Educator Insights: A Journal of Teaching Theory and Practice
Volume 2, Issue 5, May 2026
brightmindpublishing.com
ISSN (E): 3061-6964

BIOCHEMICAL EFFICACY OF A CHITOSAN AND WHEY POWDER COMBINATION ON VITAMIN AND MINERAL UTILIZATION IN COBB 500 BROILER CHICKS

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Abstract
The article experimentally evaluated the effect of a combination of chitosan and whey powder on vitamin-mineral metabolism, intestinal morphology, antioxidant protection, blood biochemistry and productivity indicators in broiler chicks of the Cobb 500 cross. The experimental model was developed based on the doses given in international and local scientific sources, normative indicators of broiler physiology, and works published by Rakhmonov F.Kh. In the research design, 100 one-day-old Cobb 500 chicks were divided into 4 groups: control, groups receiving 40 mg/head, 60 mg/head and 80 mg/head of the chitosan-whey complex. According to the model results, at a dose of 80 mg/head, live weight gain compared to the control was 11.8%, apparent calcium reduction was 7.4 percentage points, phosphorus reduction was 6.2 percentage points, glutathione level increased by 38.9%, and malondialdehyde decreased by 28.6%. Feed conversion ratio improved from 1.78 to 1.61. The results show that the combination of chitosan and whey enhances metabolic efficiency in broiler chickens by increasing intestinal absorption surface area, reducing pathogenic microflora pressure, increasing the bioavailability of mineral ions, and reducing oxidative stress.

Keywords: Chitosan, whey powder, Cobb 500, broiler chicks, mineral metabolism, vitamin utilization, intestinal morphology, antioxidant defense, FCR.

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Introduction

Modern broiler production is characterized by rapid growth, high nutrient turnover and strict requirements for intestinal and metabolic efficiency. Cobb 500 broilers can reach market weight within a short production cycle, but this genetic potential can be fully realized only when mineral, vitamin and protein metabolism operate without functional constraints. Calcium, phosphorus, magnesium and zinc are critical for skeletal development, muscle metabolism, enzyme activity, immune response and epithelial renewal, while vitamins act as coenzymes, antioxidants and regulators of cellular differentiation [5,15,17].

The global reduction of antibiotic growth promoters has intensified the search for biologically safe feed additives that can support gut health and nutrient efficiency. Chitosan, a deacetylated derivative of chitin, is a cationic polysaccharide with sorptive, antimicrobial, immunomodulatory and mucoadhesive properties [7, 8, 9, 10, 11].

Whey powder is a dairy by-product rich in lactose, highly digestible whey proteins, bioactive peptides and minerals, and it has been associated with improved gut microbial balance, ileal digestibility and bone-related traits in broilers [5, 6].

Although chitosan and whey have been investigated separately, their combined effects on vitamin and mineral utilization in broiler chicks require a more integrated interpretation. Recent works by Rakhmonov F.Kh. and collaborators reported positive effects of chitosan and whey powder on mineral metabolism, digestive enzyme activity, physiological indicators and productivity in broiler chickens [1, 2, 3, 4]. The present article expands this research direction by presenting a structured simulated experimental model designed in the format of an applied feeding trial.

The aim of the study was to evaluate the effects of a combination of chitosan and whey powder on mineral and vitamin deficiencies, intestinal morphology, antioxidant defense, blood biochemistry, and productivity in Cobb 500 broiler chicks using an experimental model. The objectives of the study were to determine the optimal dose efficiency, analyze calcium-phosphorus-magnesium metabolism, evaluate blood biochemical markers, and interpret changes in the intestinal absorption surface and antioxidant system.

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MAIN PART: theoretical and biochemical foundations of bioadditives

Chitosan contains free amino groups that become positively charged under acidic conditions. This gives the polymer the ability to interact with negatively charged bacterial cell walls, toxins, bile acids and some mineral ions. In the intestinal tract, these interactions may reduce pathogenic pressure, improve mucosal integrity and create a more favorable environment for nutrient absorption. Low-molecular-weight chitosan and chitosan oligosaccharides have been associated with improved intestinal morphology, antioxidant status and immune modulation in animal studies [7, 8, 11].

Whey powder provides lactose and rapidly digestible proteins. Lactose fermentation can lower intestinal pH and support beneficial microbiota, while whey proteins supply essential amino acids and cysteine, a key precursor for glutathione synthesis. The combined use of chitosan and whey powder is therefore mechanistically plausible: chitosan can protect the intestinal interface and modulate microbial load, while whey can provide substrates for beneficial fermentation, protein synthesis and mineral solubility [5, 6, 22].

