Effect of Different Commercial Garlic Preparations Added to Drinking Water on Physiological and Immunological Traits of Broiler Chickens (Ross 308)

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Abstract

The experiment was conducted in the broiler hall of the Animal Production Department at the College of Agriculture / Tikrit University for the period from 17/2/2025 to 24/3/2025 to study the effect of using different types of commercial garlic in drinking water and its impact on some productive traits of broiler chickens. A total of 225 one-day-old Ross 308 broiler chicks were used, placed in floor pens, and divided into five treatments. Each treatment contained three replicates, and within each replicate, there were fifteen birds. The first treatment was the control group. The birds were fed a ready-made starter diet from the Middle East factory until 10 days of age. Subsequently, experimental diets mixed in the feed mill of the College of Agriculture / Tikrit University were used. A grower diet was used from 10 to 24 days of age, and then the diet was switched to a finisher diet, fed from 25 to 35 days of age. Feeding was *ad libitum*. The experimental materials were added to the drinking water starting from 10 days to 35 days of age. The additions were 500 ml/1000 liters for T2, 350 ml/1000 liters for T3, 200 ml/1000 liters for T4, and 200 g/1000 liters for T5, according to the recommendations of the manufacturing companies, using plastic manual feeders and drinkers.

The results of the statistical analysis showed no significant differences in blood serum proteins (albumin, globulin, and total protein). The experiment showed a significant superiority in the third treatment regarding humoral immunity (IgM) compared to the control treatment, as it gave the highest value compared to the rest of the addition treatments, recording 17.426. As for the lymphoid organs, the experiment recorded no significant differences in the relative weight of the spleen, Thymus gland

) Compared to the control treatment, while the third treatment recorded a significant decrease compared to the control group regarding the relative weight of the bursa of Fabricius. As for the blood picture indicators, no significant differences were observed in the white blood cell counts between the addition

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treatments and the control treatment, while a decrease was observed in the third treatment regarding heterophils compared to the control treatment, a significant increase in the third treatment in the percentage of lymphocytes compared to the control, and a significant decrease in the H/L% (oxidative stress indicator) for the third treatment compared to the control.

Keywords: Blood parameters, Humoral immunity, Phytogenic additives, Poultry nutrition.

Introduction

Broiler production is considered one of the most important pillars for providing high-quality animal protein, given its fast production cycle and high efficiency in converting feed into meat, making it an effective economic sector and a fundamental contributor to achieving food security [1]. The success of this industry relies on the application of precise feeding programs that contribute to meeting nutritional requirements and enhancing the birds' health and production quality [2].

Poultry nutrition faces several challenges related to the environment and climatic conditions, and heat stress is one of the most impactful challenges on the birds, as it leads to a decrease in feed consumption and disturbances in metabolic processes. As indicated [3], there is a reliance on modern nutritional strategies such as using natural antioxidants and certain vitamins and organic minerals to reduce the negative effects of stress and improve the Poultry immunity, which contributes to maintaining adequate performance under various conditions.

With the global trend towards producing poultry meat free of antibiotics, gut health has become a central focus in raising production efficiency and reducing economic losses resulting from infections and immune challenges [4]. The intestine is the vital organ most closely associated with nutrient absorption and the bird's immune response, and any disruption in the balance of the intestinal microbiome negatively reflects on growth, feed conversion ratio, and meat quality [5].

Hence, the importance of using natural additives like garlic arises, as its active sulfur compounds work to inhibit harmful microbes and promote the growth of beneficial bacteria such as *Lactobacillus*, which improves intestinal integrity and reduces the need for prophylactic antibiotics [6].

Regarding diet components, the focus is no longer solely on energy or crude protein sources, but has also shifted to functional feed

additives, which have gained importance following restrictions on the use of antibiotics as growth promoters. Supporting gut health is a primary objective in this regard, as the intestine is currently viewed not only for absorption and digestion but also as a major part of the bird's immune system.

[7] Also pointed out the possibility of improving microbial balance within the intestine using probiotics, organic acids, and some plant extracts, which helps boost immunity and improve feed conversion efficiency.

