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Article

Impact of Dietary Nutmeg (*Myristica fragrans*) Powder Supplementation on Hematological and Biochemical Parameters of Ross 308 Broiler Chickens

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Abstract: This research was carried out at the Animal Production Field, College of Med. & Ind. Plants, Uni. of Kirkuk, between March 17 and April 20, 2024, to assess the impact of dietary nutmeg powder supplementation on specific blood parameters of Ross 308 broilers. Birds were randomly assigned to four treatments: T1 (control), T2 (1 g/kg), T3 (2 g/kg), and T4 (3 g/kg) diets. There was no significant difference ($P > 0.05$) for the hematological parameters, where the highest number of red blood cells was recorded for the control group ($12.01 \times 10^6/mm^3$), the highest value of hemoglobin was recorded for T4 (11.17 g dL^{-1}), and the highest packed cell volume was recorded for T2 (29.25%). This indicated that the diet supplemented with up to 3 g/kg of nutmeg powder did not adversely affect the blood health status of the birds. Conversely, significant differences ($P<0.05$) were observed in the biochemical parameters, where total cholesterol decreased from 108.3 mg dL^{-1} in the control treatment to 84.63 mg dL^{-1} in T4, triglycerides declined from 129.74 to 89.74 mg dL^{-1}, LDL decreased from 57.73 to 30.99 mg dL^{-1}, and VLDL decreased from 25.94 to 17.94 mg dL^{-1}, while HDL increased from 47.70 mg dL^{-1} in the control to 59.54 mg dL^{-1} in T4. The improvement was more pronounced at the 2 and 3 g/kg inclusion levels, suggesting that nutmeg powder can be used as a safe and effective phytogenic feed additive to improve serum lipid profile without affecting hematological stability under the conditions of this study.

Keywords: Eco Nutmeg Powder, Ross 308 Broilers, Hematological Parameters, Serum Lipid Profile, Phytogenic Feed Additive

Introduction

The supplementation of nutmeg powder to the diet of broilers was examined for its influence on certain hematological and biochemical parameters of Ross 308 broilers. Chicken meat has been a significant source of good quality animal protein, given its richness in vital nutrients, low content of trans fatty acids, and relatively low price. Therefore, the consumption of poultry meat has continued to increase, along with the growth in the human population [1, 2]. Broiler development has been of critical importance to meet the growing demand for protein, requiring effective management to maximize the economic benefits obtained from such production [3, 4]. In the poultry industry, antibiotic growth promoters have been fed to poultry for many years to minimize the incidence of disease and maximize growth rates [5]. However, the use of antibiotics has been related to the development of antibiotic-resistant bacteria, leading to antibiotic residues in poultry products, thereby posing a danger to human health. In many countries, the use of antibiotics as growth promoters has been prohibited, and the safety of poultry products has become a concern for many people [6].

In this regard, phytogenic feed additives from medicinal and aromatic plants have gained considerable interest in the field of poultry science as a source of natural growth promoters. These plant-based additives are considered to be safe and environmentally friendly with minimal risk of any harmful residues in chicken meat products [7]. Previous studies have shown that the addition of herbal additives to the diets of broilers improves the characteristics of the carcass, metabolic activity, and meat products [8].

Nutmeg is one of the plant-based additives used in the food industry that is obtained from the seed of the Indonesian plant Myristica fragrans and is used as a spice in cooking. Nutmeg contains bioactive compounds that have antioxidant activity, including myricetin [9, 10 ,11]. Furthermore, the active compound myristicin also has antimicrobial activity and is used in different food industries as a natural preservative. However, excessive consumption of nutmeg has been known to have harmful effects on the liver and central nervous system [12, 13]. Nutmeg oil has also been used in different studies to show that it increases appetite and weight gain in animals, leading to anti-obesity activity in different models [14, 15].

Despite the known biological activity of nutmeg, little information is available on the effect of nutmeg powder on hematological and biochemical blood parameters in broiler chickens. For this purpose, the present study was conducted to investigate the effect of nutmeg powder on certain hematological and biochemical blood parameters in Ross 308 broilers.