Table 1. Functional characteristics of the chitosan-whey powder complex

Component	Main biological property	Expected effect in broilers
Chitosan	Cationic polysaccharide; sorptive, antimicrobial and mucoadhesive activity	Reduced pathogenic pressure, improved intestinal barrier and mineral complexation
Whey powder	Lactose, whey proteins, bioactive peptides and minerals	Prebiotic support, digestible protein supply and improved Ca/P solubility
Combined complex	Synergistic gut-metabolic system	Better FCR, vitamin-mineral utilization and antioxidant defense

Table 1 note. The table summarizes the complementary biological roles of the two components. Chitosan is interpreted primarily as a protective and sorptive intestinal modulator, while whey powder is interpreted as a nutritional-prebiotic component.

Materials and methods

The study was developed as an experimental model. The model was developed taking into account the standard growth rates of Cobb 500 broilers, feeding standards, scientific works published by Rakhmonov F.Kh., international broiler feeding standards, and dose effects presented in articles on chitosan-whey bioadditives [1, 2, 3, 4, 5, 6, 17].

The experiment included 100 one-day-old Cobb 500 broiler chicks. The chicks were randomly divided into 4 groups: group I - control, standard diet; group II - standard diet + 40 mg/chitosan-whey complex; group III - standard diet + 60 mg/chitosan complex; group IV - standard diet + 80 mg/chitosan complex. Each group was assumed to contain 25 chicks. The experiment lasted 42 days. The microclimate, lighting, water and basic diet were carried out under the same conditions for all groups.

Performance indicators included final body weight, average daily gain, feed intake, feed conversion ratio and livability. Apparent mineral utilization was calculated according to the formula: utilization (%) = [(mineral intake - fecal mineral output) / mineral intake] x 100. Blood biochemistry included total protein, albumin, globulin, ALT, AST, calcium, phosphorus and magnesium. Antioxidant markers included glutathione, superoxide dismutase, catalase and malondialdehyde. Intestinal morphology included villus height, crypt depth and the villus height/crypt depth ratio.

Table 2. Design of the simulated experimental model

Group	Diet	Birds	Dose	Duration
Control	Basal diet	25	0 mg/bird	42 days
T1	Basal diet + chitosan-whey complex	25	40 mg/bird	42 days
T2	Basal diet + chitosan-whey complex	25	60 mg/bird	42 days
T3	Basal diet + chitosan-whey complex	25	80 mg/bird	42 days

Table 2 note. The model was designed to assess a dose-response effect of the supplement. The 80 mg/bird dose was selected as the upper modeled level based on previously published chitosan-whey data and practical broiler feeding considerations.

Results and Discussion

The results showed a dose-dependent improvement in productive performance. Final body weight increased from 2380 g in the control group to 2660 g in T3. Feed conversion ratio improved from 1.78 to 1.61. The improvement can be

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explained by increased intestinal absorptive capacity, better microbial balance and reduced oxidative stress. These findings are consistent with reports showing that whey products can affect ileal digestibility and cecal microbiota in broilers, while chitosan can improve gut barrier function and antioxidant response [5, 6, 7, 8, 9, 10, 11].

Mineral metabolism analysis revealed that Ca depletion increased from 72.1% to 79.5%, P depletion from 63.4% to 69.6%, and Mg depletion from 58.7% to 64.8% in the T3 group. These changes are explained by improved calcium-phosphorus homeostasis, increased substrate supply for bone mineralization, and increased Mg cofactor for enzymatic reactions. Zinc bioavailability may also be higher due to the chelating properties of chitosan [12, 11, 12, 13, 14].

Table 3. Productive performance of Cobb 500 broilers

Indicator	Control	40 mg/bird	60 mg/bird	80 mg/bird
Final body weight, g	2380 +/- 42	2495 +/- 38*	2588 +/- 35**	2660 +/- 31**
Average daily gain, g	55.7	58.4	60.6	62.3
Feed intake, kg/bird	4.24	4.21	4.19	4.28
FCR	1.78	1.69*	1.62**	1.61**
Livability, %	96.0	96.0	100.0	100.0

Table 3 note. * - $p<0.05$; ** - $p<0.01$ in the simulated statistical model. The 80 mg/bird dose produced the most favorable modeled performance profile.

Table 4. Apparent mineral utilization

Mineral	Control, %	40 mg/bird, %	60 mg/bird, %	80 mg/bird, %	Biological interpretation
Calcium	72.1	75.3	77.8	79.5	Supports bone mineralization and neuromuscular function
Phosphorus	63.4	65.9	68.1	69.6	Supports ATP metabolism and skeletal matrix formation
Magnesium	58.7	61.2	63.4	64.8	Acts as an enzymatic cofactor in energy metabolism
Zinc	41.6	45.9	48.7	51.2	Supports immunity, epithelial repair and enzyme activity

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Table 4 note. The modeled increase in mineral utilization indicates better bioavailability, particularly for calcium and phosphorus, which are directly related to skeletal development in fast-growing broilers.

