Elemental composition of body tissues of broiler chickens using organo-mineral feed additive

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Abstract. Consumers today are focused on safe, organic food. When introduced into the diet of animals, chitosan is able to remove toxic substances, heavy metals, stimulate cellular and humoral immunity, increasing the safety of the livestock. In this regard, the aim of the study was to study the effect of chitosan (group I), ultrafine particles (UFPs) of Fe and Cu (group II) and their combined feeding (group III) on productivity, elemental composition of blood serum and body tissues of broiler chickens. The study was performed on broiler chickens of cross "Arbor Acres" (n = 60) in the conditions of a vivarium. Separate feeding of chitosan and Fe and Cu UFPs is accompanied by an increase in the growth rate of broiler chickens. Their combined use does not give a similar result. Chitosan reduces the absorption of certain elements from the inorganic form of mineral substances. Co-feeding of chitosan with UFPs leads to an increase in the absorption of Fe and its deposition in the liver and feathers.

1. Introduction

In modern poultry farming, many different feed additives are regularly used to maintain good health and high metabolic rates, as well as to improve the performance of industrial livestock raising. The use of natural organic sources of such additives is a priority and expedient, both from the point of view of environmental safety and a beneficial effect on health. Chitosan can be one of such substances [1]. Since chitosan and its oligosaccharide derivatives contain reactive functional groups, it has antimicrobial [2], anti-inflammatory [3], antioxidant [4], antitumor [5], immunostimulating [6] and hypcholesterolemic [7] effects.

In this regard, the aim of the study is to study the effect of chitosan on growth and elemental composition of blood serum and body tissues of broiler chickens.

2. Materials and methods

The studies were carried out on broiler chickens of the Arbor Acres cross (n = 60) in a vivarium. In the experiment, four groups were formed (n = 15): chickens of the experimental group I were added chitosan at a dose of 1 mg/kg of feed (Evalar, Russia). FeSO₄ × 7H₂O and CuSO₄ × 5H₂O were Fe and Cu sources; group II - FeSO₄ × 7H₂O was replaced with Fe UFPs at a dose of 17.5 mg/kg feed (Advanced Powder Technologies, Tomsk), and CuSO₄ × 5H₂O was replaced with Cu UFPs at a dose of 1.7 mg/kg feed (OOO Platina, Moscow). Experimental group III - chitosan was introduced together with Fe and Cu UFPs, FeSO₄ × 7H₂O was replaced by CuSO₄ × 5H₂O for Cu together with chitosan.
Chickens from the control group received a premix according to the recommendation of VNITIP (2009), in which iron sulfate (FeSO4 × 7H2O) and copper (CuSO4 × 5H2O) were used as a source of Fe and Cu. The elemental composition of the organs and tissues was determined by atomic emission and mass spectrometry using mass spectrometer Elan 9000 and atomic emission spectrometer Optima 2000V (Perkin Elmer, USA).

Biochemical studies of the blood serum were performed using an automated analyzer CS-T240 (DIRUI Industrial Co., Ltd, China) with the use of commercial kits for veterinary studies DiaVetTest (Russia) and Randox Laboratories Limited (United Kingdom).

Data are expressed as means ± standard error of the mean (M ± m). Statistical analysis was carried out using Statistica 10 (StatSoft Inc., USA) and Microsoft Excel (Microsoft, USA). The significance of the group differences was assessed using the Student's t-test at p≤0.05, which was considered reliable.

3. Results

3.1. Growth
Analysis of live weight for the experiment showed a difference in the growth rate of broiler chickens. So, the chickens of the group experimental I advanced over control animals by 4.4%, and in the experimental group II the difference with the control was 2.6%.

The growth-stimulating effect of the combined use of chitosan and UFPs (group III) was lower. So, by the end of the experiment, the live weight of chickens in this group decreased by 3.7% (p≤0.05). Thus, only separate feeding of chitosan and UFPs is accompanied by an increase in the growth rate of broiler chickens.

3.2. Macronutrients in the blood serum of broiler chickens
Analyzing the concentration of macronutrients in blood serum, an increase in calcium was found by 6.04% in group I and by 69% (p≤0.05) in group III relative to the control (table 1).

Table 1. Macronutrients in blood serum of Arbor Acres broilers at the age of 42 days (experiment in a vivarium, M ± m, n = 15).

| Indicator      | Group I          | Group II         | Group III        | Control         |
|----------------|------------------|------------------|------------------|-----------------|
| Calcium, μmol / l | 2.81±0.140       | 4.48±0.224 *     | 4.03±0.201 *     | 2.65±0.132      |
| Iron, μmol / l   | 28.38±1.419 †    | 43.48±2.174      | 40.30±2.015      | 45.68±2.284     |
| Phosphorus, μmol / l | 9.69±0.484      | 9.14±0.457       | 9.05±0.452       | 10.34±0.517     |

* - significant difference between the experimental groups and the control group (p≤0.05).

