

Effects of *Boswellia Carterii* And *Boswellia Serrata* in Drinking Water on the Growth Performance, Hematology Traits and Immune Response of Broiler Chicken

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ABSTRACT

A total of 300 Ross 308 broiler chickens, 7 days old, comprised the subjects in this study. They were randomly distributed into 5 treatments of 3 replicates each (15 bird/rpt). *Boswellia* was ground and mixed in drinking water of chickens during the whole period of the experiment, which lasted for 35 days, at levels of 0g (T1: Control), 2 g *Boswellia carterii*/l liter water (T2), 3 g *Boswellia. carterii*/l liter water (T3), 2 g *Boswellia. serrata*/l liter water (T4), and 3 g *Boswellia.serrata* / l liter water (T5). Growth performance parameters which included body weight, weight gain, feed intake, feed conversion ratio, mortality and EBI were reported every week during the experiment. Moreover, three birds per replicate, 35 days old, were slaughtered to assay the hematological characteristics and immune response parameters. The results revealed that the fifth treatment (3gBs/l water) showed considerable improvement ($P<0.05$) in productivity traits (body weight, weight gain), hematological traits (RBC count, Hb concentration, PCV volume, total protein, globuline), and immune responses (Antibody titers against avian influenza virus, DTH test and bursa ratio) and decreased ($p>0.05$) feed intake, feed conversion ratio, albumin, glucose and cholesterol, followed by T4, T3, T2 respectively in comparison with control (T1). It was concluded that broilers that received *Boswellia* (BC, BS) especially T5 could have positively affected growth performance and well being of the broiler chickens.

INTRODUCTION

Medicinal plants and herbs have been used for centuries as traditional medicine for the healing of human ailments and as health promoters. It is therefore no surprising that in recent years the use of medicinal plants and herbs has grown in popularity due to their low side effects and the presence of a wide range of beneficial compounds, as has been recommended by the World Health Organization (WHO) [1].

The acceptance and use of alternative medicine is on the rise, especially in developing countries following the directives from the WHO, resulting in the publication of numerous pre-clinical and clinical studies that have given scientific credence to the efficacy of numerous plants and herbs used in traditional folk medicine to treat infections [2]. In this context therefore, vegetables, herbs, spices and edible plants have been suggested as non-traditional feed additive or growth promoters in broiler diets to enhance the growth feed conversion efficiency and also, minimize feed cost [3,4]. Furthermore, the application of herbal plants in broiler diets to promote growth has been observed to significantly improve broilers' body weight gain, reduce the mortality rate and significantly improve feed conversion [5]. Earlier research has shown that the supplementation of herbs in broiler diets has had

significant positive effect on production performance and carcass quality [6-8]. Besides, several countries have limited or prohibited the use of chemicals due to their negative side effects on both the animals and on humans. This is another steps towards the encouraging the use of natural supplements as growth promoters [9].

Boswellia, also known as frankincense or olibanum is an aromatic material that has been used through the ages as incense in Mesopotamia, Egypt, Greek and Rome. It comes from the resin of trees of the genus *Boswellia* (Burseraceae family), and mainly produced by four species, including *Boswellia serrata* in India, *Boswellia carterii* in East Africa and China, *Boswellia frereana* in Somalia and *Boswellia sacra* in Northern Africa and Arabia. Each of these produces a resin that is different due to variations in soil and climate. *Boswellia serrata* is an important component in Ayurvedic medicine as an antiarthritic, astringent, stimulant, expectorant, and antiseptic agent [10] while *Boswellia carterii* is primarily used in traditional Chinese medicine to ease pain and treat inflammation and for cancer [11]. The last 20 years have seen *Boswellia* gaining the attention of scientists and the pharmaceutical industry to better define its medical effects and establish the constituents responsible for these effects. As a result, there have been numerous researches that have reported on the anticancer, anti-inflammatory, immunomodulatory, antimicrobial and antiviral activities of various *Boswellia* species [12-19]. *Boswellia* resin is a mixture containing more than 200 different substances [20] for instance: resin, sugar, essential oils, proteins, and inorganic compounds [21]. *Boswellia carterii* and *Boswellia serrata* possess the active pentacyclic triterpene acids α - and β - boswellic acid, 11-keto- β -boswellic acid, acetyl-11-keto- β -boswellic acid, acetyl-11-dien- β -boswellic acid, acetyl- α -boswellic acid and acetyl- β -boswellic acid. Acetyl-11-keto- β -boswellic acid (AKBA) is known to be the main constituent in *Boswellia* [22,23]. The primary difference between resin *B. carterii* and *B. serrata* is the higher content of proteins (22%) for *B. serrata* than (6%) for *B. carterii* [24]. On the other hand, to our best knowledge, there have been no studies on the effects of *Boswellia* on broilers, and this current study was undertaken to determine the effect of supplementation of *Boswellia carterii* and *Boswellia serrata* on growth performance, Blood profile and immune responses in broiler chickens when used as supplements in drinking water.