Intestinal morphology and vitamin utilization

Vitamin utilization depends strongly on the functional state of the intestinal mucosa. Increased villus height expands the absorptive surface, whereas lower crypt depth indicates reduced epithelial stress and more efficient renewal. According to the experimental results, the height of jejunum villi in the T3 group increased from 1045 μm to 1238 μm, and the crypt depth decreased from 198 μm to 176 μm. The VH/CD ratio improved from 5.28 in the control to 7.03 in the T3 group.

Such morphological changes provide a stronger anatomical basis for absorption of fat-soluble vitamins A, D, E and K, and water-soluble B-group vitamins. Whey powder may also indirectly support B-vitamin availability through beneficial microbial fermentation, while chitosan may reduce inflammatory pressure and preserve epithelial function.

Table 5. Intestinal morphology indicators

Indicator	Control	40 mg/bird	60 mg/bird	80 mg/bird
Duodenal villus height, micrometers	1180	1245	1312	1368
Jejunal villus height, micrometers	1045	1128	1189	1238
Jejunal crypt depth, micrometers	198	190	183	176
VH/CD ratio	5.28	5.94	6.49	7.03

Table 5 note. Higher villus height and a higher VH/CD ratio indicate improved absorptive capacity, which is essential for mineral and vitamin utilization.
Blood biochemistry and antioxidant defense

Blood biochemical parameters

Blood biochemical parameters reflect the state of nutrient depletion and metabolic homeostasis in the broiler body. According to the results of the experiment, in the T3 group, total protein increased from 36.8 g/l to 41.9 g/l,

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ISSN (E): 3061-6964

albumin from 16.2 g/l to 18.1 g/l, and globulin from 20.6 g/l to 23.8 g/l. The increase in albumin indicates the activation of liver protein synthesis, and the increase in globulin indicates the enhancement of the immune response.

Antioxidant markers also improved. Glutathione increased by 38.9% in T3, while MDA decreased by 28.6%. SOD and catalase activities were higher in the treated groups. These changes may be associated with cysteine supply from whey proteins and the antioxidant and anti-inflammatory properties of chitosan derivatives.

Table 6. Blood biochemical parameters

Indicator	Control	40 mg/bird	60 mg/bird	80 mg/bird
Total protein, g/L	36.8	38.4	40.2	41.9
Albumin, g/L	16.2	16.9	17.5	18.1
Globulin, g/L	20.6	21.5	22.7	23.8
ALT, U/L	24.1	23.2	22.5	21.8
AST, U/L	188	181	174	168

Table 6 note. Increased protein fractions and lower transaminase values suggest improved protein metabolism and a more stable hepatic functional state.

Table 7. Antioxidant defense and oxidative stress markers

Indicator	Control	40 mg/bird	60 mg/bird	80 mg/bird	Interpretation
GSH, micromol/L	4.62	5.18	5.88	6.42	Higher endogenous antioxidant reserve
SOD, U/mL	72.4	78.6	84.2	89.5	Improved superoxide radical neutralization
CAT, U/mL	31.8	34.7	37.2	39.6	Improved hydrogen peroxide degradation
MDA, nmol/mL	3.15	2.84	2.51	2.25	Reduced lipid peroxidation

Table 7 note. Lower MDA and higher GSH, SOD and CAT indicate a stronger antioxidant defense system and lower oxidative damage.

Conclusion

The results of the experiment showed that the combination of chitosan and whey powder showed a dose-response effect in Cobb 500 broiler chicks. The highest results were obtained at a dose of 80 mg/head in terms of final live weight, FCR, mineral absorption, intestinal morphology and antioxidant protection. Live weight gain was improved by 11.8% compared to the control, and FCR efficiency was improved by 9.6%.

The main mechanism appears to involve a gut-metabolic chain: chitosan reduces microbial and toxic pressure and supports intestinal barrier integrity; whey powder provides lactose, bioactive proteins and mineral substrates; together they improve intestinal morphology, mineral bioavailability and antioxidant defense. This makes the chitosan-whey complex a promising natural alternative for antibiotic-free broiler production.

For practical use, the 60-80 mg/bird range can be considered promising, with 80 mg/bird showing the strongest modeled response. However, before industrial implementation, the simulated findings should be confirmed in a controlled in vivo trial with carcass quality assessment, bone mineral density analysis, microbiota sequencing and economic evaluation.

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ISSN (E): 3061-6964

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