochol') in combination with sodium selenite at similar doses.

Blood samples were collected from the laying hens in vivo from the subclavian vein, with all the rules of asepsis and antiseptics. The following indicators were taken into account: total protein (biuret reaction), protein fractions of blood serum (electrophoresis on paper), Serum creatinine (the kinetic method of IFCC (KF 2.7.3.2)) (Horiachkovskyi, 2005). Lysozyme activity of blood serum (LASK) was determined, using the photoelectrocholmometric method (A. G. Dorefaychuk), with a change in the temperature regimen of the reaction of blood serum of laying hens with culture of *Micrococcus lysodeikticus*; bactericidal activity serum of blood (BASB) was determined by method of Michelle Teffer in modification of O. V. Smirnova and T. A. Kuzmina (Horiachkovskyi, 2005).

During the organization of scientific research, the principles of bioethics were observed in accordance with the requirements of the European Convention for the Protection of Experimental Animals (EC 86/609/EEC) (CEC, 1986).

Determination of total bacterial contamination and contamination with moldy mushrooms performed using the method number 3 (testing of non-injectable preparations intended for introduction into animals with drinking water and/or aerosol) according to DSTU 4483:2005 'Veterinary immunobiological preparations. Methods of determination of bacterial and fungal contamination' (DSSU, 2005). Bacterial sowing of investigated drugs at a determined total bacterial contamination were carried out on meat-peptone agar (the manufacturer of the drug — Himedia, India, No. M 001) at a temperature of 35 ± 1 °C for 7 days. Determination of contamination by mold mushrooms using superficial cropping on a dense nutrient medium Saburo with glucose (Himedia, India, No. M 063) at the incubation temperature of 24 ± 1 °C for 14 days. The incubation was maintained in the HT-3/40 thermostat. At the end of the cultivation period, the results of the research were recorded visually, and the average number of increased colonies per dose was recounted.

The research results are presented in accordance with the requirements of the International System of Units and statistically processed using the MS Excel. The probability of differences between the indicators was estimated according to Student's criterion.

Results and discussions. The total protein content in the blood serum is one of the most important indicators of the quality of protein feeding of farm birds. According to the results of the research on the Adler's silver breeders, it should be noted that the usage of sodium selenite, 'Phytopank' and 'Phytochol' as supplements positively affects on metabolism, including protein metabolism and enzymatic processes (Fig. 1).

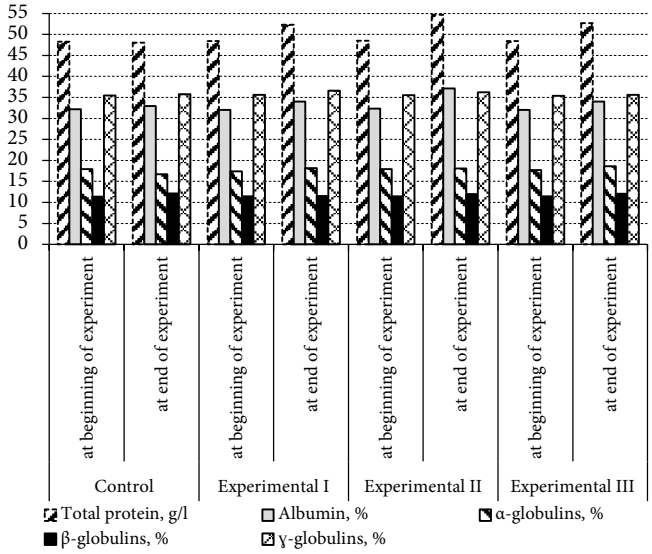

Figure 1. Indicators of protein metabolism in laying hens

The analysis of the results of the research at the end of the experiment shows that when used for feeding laying hens of the first experimental group of sodium selenite, the content of total protein increased by 8.9% compared with the control group. At the end of the experiment, in the laying hens of the second experimental group, which received feed phytomedications 'Phytopank' and 'Phytochol', there was an increase in the total protein content by 13.7% compared with the control group. A similar situation was observed in the laying hens of the third experimental group receiving sodium selenite in combination with phytomedications, in particular, the total protein content increased by 9.8%.

According to other researchers, it has been proved that the total protein correlates with the performance indicators in the laying hens, as well as a marker for the absence of chronic diseases, accompanied by a negative nitrogen balance. In addition, the content of total protein and its fractions reflects the activity of synthetic liver function.

Statistically significant increases in the albumin fraction by 3.2% were observed in the laying hens of the first experimental group under conditions of feeding with sodium selenite In the laying hens of the second and third experimental groups, when applying phytoadditives separately and simultaneously with the addition of sodium selenite in combination with feed phytoadditives, an increase in the albumin fraction was noted by 12.8% ($p < 0,001$) and 3.3% respectively. This fact can be explained by the fact that there is an improvement in metabolism and enzymatic processes, in particular liver with the help BAS, macro- and trace elements, vitamins, etc., which are part of feed phytoadditives. In addition, the increase in α-globulins in the first experimental group was determined by 8.6%, in the second by 8.1% and by the third by 11.3%, and γ-globulins by the first by 2.2%, the second by 1.2%, the third by 0.5%.