MATERIALS AND METHODS

Animals and Dietary Treatments

A total of 300 day-old broiler chickens of mixed sex (Ross-308) were weighted and randomly allocated to five treatment groups, each with 3 replicates of 20 chicks. *Boswellia* was ground into powder and mixed with drinking water 12 hours before (to ensure solubility) and supplemented to the drinking water of chicks during the whole period of the experiment, which lasted 35 days, at levels of 0 g (T1; Control), 2 g Bc/ l liter water (T2), 3 g Bc / l liter water (T3), 2 g Bs/ l liter water (T4), and 3 g Bs / l liter water (T5). *Boswellia* (Bc, Bs) was added to drinking water from the age of 7 days and until the end of the experiment. The basal diet (**Table 1**) was formulated according to nutrient requirements of broilers recommended by National Research Council (NRC) [25]. From day 1 to day 14 the birds were fed a starter diet, and from day 15 to day 35. All the dietary treatments were added to the basal diets in place of corn. Chicks were raised on floor pens (120 cm x 120 cm x 80 cm) for 5 weeks and throughout the whole experimental period, there was free access to feed and water. The lighting regime was 23 h light followed by 1 h of darkness while ambient temperature in experimental house was a constant 32 °C for the first week and gradually lowered by 3 °C in weeks 2 and 3 before being fixed at 22 °C until the end of the experiment.

Table 1. Composition of basal diet.

| Item,% | Starter% | Grower% |
|---------------------------|-----------|---------|
| | 1 to 21 d | 22-35 d |
| Yellow corn, ground | 56.0 | 64.80 |
| Soybean meal (44%CP) | 29.0 | 23.65 |
| Corn gluten meal (60%CP) | 8.20 | 5.80 |
| Vegetable oil | 3.40 | 2.40 |
| Di - calcium phosphate | 1.80 | 1.30 |
| Calcium carbonate | 0.90 | 1.30 |
| Sodium chloride | 0.30 | 0.30 |
| Vit. and Min. premix * | 0.30 | 0.30 |
| DL - Methionine | 0.10 | 0.05 |
| L-Lysine | 0.00 | 0.10 |
| Total | 100 | 100 |
| Calculated analysis (%)** | | |
| CP Crude protein | 22.51 | 19.53 |
| EE Ether extract | 2.620 | 2.840 |
| CF Crude fibre, | 2.470 | 2.420 |
| Ca | 0.869 | 0.906 |
| Available P | 0.444 | 0.346 |
| Methionine | 0.515 | 0.411 |
| Methionine +Cystine | 0.944 | 0.781 |

| | | |
|--------------|-------|-------|
| Lysine | 1.088 | 1.010 |
| ME, K cal/Kg | 3109 | 3150 |

Each 3.0 kg of the Vit. and Min. premix contains : Vit. A, 12000000 IU; Vit. D3 2500000. IU; Vit. E, 10 g; Vit. K, 2.5 g; Vit. B1, 1.5 g; Vit. B2, 5 g; Vit. B6, 1.5 g; Vit. B12, 10 mg; Choline chloride, 1050 g; Biotin, 50 mg; Folic acid, 1 g; Nicotinic acid, 30 g; Ca pantothenate, 10 g; Zn, 55 g; Cu, 10 g; Fe, 35 g; Co, 250 mg; Se, 150 mg; I, 1 g; Mn, 60 g and anti-oxidant, 10 g.

**According to NRC [25].

Performance

Body weights of broilers were recorded at 14, 21, 28, and 35 days of age. Feed intake and weight gain were documented in different periods, and FCR was computed. Mortality was accordingly documented as it occurred and adjustments made on the basis of the total number of birds to determine the total feed intake per bird and FCR.

Blood Parameters

The prepared emulsions were fish/algal oil with antioxidants in combinations transformed into microencapsulated powder using the table top spray drier equipped with an anti-blocking device operated under the influence of compressed air (to avoid the blockage during spray drying) (M/s. Basic Technologies Pvt. Ltd., Kolkata) under standard conditions. The emulsion was sprayed into the chamber at a pressure of 2 kg/cm² (30 psi) through a one way nozzle (0.8 mm). The inlet and outlet temperature was maintained at 160 °C and 70 °C respectively. The microencapsulated powder (5g) each was filled in brown plastic containers and stored in refrigerator for 7 days.

