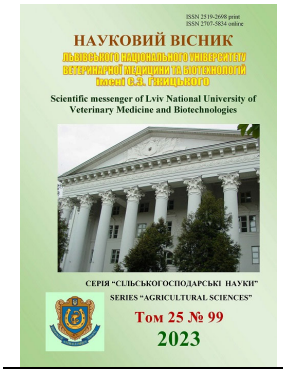

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Functional state and protein-synthesizing function of the liver of laying hens under conditions of cadmium loading

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The toxicity of cadmium in the bird's body depends on the penetration method, the total dose, and the duration of poisoning. That is why the work aimed to study the effect of Cadmium on liver function disorders. To achieve the set goal in experiments on chickens under cadmium load, it was necessary to solve the following tasks: to study the effect of cadmium on the protein synthesis function of the liver of laying hens; to study the effect of cadmium on the functional state of the liver of laying hens. 24 laying hens aged 78 weeks were selected for research. These chickens were divided into three groups: control and two experimental, taking into account their age and weight. Chickens from the control group received compound feed and clean water without adding cadmium sulfate. In the chickens of the experimental groups, cadmium sulfate was added to the drinking water in different concentrations for 30 days: the first group (R1) – 2 mg per kilogram of body weight, the second group (R2) – 4 mg per kilogram of body weight. According to the conducted studies, it was found that in laying hens under conditions of cadmium load, the functional state of the liver is disturbed, as evidenced by the increased activity of aminotransferases in their blood serum. It is worth noting that the highest activity of alanine and aspartate aminotransferases was in the blood serum of laying hens of the second experimental group on the 21st day of the experiment. The results indicate an increase in destructive processes in the body of laying hens due to cadmium loading. In laying hens under cadmium load, the liver's protein synthesis function is suppressed, manifested by a decrease in the level of total protein and albumins, with a simultaneous increase in the level of globulins. Drinking cadmium sulfate with water in a larger dose (4 mg/kg of body weight) was accompanied by a more probable decrease in total protein and albumin level than drinking cadmium sulfate in a smaller dose (2 mg/kg of body weight). Under cadmium load, an albumin-globulin disproportion occurs, which is indicated by the value of the A/G ratio. In the chickens of the first experimental group, it was found that the value of the A/G ratio on the 21st day of the experiment was 0.42, while in the second experimental group, it was lower and was 0.39, respectively, against the control 0.52.

Key words: *heavy metals, cadmium, poultry, toxicosis, protein, aminotransferases.*

Introduction

It is known that all environmental factors and conditions affect the health of animals and birds. Today, the natural environment is changing faster than the adaptability of animals, which hurts their health. The anthropogenic burden of urban and rural areas today is characterized by multicomponent chemical pollution of the environment (atmospheric air, natural waters, and soils) (Bashchenko et al., 2020; Nefodova et al., 2021; Kolosova & Shatorna, 2022).

Recently, a significant amount of data has been collected, confirming the influence of the composition of the environment on the elemental composition of living or-

ganisms. Several scientific studies have demonstrated the threat to human and animal health due to the growing pollution of heavy metals in the environment. In addition, their concentration in soil, water, and air is related to their content in the biological environments of humans and animals, which may indicate possible poisoning (Ostapiuk et al., 2018; Zemlianyi et al., 2021; Kolosova et al., 2021; Gutyj et al., 2022, 2023).

Cadmium compounds deserve special attention from researchers, as this element belongs to the first class of environmental hazards. It is considered a dangerous pollutant of the environment since its toxic effect can lead to various disorders in the functioning of animal and bird organisms. With small doses, this element is deposited in organs and tissues for a long time, causing the possibility of developing toxicosis and violations of biochemical processes, structure, and functions of cells. Cadmium toxicity depends on its form, solubility, and interaction with other active substances. The biological reaction to this toxicant also depends on the age, sex, and general condition of the animal and bird organism (Lavryshyn et al., 2020; Nazaruk et al., 2021; Ostapyuk et al., 2021, 2023).

It should be noted that the circulatory system is the most sensitive to the influence of environmental factors on the animal body; it is one of the first to respond to changes in animal feeding, and even more so to changes in the macro-, microelement, and vitamin supply of the body. Among the changes that occur in the peripheral blood of animals, the first and most convenient for detecting the toxic effect of heavy metals are changes in their morphological and biochemical composition (Slobodian et al., 2019, 2020, 2021, 2022).