Materials and Methods

1. Experimental Location, Birds, and Management

The experiment was conducted at the College of Medicinal and Industrial Plants, Animal Production Field, for a period of 35 days. A total of 240 ten-day-old Ross 308 broiler chicks (unsexed) were used in this study. The chicks were obtained from a private hatchery in Kirkuk and randomly assigned to four dietary treatments using a completely randomized design (CRD). Each treatment consisted of four replicates, with 15 birds per replicate.

Dietary treatments were the equationted as follows:

- **T1 (Control):** Basal diet without supplementation
- **T2:** Basal diet + 1 g/kg nutmeg powder
- **T3:** Basal diet + 2 g/kg nutmeg powder
- **T4:** Basal diet + 3 g/kg nutmeg powder

Nutmeg powder derived from *Myristica fragrans* was incorporated into the basal diet according to treatment levels. Birds were fed starter, grower, and finisher diets throughout the experimental period, as shown in Table 1. Feed and water were provided ad libitum, and birds were managed under standard commercial rearing conditions.

At 35 days of age, two birds were randomly selected from each treatment group for blood sampling. Blood samples were collected from the jugular vein into anticoagulant-free tubes. Samples were centrifuged at 3000 rpm to obtain serum, which was subsequently used for biochemical analyses.

Table 1. Proportions of feed ingredients and calculated chemical composition of the experimental diets.

S.No	Ingredient Name	Starter1	Grower2	Finisher3
1	Corn	531.000	523.000	554.000
2	Oil-Soyabean	6.000	8.500	18.000
3	Wheat Flour(Ard)	50.000	100.000	100.000
4	Soya DOC 46%	370.000	328.000	290.000
5	Birmix m 111 plus l str	25.000	0.000	0.000
6	Lime Stone(kis) 33%	18.000	15.500	13.000
7	Birmix m 112 plus l gro	0.000	25.000	25.000
	Total	1000.000	1000.000	1000.00
	Nutrient Menu			
S.No	**Nutrient Name**	**Starter1**	**Grower2**	**Finisher3**

1	Crude Protein (%)	22.5150	21.0290	19.5135
2	M.E (kcal/kg)	3005.3300	3051.1300	3150.9900
3	EE (%)	2.9956	3.2923	4.3064
4	Crude Fiber (%)	3.4988	3.2709	3.1142
5	Calcium (%)	1.0612	0.9327	0.8383
6	Available Phosphorus (%)	0.5141	0.4613	0.4549
7	Sodium (%)	0.1625	0.1635	0.1634
8	Chloride (%)	0.2077	0.2083	0.2077
9	Potassium (%)	0.9082	0.8431	0.7775
10	Dig. Lysine (%)	1.4558	1.2810	1.1828
11	Dig. Methionine (%)	0.6231	0.5801	0.5648
12	Dig. Meth+Cyst (%)	0.9348	08787	0.8492
13	Dig. Threonine (%)	0.7896	0.7154	0.6671
14	Choline (mg/kg)	1791.1920	1746.1560	1656.6280
15	Elec. Bal.[dEB(mEq/Kg)]	244.4188	228.0303	211.3881

a. PCV

To obtain packed cell volume, packed cell volume determination tubes were used, which were heparinized capillary tubes. These tubes were filled with blood to about two-thirds of their length, sealed with clay, and then centrifuged at 12,000 rpm for 5-10 minutes in a micro hematocrit centrifuge. The packed cell volume percentage was then measured using a micro hematocrit reader as described in [19].

b. Hb Concentration

Hemoglobin concentration was measured using a commercial diagnostic kit (Vitro Scient, Germany) according to the manufacturer's guidelines.

c. MCV

Mean corpuscular volume (MCV) was calculated according to [19] using the following the equation:

$$MCV\ (fL) = \frac{PCV}{Total\ Red\ Blood\ Cell\ Count}\ x\ 10$$

where fL = femtoliters (10^{-15} liters).