Immune Responses

Humeral immunity, newcastle disease antibody titer: The commercially available oil-adjuvant injectable emulsion against Newcastle disease virus (NDV) was used to vaccinate broiler chicks. Injection was administered subcutaneously with 0.2 mL per chick at 9 day of age. Antibody titers against NDV, and relative weight of lymphoid organs (spleen, bursa of fabricius) were recorded as immune responses. At 35 days of age, three male broilers from each pen were randomly selected, and blood samples collected by puncturing the brachial vein and centrifuging the blood at 2000×g for 10 min. Then, the serum was collected and kept at -20 °C until needed for analysis of antibody titers against NDV. Serum antibody titers against NDV were recorded using the hemagglutination inhibition test [26]. Hemagglutination inhibitions were then converted to log. Spleen and bursa were collected, weighed and computed as a percentage of live body weight after slaughtering three birds per replicate.

Delayed Type hypersensitivity (DTH) test: On day 35, 10 chickens from each group were injected with 100 µg/0.1 mL PHA antigen (Phytohemagglutinin PHA-P, Sigma) in PBS (Phosphate-Buffered Saline) Buffered Saline in the right wattle. The extent of swelling at the injection site was recorded after 24 h with a micrometer [27].

Statistically Analysis

The approach adopted was a completely randomized experimental design with the data statistically analyzed employing the General Linear Models (GLM) procedure of SAS software [28] for analysis of variance [29]. Multiple range test was used to establish the variances among treatment means, which were considered different at p<0.05 and p<0.01.

RESULTS

Performance

Body weight (BW)

The average live body weight at the periods of 14, 21, 28 and 35 d are shown in **Table 2**. Chickens that received two different levels of BC, BS in their drinking water showed no significant differences in BW at 14 days of age. However, the chicks that received 3 g Bs/l water showed significant differences (p<0.05) in BW at 14, 21,28 and 35 days of age in comparison with other treatments except T4, which did not differ from T5 at 21d. There were no significant differences of BW observed between T3, T4 at 28 and 35 d also between T1, T2, T3 at 21 d., but T1 was significant differences with T2 at 28, 35 d.

Table 2. Performance parameters of the broiler drinking water different levels of *Boswellia* (Bc, Bs) (Mean and SE).

| Parameters | Day | Treatments | | | | |
|------------|-----|------------------------------|------------------------------|------------------------------|------------------------------|------------------------------|
| | | T1 | T2 | T3 | T4 | T5 |
| BW (g) | 14d | 328.83 ± 1.67 | 333.14 ± 2.09 | 332.97 ± 1.87 | 329.91 ± 1.90 | 330.77 ± 2.01 |
| | 21d | 725.73 ± 7.01 ^c | 728.35 ± 6.13 ^c | 734.64 ± 6.24 ^{bc} | 735.92 ± 5.99 ^a | 746.33 ± 6.86 ^a |
| | 28d | 1169.90 ± 10.02 ^d | 1191.22 ± 9.66 ^c | 1204.76 ± 11.23 ^b | 1207.14 ± 10.56 ^b | 1223.06 ± 10.78 ^a |
| | 35d | 1700.30 ± 13.57 ^d | 1766.43 ± 13.70 ^c | 1785.11 ± 13.88 ^b | 1791.23 ± 14.46 ^b | 1832.68 ± 14.17 ^a |