The regularities of changes in the functional state and protein-synthesizing function of the liver of laying hens under cadmium load have not yet been sufficiently elucidated in the literature. The study of these processes will allow us to reveal hitherto unknown features of metabolic processes in chickens under the conditions of the development of cadmium toxicosis. Conducting research in this aspect is relevant.

The aim of the research

The research was aimed at studying the effect of Cadmium on the functional state and protein synthesis function of the liver of laying hens.

Materials and methods

24 laying hens aged 78 weeks were selected for research. These chickens were divided into three groups: control and two experimental, taking into account their age and weight. To track their metabolism, all chickens were labeled with persistent organic dyes. Chickens from the control group received compound feed and clean water without adding cadmium sulfate. In the chickens of the experimental groups, cadmium sulfate was added to the drinking water in different concentrations for 30 days: the first group (R1) – 2 mg per kilogram of body weight, the second group (R2) – 4 mg per kilogram of body weight.

The blood of laying hens was collected from the sub pterygoid vein in the following periods: before the administration of the experimental drugs and the toxicant, on the first, seventh, fourteenth, twenty-first, and thirtieth days of the experiment.

In blood serum, the activity of aspartate aminotransferase (AST; K.F. 2.6.1.1) and alanine aminotransferase (ALT; K.F. 2.6.1.2) was studied - according to the method of Reitman and Frenkel as modified by K. G. Kapetanaki (1962). The protein synthesis function of the liver of laying hens was determined by the level of total protein (biuret reaction) and protein fractions (polyacrylamide gel electrophoresis) in blood serum (Vlizlo et al., 2012).

All experimental interventions and animal slaughter were carried out in compliance with the requirements of the “European Convention for the Protection of Vertebrate Animals Used for Experimental and Scientific Purposes” (Strasbourg, 1985) and the resolutions of the First National Congress on Bioethics (Kyiv, 2001).

The research results were analyzed using the Statistica 7.0 software package. Students' t-tests assessed the probability of differences. The results of ordinal values were considered statistically significant when * – $P < 0.05$, ** – $P < 0.01$, *** – $P < 0.001$ (ANOVA).

Results and discussion

Since cadmium harms the function of the liver of animals and birds, it is essential to conduct a study of its functional state. For this, the activity of aminotransferases in the blood of chickens was determined since these enzymes reflect the functional state of the liver. Aminotransferases participate in peramination processes, transferring amino groups from amino acids to keto acids. One of the enzymes belonging to this group is alanine aminotransferase. It is one of the leading indicators of damage to liver cells under conditions of cadmium intoxication.

According to the data in Table 1, it can be seen that the activity of alanine aminotransferase in the blood serum of laying hens of the control and experimental groups at the beginning of the experiment fluctuated within physiological values. After drinking cadmium sulfate with water, an increase in the activity of this enzyme was established on the 7th day of the experiment by 12.9 % in the blood serum of the first experimental group and by 19.4 % in the second experimental group compared to the control group. On the 14th day of the experiment, the activity of alanine aminotransferase in the blood serum of laying hens of the first experimental group was 0.39 ± 0.005 mmol/g/l, and in the second experimental group – 0.42 ± 0.007 mmol/g/l. In contrast, this indicator was significantly lower in the control group and, accordingly, was 0.32 ± 0.006 mmol/g/l.

On the 21st day of the experiment, an increase in the activity of alanine aminotransferase in the blood serum of laying hens of the first experimental group was established by 24.2 % and in the second experimental group by 42.4 % compared to the indicators of the control group. It is worth noting that the highest activity of the enzyme was in the blood of laying hens of the second experimental group, where, accordingly, on the 30th day of the experi-

ment, it fluctuated within the range of 0.44 ± 0.006 mmol/g/l, which is 41.9 % higher than the control and 12.8 % – for the first experimental group.

When studying the activity of the second aminotransferase enzyme, namely aspartate aminotransferase, it was established that at the beginning of the experiment, this enzyme in the blood serum of the control and two experimental groups fluctuated within the physiological values of 4.29 ± 0.16, 4.30 ± 0.17 and 4.28 ± 0.15 mmol/g/l. After drinking Cadmium with water, an increased activity of aspartate aminotransferase was established starting from the 7th day of the experiment.