d. Total Cholesterol

Total cholesterol concentration was determined using the enzymatic hydrolysis method described by [16]. Cholesterol concentration was calculated as follows:

$$Cholesterol\ \ mg/100ml = \frac{Sample\ Absorbance}{Standerd\ Absorbance}\ x\ 100$$

e. Triglycerides

Triglyceride concentration was determined using the enzymatic method described by [17], according to the following the equation:

$$Triglycerides\ mg/100ml = \frac{Sample\ Absorbance}{Standerd\ Absorbance}\ x\ 100$$

f. (HDL)

HDL concentration was measured enzymatically according to the method of [18]. Concentration was calculated as:

$$HDL\ mg/100ml = \frac{Sample\ Absorbance}{Standerd\ Absorbance}\ x\ 50\ x\ 101$$

where 50 represents the standard solution concentration and 101 represents the dilution factor.

g. LDL

LDL concentration was calculated according to [19] using the following equation:

$$LDL = Total\ Cholesterol - (VLDL + HDL)$$

h. VLDL

VLDL concentration was estimated using the following equation:

$$VLDL = \frac{Triglycerides}{5}$$

2. Statistical Analysis

Data were analyzed using a Completely Randomized Design (CRD). Differences among treatment means were evaluated using Duncan's multiple range test [20] at a significance level of ($P \leq 0.05$) using SAS software [21].

The statistical model applied was:

$$Y_{ij} = \mu + T_i + E_{ij}$$

Where:

- Y_{ij}= observed value of the jth bird under the ith treatment
- μ= overall mean
- T_i= effect of dietary treatment
- E_{ij}= random error, normally distributed with mean zero and variance σ_e^2

Results and Discussion

The results obtained from the experiment and presented in Table 2 reveal that the dietary supplementation with nutmeg extract failed to show any significant differences ($P > 0.05$) in any of the hematological traits. Numerically varying results were obtained in this experiment. For example, the count of red blood cells (RBC) was maximum in T1 (control) with 12.01 x 10^6/mm^3 and minimum in T2 with 10.67 x 10^6/mm^3. For hemoglobin concentration (Hb), the maximum concentration was recorded in T4 (11.17 g dL^{-1}), followed by T3 (11.16 g dL^{-1}), while the minimum concentration was recorded in T1 (10.25 g dL^{-1}). For the determination of packed cell volume (PCV), T2 recorded the maximum (29.25%) and T3 recorded the minimum (24.76%). For mean corpuscular volume (MCV), T4 recorded the maximum (115.83 fL) and T3 recorded the minimum (113.18 fL). For mean corpuscular hemoglobin (MCH), T2 recorded the maximum (37.25 pg) and T1 recorded the minimum (31.69 pg). For mean corpuscular hemoglobin concentration (MCHC), T1 recorded slightly higher (35.40%) than the other treatments and T4 recorded the minimum (34.97%). These results reveal that the dietary supplementation with nutmeg extract at 3 g/kg body weight failed to show any significant effect on hematological traits.

Table 2. Effect of dietary nutmeg powder supplementation on hematological parameters of Ross 308 broiler chickens (mean ± SE)

Treatment	RBC (x10^6/mm^3)	Hb (g dL^{-1})	PCV (%)	MCV (fL)	MCH (pg)	MCHC (%)
T1	12.01 ± 0.12a	10.25 ± 0.26a	25.10 ± 1.47a	115.25 ± 0.88a	31.69 ± 1.45a	35.40 ± 0.60a
T2	10.67 ± 0.33a	10.27 ± 0.30a	29.25 ± 0.22a	113.56 ± 1.35a	37.25 ± 0.04a	35.35 ± 0.46a
T3	11.09 ± 0.54a	11.16 ± 0.54a	24.76 ± 1.19a	113.18 ± 1.42a	35.92 ± 0.15a	35.29 ± 0.42a
T4	11.23 ± 0.24a	11.17 ± 0.04a	27.90 ± 1.52a	115.83 ± 1.45a	33.32 ± 0.55a	34.97 ± 0.06a

The results obtained from the data presented in the table show that there is a significant effect ($P < 0.05$) of dietary nutmeg powder supplementation on the serum lipid profiles. The cholesterol concentration was the highest in the control treatment (T1) at 108.3 mg dL^{-1}. However, this concentration decreased significantly in the subsequent treatments and was lowest in T4 at 84.63 mg dL^{-1}. This effect may be attributed to the active compounds present in nutmeg powder, such as flavonoids, which have the ability to inhibit the activity of HMG-CoA reductase, the major hepatic enzyme that controls cholesterol synthesis from the conversion of β-hydroxy-β-methylglutaryl-CoA to mevalonic acid. These compounds also have the ability to influence cholesterol transport proteins to facilitate its excretion from the liver to the blood.