| | | | | | | |
|--------------------|-----------------------------|-------------------------------|-------------------------------|-------------------------------|--------------------------------|-------------------------------|
| BWG(g) | 14d | 212.83 ± 2.29 | 213.08 ± 1.72 | 211.61 ± 2.06 | 214.25 ± 1.15 | 211.28 ± 1.20 |
| | 21d | 396.95 ± 2.29 ^c | 395.21 ± 1.72 ^c | 401.67 ± 2.06 ^{bc} | 406.01 ± 1.15 ^b | 415.56 ± 1.20 ^a |
| | 28d | 444.17 ± 3.26 ^c | 462.87 ± 3.17 ^b | 470.12 ± 2.84 ^a | 471.22 ± 2.97 ^a | 476.73 ± 2.83 ^a |
| | 35d | 530.40 ± 5.01 ^d | 570.21 ± 5.13 ^c | 580.35 ± 4.89 ^b | 584.09 ± 4.74 ^b | 609.62 ± 4.30 ^a |
| | 1-35 | 1371.52 ± 11.03 ^e | 1433.29 ± 12.15 ^c | 1452.14 ± 12.44 ^b | 1461.32 ± 12.68 ^b | 1501.91 ± 11.77 ^a |
| FI (g) | 14d | 333.53 ± 29.17 | 332.39 ± 30.55 | 333.50 ± 30.71 | 335.43 ± 27.88 | 334.09 ± 29.72 |
| | 21d | 653.87 ± 61.12 | 654.74 ± 60.31 | 657.23 ± 59.98 | 660.02 ± 61.18 | 665.34 ± 61.36 |
| | 28d | 773.60 ± 70.19 ^b | 780.280 ± 70.31 ^{ab} | 785.640 ± 71.56 ^a | 786.11 ± 71.34 ^a | 788.52 ± 71.46 ^a |
| | 35d | 991.85 ± 90.55 ^c | 1006.38 ± 94.18 ^b | 1007.13 ± 92.48 ^b | 1012.59 ± 90.25 ^b | 1017.40 ± 92.30 ^a |
| | 1-35d | 2752.67 ^c ± 112.07 | 2773.79 ^b ± 145.08 | 2783.50 ^b ± 109.45 | 2794.15 ^{ab} ± 127.59 | 2805.44 ^a ± 150.43 |
| FCR | 14d | 1.57 ± 0.01 | 1.56 ± 0.02 | 1.58 ± 0.02 | 1.57 ± 0.01 | 1.58 ± 0.02 |
| | 21d | 1.65 ± 0.02 | 1.65 ± 0.01 | 1.64 ± 0.03 | 1.63 ± 0.03 | 1.60 ± 0.02 |
| | 28d | 1.74 ± 0.02 ^a | 1.69 ± 0.03 ^{ab} | 1.67 ± 0.03 ^b | 1.67 ± 0.04 ^b | 1.65 ± 0.02 ^b |
| | 35d | 1.87 ± 0.03 ^a | 1.76 ± 0.02 ^b | 1.74 ± 0.03 ^b | 1.73 ± 0.02 ^b | 1.67 ± 0.01 ^c |
| | 1-35d | 1.71 ± 0.05 ^a | 1.67 ± 0.03 ^{ab} | 1.66 ± 0.04 ^b | 1.65 ± 0.03 ^b | 1.63 ± 0.03 ^b |
| Mortality (%)1-35d | 1.67 ± 0.72 ^a | 5.00 ± 0.39 ^a | 3.33 ± 0.26 ^b | 3.33 ± 0.21 ^b | 1.67 ± 0.13 ^b | |
| EBI | 265.14 ± 24.11 ^d | 287.10 ± 20.18 ^c | 297.02 ± 25.22 ^b | 299.85 ± 24.58 ^b | 315.88 ± 21.79 ^a | |

Mean values with (a-d) superscript in row differ significantly at (p<0.05).

T1 served as control while T2, T3, T4 and T5 were kept on drinking water containing T2: 2 g (Bc), T3: 3 g (Bc), T4: 2 g (Bs), T5: 3 g (Bs).

Values (Mean ±SD) of each experimental day in each row followed by different letters differ significantly (p<0.05, p <0.01).

Body weight gain (BWG)

Table 2 shows that different levels of Bc, Bs added to drinking water made no significant differences in BWG at 14 days of age. Chicks that received 3 g Bs/l water) showed higher BWG (p<0.05) than other treatments during the period 21 to 35 days of age. However, there were no significant effects of BWG between T5 and T3, T4 at 28 d. but the highest value (p<0.01) of BWG was recorded in T5 (1501.91 g) at the total period of 1 to 35d.

Feed Intake (FI)

Data presented in **Table 2** shows no significant effects on FI for the period from 14 to 21 days of age. Birds receiving 3 g Bs/l in drinking water had higher FI (p<0.05) during the period of 28 to 35 days of age (788.52, 1017.40, respectively). However, the lowest FI was in control treatment (773.6, 991.85, respectively) during the period from 28 to 35 days of age. Whereas, no significant differences in FI among chicks that received 2,3 g Bc /l, 2 g Bs /l in drinking water from 28 to 35 days of age.

Feed Conversion ratio (FCR)

Table 2 shows no significant effects on FCR during the period from 14 to 21 days of age. Control treatment had higher FCR (p<0.05) during the period 28 to 35 days of age (1.74, 1.87, respectively). Nevertheless, the lowest FCR was in birds that received 3 g Bs/l in drinking water (1.65, 1.67, respectively) for the period from 28 to 35 days of age. Whereas, no significant differences in FCR for birds that received 2,3 g/l 3 g/l BC, 2 g/l BS in drinking water from 28 to 35 days of age.