Table 1

Activity of serum aminotransferases of laying hens under conditions of cadmium loading (M ± m, n = 8)

Research groups	Before drinking	Days of research			
		7	14	21	30
ALT, mmol/g/l					
C	0.32 ± 0.007	0.31 ± 0.006	0.32 ± 0.006	0.33± 0.005	0.31 ± 0.006
R1	0.30 ± 0.009	0.35 ± 0.008*	0.39 ± 0.005***	0.41 ± 0.006***	0.39 ± 0.007***
R2	0.31 ± 0.007	0.37 ± 0.009**	0.42 ± 0.007***	0.47 ± 0.008***	0.44 ± 0.006***
AST, mmol/g/l					
C	4.29 ± 0.16	4.31 ± 0.17	4.30 ± 0.15	4.34 ± 0.16	4.32 ± 0.12
R1	4.30 ± 0.17	4.48± 0.19	4.83 ± 0.22	5.15 ± 0.17**	4.96 ± 0.21*
R2	4.28 ± 0.15	4.61 ± 0.20	5.17 ± 0.19**	5.72 ± 0.23***	5.56 ± 0.24***

On the 14th day of the experiment, an increase in the activity of the enzyme under investigation in the blood serum of laying hens that were given cadmium sulfate at a dose of 2 mg/kg of body weight was found to increase by 12.3 %, respectively. Aspartate aminotransferase activity was somewhat lower in the blood serum of the birds of the second experimental group, where it was 5.17 ± 0.19 mmol/g/l. At the same time, in control, this enzyme fluctuated within the range of 4.30 ± 0.15 mmol /g/l. In the subsequent 21 days of the experiment, the activity of aspartate aminotransferase in the serum of the birds of both experimental groups continued to increase and, relative to the control indicators, increased by 18.7 and 31.8 %, respectively. The activity of the studied enzyme on the 30th day of the experiment in the blood of the first experimental group slightly decreased to 4.96 ± 0.21, and in the second experimental group – to 5.56 ± 0.24 mmol/g/l.

High activity of the above-mentioned enzymes in the blood serum of laying hens under cadmium load indicates destructive processes in the liver, which cause increased release of aminotransaminases from cell organelles in the blood of laying hens of the research groups. Therefore, the obtained results indicate an increase in destructive processes in the body of laying hens due to exposure to heavy metals, including cadmium.

It is important to note that the increase in alanine aminotransferase activity was more pronounced than the increase in the activity of aspartate aminotransferase. This is explained by alanine aminotransferase quickly released from hepatocytes upon cadmium loading, even with minor destructive damage to their membranes. While aspartate aminotransferase is located in the mitochondria of hepatocytes, and the penetration of this enzyme into the blood is complicated not only due to the destruction of the surface membrane of the cell but also due to the need to penetrate through the mitochondrial membrane, which is possible only with large doses of cadmium.

Protein is considered the primary component that ensures the internal processes of "building" in the body. It contributes to maintaining the fluidity and viscosity of blood and determines the necessary blood volume in vessels. Proteins keep formed elements in suspension and are also responsible for transporting the most important exogenous and endogenous substances. These proteins also regulate the pH level of the blood environment and take an active part in immune reactions.

During the feeding of laying hens of the research groups with cadmium sulfate, inhibition of the function of protein synthesis in the liver was detected, as indicated by a low level of total protein in their blood. Thus, on the 14th day of the experiment, a probable decrease in the level of total protein was observed in the blood of the laying hens of the first experimental group, namely by 6.8 %, compared to the indicators of the control group of chickens. On the 21st day of the experiment, the level of total protein in the blood of the first experimental group continued to decrease. It amounted to 42.12 ± 1.21 g/l; in the control group, this indicator fluctuated within 46.70 ± 1.00 g/l (table 2). On the 30th day of the experiment, a 9 % decrease in the level of total protein was found in the blood of the chickens of the first experimental group compared to the control group.

When drinking laying hen cadmium sulfate in a dose of 4 mg/kg of body weight, a negative effect on the protein synthesis function of the liver was recorded. Thus, on the 7th day of the experiment, a probable decrease in total protein level was noted, where, in accordance with the indicators of the control group, it decreased by 4.2 %. On the 14th day of the experiment, a decrease of this indicator in the blood of chickens of the second experimental group was established to 42.33 ± 1.20 g/l. In contrast, this indicator was 46.57 ± 0.98 g/l in the control group. On the 21st and 30th day of the experiment, the lowest indicators of the level of total protein were found in the blood of the birds of the second experimental group, where, compared to the control group, it decreased by 16.4 and 14.9 %, respectively.