With escalating global interest in reducing the use of antibiotics in poultry farming due to bacterial resistance and health risks they cause, researchers have turned to safe, natural alternatives that enhance bird health and maintain productivity without leaving drug residues in animal products. Garlic (*Allium sativum*) is among the most prominent of these alternatives due to its active compounds, primarily allicin, which possess antimicrobial and antioxidant properties, thereby enhancing digestive health and immunity [8].

Studies have proven that adding garlic extracts to drinking water is a practical and easy-to-apply method, as it allows for the rapid absorption of vital compounds without the need to significantly modify the diet. This addition also contributes to improving intestinal structure by increasing villus height and crypt depth, which positively reflects on the efficiency of nutrient utilization and the feed conversion ratio [9].

Recent research indicates that antibiotic alternatives, including garlic extracts, contribute to reducing reliance on antibiotics, supporting gut health and immunity, and improving the quality of the final product, thereby enhancing sustainability in the poultry industry [10].

The study aimed to obtain commercial garlic extracts and Active compounds available in local markets that are used by breeders, to study and use them in broiler rearing, to demonstrate their effect on the physiological and immunological performance of broiler chickens, and to determine which formulation is best without affecting the health and quality of the chickens.

Materials and Methods

A total of 225 one-day-old Ross 308 broiler chicks, obtained from Al-Alamein hatchery in Saladin Governorate, were used. They were placed in floor pens and divided into five treatments. Each treatment contained three replicates, with fifteen birds per replicate. The first treatment was the control group. The birds were fed a ready-made starter diet from the Middle East factory until 10 days of age. Subsequently, experimental diets mixed in the feed mill of the College of Agriculture /

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Tikrit University were used. A grower diet was used from 11 to 24 days of age, and then the diet was switched to a finisher diet, fed from 25 to 35 days of age (Table 1). Feeding was *ad libitum*.

The experimental materials were added to the drinking water starting from 10 days of age until 35 days of age using plastic manual feeders and drinkers. The lighting program was 23 hours of light / 1 hour of darkness. The temperature was 34°C at the beginning of the experiment and was reduced to 24°C at the end of the experiment. The following traits were studied: blood serum proteins, humoral immunity (IgM), lymphoid organs, and some blood picture indicators. Data were statistically analyzed using a Completely Randomized Design (CRD) to study the effect of the treatments on the studied traits. Significant differences among means were compared using Duncan's multiple range test [11], and the [12] was used for statistical analysis.

Table (1): Illustrates the materials used in each treatment

Treatment	Dosage	Components
T1	No addition	Control treatment – no additive in drinking water
T2	500 ml/1000 L	Garlic 40,000 mg/l, Thyme 40,000 mg/l, Thymol 16,000 mg/l, Cinnamaldehyde 14,000 mg/l
T3	350 ml/1000 L	Garlic Oregano, Vitamin E 5,000 mg/l, Azadirachta indica 20,000 mg/l, Ocimum sanctum 10,000 mg/l, Phyllanthus emblica 5,000 mg/l, Vitamin B12 1,000 mg/l, Betaine 30,000 mg/l, Methionine 30,000 mg/l, Magnesium Sulfate 15,000 mg/l
T4	200 ml/1000 L	Garlic oil extract, Coriander extract, Vitamin A 6,000 IU, Vitamin E 12,000 mg/l
T5	200 g/1000 L	Pure Allicin Powder 250 g/kg

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Table (2): Components of broiler chicken diets (Grower and Finisher) with calculated chemical composition

Diets: Finisher	Diets: Grower	Feed Ingredients
65.82	60.23	Yellow Corn (%)
28.45	33.62	Soybean Meal (48%)
0.14	0.13	Lysine
2.5	2.5	*Premix (%)
2.03	2.15	Sunflower Oil (%)
0.6	0.9	Dicalcium Phosphate (%)
0.1	0.1	Limestone (%)
0.11	0.12	DL-Methionine (%)
0.25	0.25	Table Salt (%)
100	**100**	Total
**Calculated Chemical Composition		
3100	3050	Metabolizable Energy (kcal/kg feed)
19.5	21.51	Crude Protein (%)
2.28	2.3	Crude Fiber (%)
1.08	1.18	Lysine (%)
0.56	0.59	Methionine (%)
0.86	0.92	Methionine + Cystine (%)
0.79	0.87	Calcium (%)
0.36	0.42	Phosphorus (%)

*Premix (%): Calcium 20.40%; Phosphorus 4.90%; Sodium 5.00%; Chloride 7.79%; Crude Protein 10.00%; Crude Fat 2.65%; Crude Fiber 0.25%; Ash 78.09%; Lysine 1.60%; Methionine 6.00%; Threonine 0.16%; Methionine+Cystine 6.05%.