Concerning the β-globulin fraction, there was a slight decrease in the laying hens in all experimental groups from 0.3% to 4.5% in compared with the control group, but all parameters fluctuated within the limits of physiological norms. This is probably due to the fact that most beta-globulins are transport proteins, and they are involved in the transfer of hormones, vitamins, minerals and other substances, including those that are part of phytoadditives, which was confirmed after performing the research.

The great attention is paid to the humoral factors of protection (BASK, LASK), when studying the support of the level of natural resistance of the organism to the functional state of poultry and the persistence of diseases (Table 1).

Table 1 — The indicators of natural resistance of laying hens (M ± m, n = 6)

Indicators	Control group	Experimental groups		
		I	II	III
LASK, %	25.36 ± 0.51	27.9 ± 0.99*	30.45 ± 1.24**	28.89 ± 0.95**
BASK, %	47.71 ± 1.17	51.74 ± 2.72	57.22 ± 2.24**	54.7 ± 1.16**

Notes: * — $p < 0.05$; ** — $p < 0.01$ compared to the control group.

The analysis of the results showed that the combined usage of phytoadditives 'Phytopank' and 'Phytochol' and sodium selenite for the laying hens is to the improvement of overall resistance, as evidenced by their BASK, LASK and total albumin levels. With the use of sodium selenite in the feeding of laying hens of the first experimental group, the level of lysozyme activity of blood serum increased by 9.1% ($p < 0.05$), while in the second experimental group the increasing of the serum lysozyme activity by 16.7% ($p < 0.01$) compared with the control group. The same situation was observed in the laying hens in the third experimental group, where sodium selenite and phytoadditives added during feeding. In this group, the level of lysozyme activity of blood serum increased by 12.2%.

There was no statistically significant increase in the bactericidal activity of the serum of 7.7% in the laying hens of the first experimental group under conditions of feeding with sodium selenite. In the laying hens of the second and third experimental groups, when using phytoadditives separately and simultaneously with the addition of sodium selenite in combination with feed phytoadditives, a statistically significant increase in serum blood bactericidal activity was observed at 16.62% ($p < 0.01$) and 12.8% respectively. Furthermore, in the second experimental group (which was added only phytoadditives) of total protein was an increase of 13.7% and protein fractions by 12.8 albumins ($p < 0.01$), α-globulins — 11.3%, γ-globulin — 2.2% and β-globulin — 4.5%.

This can be explained with the correlation of the total albumin and its fractions with vitals, as well as marker of the absence of chronic diseases and reflection the activity of the synthetic function of the liver. It is known that most β-globulins are transport proteins that participate in the transport of hormones, vitamins and other BAR to cells, and -globulins support the overall resistance of the organism, which is confirmed by the results of research (see Fig. 1).

For non-inoculated phytoadditives such as 'Phytochol', 'Phytopank' and sodium selenite, which are recommended to use in poultry farming, a small number of bacterial and/or non-pathogenic fungal cells no more than one colony forming unit (CFU) for one dose is acceptable (Table 2).

Table 2 — Tests of non-injectable drugs intended to be administered to laying hens

Indicators	Phytopank	Phytochol	Sodium selenite
Total bacterial contamination, CFU/dose	< 1	< 1	< 1
Contamination with mold mushrooms, CFU/dose	< 1	< 1	< 1

Analysis of the results of the research show that sodium selenite and phytoadditives 'Phytopank' and 'Phytochol',

which were used during experiments for laying hens, have a total bacterial contamination and mushroom contamination of less than one, which is, the average number of non-pathogenic bacteria and (or) fungi per one dose and not exceed the permissible norm. This indicates that preparations of sodium selenite and phytoadditives 'Phytopank' and 'Phytochol' are sterile, which meets the requirements and can be used in domestic poultry farming for the intended purpose.

Conclusions. 1. The complex application of phytoadditives 'Phytopank', 'Phytochol' and sodium selenite for laying hens during period of intensive feeding had positive affect on the studied indicators of natural resistance, which was confirmed by the increase per 13.7% of total albumin and albumin fractions (albumin raise by 12.8% ($p < 0.001$), α-globulins by 11.3%, γ-globulins by 2.2%, β-globulins by 4.5%); the level of BASK raised by 16.62% ($p < 0.01$) and the level of LASK raised by 16.7% ($p < 0.01$). In addition, there was an improvement in metabolism, liver function, feed intake and the absorption of its nutrients.

2. Determination of the total bacterial contamination and contamination of mold fungi of feed phytoadditives 'Phytopank' and 'Phytochol' and sodium selenite showed that it does not exceed the permissible dose rate per dose.

The prospects for further researches is to study the influence of feed phytoadditives and sodium selenite concentration of carotenoids and vitamins in poultry eggs during the intensive productivity of laying hens under the influence of anthropogenic impact.

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