The triglyceride concentration was also highest in the control treatment (T1) at 129.74 mg dL^{-1} and decreased in the subsequent treatments. The lowest concentration was recorded in T4 at 89.74 mg dL^{-1}. This effect may also be attributed to the enhancement of lipid metabolism. This effect is also supported by the reduction in cholesterol concentration in the subsequent treatments. These results also show that the concentration of low-density lipoproteins (LDL) decreased from 57.73 mg dL^{-1} in T1 to 30.99 mg dL^{-1} in T4. This effect may be attributed to the antioxidant property of flavonoids that prevents LDL oxidation in addition to their therapeutic property in preventing atherosclerosis.

In contrast, the HDL level showed significant increases in all supplemented groups, with the highest level recorded for T4 at 59.54 mg dL^{-1} compared to 47.70 mg dL^{-1} for the control. HDL is known to be a good lipoprotein that plays a significant role in the transport of lipids from the peripheral tissues to the liver for metabolic activity. Therefore, the increase in HDL level indicates an improvement in the lipid profile. In addition, the level of VLDL was found to be the highest for T1 at 25.94 mg dL^{-1}, while the level for T4 was found to decrease to 17.94 mg dL^{-1}, similar to the decrease in triglycerides, as the two are metabolically related.

These findings can be attributed to the variety of bioactive compounds found in medicinal plants, which, when acting synergistically, produce favorable effects on the metabolic system compared to the use of single compounds, as shown for pharmaceutical drugs [22, 23]. In conclusion, dietary supplementation of the nutmeg powder was found to improve the lipid profile, as the cholesterol, triglyceride, LDL, and VLDL levels were found to decrease, while the HDL level increased[24].

Table 3. Effect of dietary nutmeg powder supplementation on serum lipid profile of Ross 308 broiler chickens (mean ± SE)

Treatment	Cholesterol (mg dL^{-1})	Triglycerides (mg dL^{-1})	HDL (mg dL^{-1})	LDL (mg dL^{-1})	VLDL (mg dL^{-1})
T1	108.30 ± 1.01a	129.74 ± 0.87a	47.70 ± 1.88b	57.73 ± 1.74a	25.94 ± 0.17a
T2	93.80 ± 1.49b	104.21 ± 1.70b	55.53 ± 1.78a	45.81 ± 1.92b	20.84 ± 0.34b
T3	88.78 ± 1.48bc	99.42 ± 0.76c	54.75 ± 1.50a	42.40 ± 1.84bc	19.88 ± 0.15c
T4	84.63 ± 1.19c	89.74 ± 0.77d	59.54 ± 0.62a	30.99 ± 0.84c	17.94 ± 0.15d

Conclusion

The results obtained from this study have shown that the addition of nutmeg powder in chicken feed did not have a significant effect on the hematological values in Ross 308 broiler chickens. These values include red blood cells (RBC), hemoglobin (Hb), packed cell volume (PCV), mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), and mean corpuscular hemoglobin concentration (MCHC). These values were not affected in any manner by the addition of nutmeg powder in chicken feed. However, the addition of nutmeg powder in chicken feed had a significant effect on the serum lipid profiles. Nutmeg powder addition in chicken feed at 2 and 3 g/kg resulted in a significant reduction in total cholesterol, triglycerides, LDL cholesterol, and VLDL cholesterol. Nutmeg powder addition in chicken feed also resulted in a significant increase in HDL cholesterol. Nutmeg powder addition in chicken feed can therefore be considered safe and effective in improving lipid metabolism in broiler chickens without any effect on their hematological values.

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