**The chemical composition values of the feed ingredients included in the diet formulation were calculated [13].

Statistical Analysis

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Data were statistically analyzed using a Completely Randomized Design (CRD) to study the effect of treatments on the studied traits. Significant differences among means were compared using Duncan's multiple range test [11], and SAS version 9.1 was used for statistical analysis [12].

Results and Discussion

Blood Serum Proteins and Humoral Immunit

The results presented in Table (3) showed that the supplementation of garlic in drinking water had no significant effect ($P > 0.05$) on serum total protein, albumin, and globulin concentrations. This indicates that the levels of garlic preparations used in this study did not impair liver function or protein metabolism, suggesting that these additives are physiologically safe for broiler chickens. The absence of significant changes in globulin levels further reflects the stability of the immune-related protein fraction under the experimental conditions.

On the other hand, a significant improvement ($P \leq 0.05$) in humoral immunity (IgM) was observed in T3 compared to the control (T1) and other treatments. This enhancement in IgM concentration indicates a stimulatory effect of garlic-based additives on the immune system. The superior response observed in T3 may be attributed to the synergistic effect of garlic combined with other bioactive compounds such as vitamins, amino acids, and herbal extracts present in this treatment.

The immunostimulatory effect of garlic is mainly associated with its sulfur-containing compounds, particularly allicin, which has been reported to enhance lymphocyte proliferation, antibody production, and overall immune responsiveness. These findings are in agreement with those reported by Fathi and Rezaee [14], who observed improved immune parameters in broilers supplemented with garlic-derived products.

Interestingly, the increase in IgM levels occurred without significant alterations in serum protein fractions, suggesting that garlic supplementation selectively enhances immune function without disrupting general metabolic processes. This selective effect may be advantageous in poultry production systems aiming to improve immunity without compromising physiological balance.

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Table (3): Effect of adding different types of commercial garlic to drinking water on blood serum proteins and humoral immunity (M± SE)

Treatment	Total Protein		Albumin		Globulin		Humoral Immunity (IgM)
T1	4.972 0.393	±	1.359 0.070	±	3.613 0.371	±	10.423 ± 0.836 c
T2	4.792 0.244	±	1.433 0.083	±	3.359 0.253	±	11.484 ± 0.633 bc
T3	4.813 0.281	±	1.163 0.098	±	3.649 0.542	±	17.426 ± 1.648 a
T4	4.132 0.092	±	1.121 0.140	±	3.011 0.121	±	13.225 ± 0.472 bc
T5	4.727 0.297	±	1.144 0.061	±	3.359 0.302	±	14.127 ± 0.378 b
Significance	N.S		N.S		N.S		*

- T1 Control treatment, T2 Garlic extract 500ml/1000L, T3 Garlic and herbs 350ml/1000L, T4 Garlic oil 200ml/1000L, T5 Garlic powder 200g/1000L.

***** Indicates the presence of significant differences among treatments.

- Different letters in the same column indicate significant differences among treatments at a probability level of ($P \leq 0.05$).

- N.S indicates no significant differences among treatments.

Lymphoid Organs

The results of Table (4) showed no significant differences ($P \leq 0.05$) in the relative weights of both spleen weight and thymus weight. This indicates that using garlic at the employed concentrations was physiologically safe and did not cause abnormal enlargement or inflammation in these organs.

As for the relative weight of the bursa of Fabricius, the third treatment achieved a significant decrease ($P \leq 0.05$) compared to the rest of the treatments, recording 0.13, while the first, second, fourth, and fifth treatments recorded 0.20, 0.21, 0.21, and 0.22, respectively. This indicates that garlic at this concentration stimulated the working efficiency of the immune system and not just an increase in size, as the lymphocytes in the gland successfully transformed into plasma cells, producing antibodies in the blood circulation.