Table 2
The total content of protein and its fractions in the blood of chickens under conditions of cadmium loading (M ± m, n = 8)

Research groups	Before drinking	Days of research			
		7	14	21	30
Total protein, g/l					
C	46.47 ± 0.93	46.61 ± 0.85	46.57 ± 0.98	46.70 ± 1.00	46.62 ± 0.93
R1	46.69 ± 0.74	45.86 ± 1.20	43.40 ± 1.18*	42.12 ± 1.21**	42.43 ± 1.15*
R2	46,57 ± 0.91	44.64 ± 0.99	42.33 ± 1.20*	39.06 ± 1.21***	39.67 ± 1.16**
Albumins, %					
C	33.60 ± 0.72	33.91 ± 0.81	33.76 ± 0.73	34.05 ± 0.79	33.83 ± 0.87
R1	34.10 ± 0.90	32.39 ± 0.70	31.15 ± 0.85*	29.64 ± 0.98**	31.65 ± 0.93*
R2	33.80 ± 0.86	31.83 ± 0.78	29.64 ± 0.96**	28.18 ± 0.82***	29.36 ± 0.95**
Globulins, %					
C	66.40 ± 1.92	66.09 ± 1.61	66.24 ± 1.72	65.95 ± 1.69	66.17 ± 1.75
R1	65.90 ± 1.75	67.61 ± 2.13	68.85 ± 1.93	70.36 ± 1.97*	68.35 ± 2.05
R2	66.20 ± 1.81	68.17 ± 2.25	70.36 ± 1.90	71.82 ± 2.12*	70.64 ± 1.89
A/G ratio					
C	0.51	0.51	0.51	0.52	0.51
R1	0.52	0.48	0.45	0.42	0.46
R2	0.51	0.47	0.42	0.39	0.42

The decrease in the level of total protein in the blood of chickens of the experimental groups occurred in parallel with the decrease in the albumin fraction. For example, on the 14th day of the experiment, the level of albumin in the blood of the first experimental group decreased by 2.61 %, and in the second experimental group – by 4.12 %, compared to the control group of laying hens. The lowest level of the albumin fraction in the blood of birds was observed on the 21st day of the experiment, where it was 29.64 ± 0.98 % in the blood of laying hens of the first experimental group and 28.18 ± 0.82 % in the blood of hens of the second experimental group. Subsequently, on the 30th day of the experiment, we observed a decrease in the albumin level in the chickens' blood in both experimental groups.

When studying the level of globulins in the blood of laying hens under conditions of cadmium load, it was established that their level in the blood of the first and second experimental groups probably increased on the 14th and 21st days of the experiment. So, in the blood of chickens of the first research group, the level of globulins increased to 68.85 ± 1.93 and 70.36 ± 1.97 %, and in the second – 70.36 ± 1.90 and 71.82 ± 2.12 %, respectively. On the 30th day of the experiment, the level of globulins in the blood of the first and the second experimental groups remained high. An increase in the level of the globulin fraction in the blood serum of chickens with cadmium toxicosis reflects the intensity of inflammatory processes in their body.

Thus, an albumin-globulin /disproportion occurs in experimental birds under cadmium load, which is indicated by the value of the A/G ratio. In the hens of the first experimental group, it was established that the value of the A/G ratio on the 21st day of the experiment was 0.42. In contrast, in the second experimental group, it was lower and was 0.39, respectively, against the control, 0.52.

Conclusion

In laying hens, the functional state of the liver is disturbed under conditions of cadmium loading, which is indicated by the increased activity of aminotransferases in their blood serum. The highest activity of alanine and aspartate aminotransferases was in the blood serum of laying hens of the second experimental group on day 21 of the experiment.

In laying hens under cadmium load, the liver's protein synthesis function is suppressed, manifested by a decrease in the level of total protein and albumins, with a simultaneous increase in the level of globulins. Drinking cadmium sulfate with water in a larger dose (4 mg/kg of body weight) was accompanied by a more probable decrease in total protein and albumin level than drinking cadmium sulfate in a smaller dose (2 mg/kg of body weight).

Conflict of interest

The authors declare that there is no conflict of interest.

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