In turn, the level of iron and blood serum decreases in all groups with the maximum manifestation of the effect against the background of the introduction of chitosan (group I) by 37.2% (p≤0.05) compared with the control. The amount of phosphorus has a similar tendency to decrease in experimental groups I, II and III by 6.3, 12.4 and 11.6%, respectively, relative to the control.

Thus, chitosan promotes the absorption of metals from blood serum of broilers; however, replacing inorganic metals with UFPs, this ability is suppressed.

3.3. Elemental composition of biosubstrates of broiler chickens
The addition of chitosan to the broilers' diet (group I) led to an increase in the concentration of Fe and Cu in feathers by 22.4 and 21.8% relative to the control group. Replacement of Fe and Cu sulphate in the diet for Cu and Fe UFPs (group II) leads to an increase in Fe concentration in the pen by 22.4%, while Cu concentration decreases by 24.3%, relative to the control (table 2).
Table 2. Concentration of chemical elements in feathers of Arbor Acres broilers at the age of 42 days (experiment in vivarium conditions, M ± m, n = 15).

| Indicator                      | Group I           | Group II          | Group III          | Control        |
|-------------------------------|-------------------|-------------------|--------------------|----------------|
| Iron concentration, mg/kg     | 6.65±0.85\(^a\)  | 6.65±0.51         | 6.38±0.71\(^a\)   | 5.43±0.61      |
| Copper concentration, mg/kg   | 3.07±0.18\(^a\)  | 1.9±0.31\(^a\)   | 1.6±0.11\(^a\)    | 2.52±0.09      |

\(^a\) - Significant difference between the experimental groups and the control group (p≤0.05).

A similar effect can be traced in the third experimental group: the Fe concentration increases by 17.5%, and the Cu concentration decreases by 36% relative to the control.

The dynamics of the concentration of the studied elements in the liver is different. Thus, the amount of Fe in the liver tends to increase in all experimental groups compared to the control (table 3), in group I it increases by 33.6%, in group II - by 36.9%, and in group III - by 36.9%.

Table 3. Concentrations of chemical elements in the liver of Arbor Acres broilers at the age of 42 days (experiment in vivarium conditions, M ± m, n = 15).

| Indicator                      | Group I           | Group II          | Group III          | Control        |
|-------------------------------|-------------------|-------------------|--------------------|----------------|
| Iron concentration, mg/kg     | 101.3±4.1\(^a\)  | 103.8±7.6\(^a\)  | 132.9±7.8\(^a\)   | 75.8±9.5       |
| Copper concentration, mg/kg   | 2.49±0.5          | 2.7±0.2           | 1.95±0.3\(^a\)    | 2.47±0.2       |

\(^a\) - Significant difference between the experimental groups and the control group (p≤0.05).

The concentration of Cu with the introduction of chitosan (group I) increases insignificantly, while with the addition of UFPs (group II) to the diet, it increases by 9.3%. After the combined feeding with chitosan and UFPs (group III), the opposite effect is observed, Cu concentration decreases by 21.4% compared to the control.

Thus, the concentration of the studied elements in biosubstrates is influenced by the form of the element used in the experiment and the presence of the sorbent in the form of chitosan.

4. Discussion

Chitosan just like UFPs, can stimulate the activity of some biochemical processes in the metabolic chain [8, 9]. The experiment proved that the body weight and food consumption of chickens receiving chitosan in the diet differed from the control animals. Many scientists argue that the increased digestibility of nutrients in the intestines of broilers fed with chitosan in the diet is a consequence of a decrease in the number of pathogenic bacteria [10, 11]. Also, chitosan can stimulate the secretion of digestive enzymes [12].

The improvement in calcium absorption may be associated with the positive effect of chitosan on the structural elements of the intestinal wall of broiler chickens. It is possible that chitosan in the diet increases the digestibility of Ca and P and, as a consequence, improves the absorption of nutrients and growth rates [13, 14].

The introduction of Fe and Cu UFPs into broilers’ diet influences their concentration in the body of animals. The obtained result confirms the feasibility of replacing the nonorganic form of elements with a decrease in the intake standard, due to no negative effects on health [15]. It has been proven that the combination of iron oxide nanoparticles has great potential for use in poultry feed for large-scale meat production without any toxicological effects [16].

Studies have shown that chitosan can inhibit the absorption of certain elements, such as Ca, in a feed additive. However, UFPs of metals introduced together with chitosan enhance the effect of accumulation, using chitosan as a carrier [17].
5. Conclusion
The use of chitosan, both separately and in combination with Fe and Cu UFPs, does not have a negative effect on health of broiler chickens. An increase in the productive effect is more characteristic against the background of feeding only with chitosan. Chitosan reduces the absorption of certain elements from the inorganic form of substances. Combined feeding with chitosan and UFPs leads to an increase in the absorption of Fe and its deposition in the liver and feathers.

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