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Table (4): Effect of adding different types of commercial garlic to drinking water on organic immune parameters (M± SE)

Bursa of Fabricius Weight %	Thymus Weight %	Spleen Weight %	Treatment
a 0.019 ± 0.20	0.028 ± 0.28	0.025 ± 0.17	T1
a 0.239 ± 0.21	0.039 ± 0.36	0.013 ± 0.20	T2
b 0.015 ± 0.13	0.035 ± 0.32	0.045 ± 0.21	T3
a 0.021 ± 0.21	0.009 ± 0.38	0.015 ± 0.22	T4
a 0.023 ± 0.22	0.037 ± 0.38	0.028 ± 0.20	T5
*	N.S	N.S	Significance

- T1 Control treatment, T2 Garlic extract 500ml/1000L, T3 Garlic and herbs 350ml/1000L, T4 Garlic oil 200ml/1000L, T5 Garlic powder 200g/1000L.

***** Indicates the presence of significant differences among treatments.

- Different letters in the same column indicate significant differences among treatments at a probability level of ($P \leq 0.05$).

- N.S indicates no significant differences among treatments.

Some Hematological Parameters

Table (5) showed that the addition of garlic to drinking water did not significantly affect the white blood cells in the blood. As for the heterophils H% , a significant decrease ($P \leq 0.05$) was observed in the addition treatments (second, third, fourth, and fifth) compared to the control group, where the third treatment recorded the lowest value of 22.750, while the second, fourth, and fifth treatments recorded (25.250, 26.000, 25.750) respectively. The decrease in this percentage often indicates an improvement in health status or a reduction in inflammatory agents.

As for the percentage of lymphocytes (L%) ,a significant increase ($P \leq 0.05$) was observed in the third and fourth treatments, recording (66.250, 64.500) respectively, while the first, second, and fifth treatments recorded (60.500, 63.750, 63.250) respectively. Since lymphocytes are primarily responsible for the immune response and the formation of antibodies, their increase indicates the enhancement of the birds' immune system as a result of garlic addition.

As for the H/L% ratio (stress indicator), a significant decrease was

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observed in the addition treatments compared to the control treatment, and the third treatment recorded the lowest value of 0.342, while the second, fourth, and fifth treatments recorded 0.395, 0.402, and 0.407, respectively. The decrease in this ratio indicates that the birds are living in a state of physiological rest, away from environmental or nutritional stress.

Table (5): Effect of adding different types of commercial garlic to drinking water on some blood picture indicators (M ± SE)

Treatment	WBC	H%	Lym%	H/L%
T1	17.950 ± 0.993	29.000 ± 0.912 a	60.500 ± 0.645 b	0.480 ± 0.019 a
T2	19.100 ± 0.759	25.250 ± 0.853 bc	63.750 ± 0.853 ab	0.395 ± 0.015 bc
T3	20.600 ± 1.003	22.750 ± 0.853 c	66.250 ± 1.314 a	0.342 ± 0.018 c
T4	18.525 ± 0.566	26.000 ± 1.080 b	64.500 ± 1.707 a	0.402 ± 0.020 b
T5	17.950 ± 0.960	25.750 ± 0.946 b	63.250 ± 0.250 ab	0.407 ± 0.013 b
Significance	N.S	*	*	*

- T1 Control treatment, T2 Garlic extract 500ml/1000L, T3 Garlic and herbs 350ml/1000L, T4 Garlic oil 200ml/1000L, T5 Garlic powder 200g/1000L.

***** Indicates the presence of significant differences among treatments.

- Different letters in the same column indicate significant differences among treatments at a probability level of ($P \leq 0.05$).

- N.S indicates no significant differences among treatments.

- WBC: White blood cells, Het%: Heterophils, Lym%: Lymphocytes, Het/Lym%: Stress ratio

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Conclusions

1. The addition of garlic to drinking water did not lead to significant differences in blood serum proteins, relative weights of the thymus and spleen, and white blood cell counts.
2. The addition of garlic to drinking water led to a significant increase in humoral immunity and lymphocytes, a decrease in the oxidative stress indicator, and also a decrease in the relative weight of the bursa of Fabricius.

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