Mortality percentage

Cumulative mortality percentage was calculated during the starting and finishing periods and presented in **Table 2**. Obtained results suggest that the percentage of mortality in T1, T2 was higher than in treatments T3, T4, T5 and statistically significant (p<0.05).

Economic Efficiency index (EBI)

Table 2 shows that EBI value at 35 days of age had a higher value (315.88) in birds that received 3 g Bs/l in drinking water in comparison with control, which had the lowest value (265.14), whereas, the other treatments T2, T3, T4 showed higher value (287.10, 297.02, 299.85, respectively) than control but lower than T5.

Blood Profile

The overall mean packed cell volume (PCV), Hemoglobin (Hb), total erythrocytes count (RBC) are given in **Table 3**. As can be seen from the table, mean PCV, Hb, RBC values were reduced significantly (p<0.05) in T1, T2, T3 and T4. However, PCV, Hb and RBC were significantly (p<0.05) increased in the birds which were offered drinking water supplemented with 3g Bs (T5) compared

to control treatment and other treatments, except T4, which did not show any significant differences with T5 in PCV.

Table 3. Haematological parameters of the broiler drinking water different levels of Boswellia (Bc, Bs) (Mean and SE).

| Parameters | Treatments | | | | |
|---|-----------------------------|------------------------------|-----------------------------|-----------------------------|-----------------------------|
| | T1 | T2 | T3 | T4 | T5 |
| PCV (%) | 3.110 ^c ± 29.53 | 2.560 ^{bc} ± 29.89 | 3.390 ^{ab} ± 30.18 | 3.220 ^a ± 30.29 | 2.160 ^a ± 30.87 |
| Hb (g/dl) | 5.914 ± 0.5780 ^d | 6.228 ± 0.6600 ^c | 6.513 ± 0.7870 ^b | 6.580 ± 0.6130 ^b | 6.893 ± 0.5440 ^a |
| RBC (x10 ³ / mm ³) | 2.853 ± 0.3430 ^d | 2.967 ± 0.4220 ^{cd} | 3.22 ± 0.2470 ^{bc} | 3.243 ± 0.3210 ^b | 3.466 ± 0.1780 ^a |

Mean values with (^{a-d}) superscript in row differ significantly at (p<0.05).T1 served as control while T2, T3, T4 and T5 were kept on drinking water containing T2: 2 g (Bc), T3: 3 g (Bc), T4: 2 g (Bs), T5: 3 g (Bs).

Values (Mean ± SD) of each experimental day in each row followed by different letters differ significantly (p<0.05). PCV: Packed cell volume, Hb: Haemoglobin, RBC: Red Blood Cell.

Blood Serum Constituents

Different biochemical parameters (total serum cholesterol, protein, albumin, globulin, glucose) are given in **Table 4**. The total serum cholesterol, albumin and glucose values decreased significantly throughout the experimental period in birds which were offered drinking water supplemented with BC, BS in comparison with control, which had high values in three qualities. However, the lowest values of cholesterol, albumin and glucose was recorded in T5 (3 g Bs/l water), whereas, total protein and globulin significantly (P<0.05) increased in the birds which were offered drinking water supplemented with 3 g Bs/l water compared to other treatments and control during the total period. However, the lowest value was recorded in control.

Table 4. Blood serum parameters of the broiler drinking water different levels of Boswellia (Bc, Bs) (Mean and SE).

| Parameters | Treatments | | | | |
|---------------------|------------------------------|------------------------------|------------------------------|------------------------------|------------------------------|
| | T1 | T2 | T3 | T4 | T5 |
| Cholesterol, mmol/L | 105.18 ± 9.330 ^a | 97.56 ± 10.020 ^b | 94.7 ± 8.450 ^c | 93.89 ± 9.340 ^c | 87.47 ± 10.560 ^d |
| Total protein, g/L | 5.875 ± 0.700 ^d | 6.246 ± 0.660 ^c | 6.518 ± 0.590 ^b | 6.543 ± 0.740 ^b | 6.988 ± 0.630 ^a |
| Albumin g/L | 1.656 ± 0.020 ^a | 1.593 ^b ± 0.030 | 1.544 ± 0.020 ^c | 1.521 ± 0.030 ^c | 1.437 ± 0.010 ^d |
| globuline g/L | 3.91 ± 0.530 ^d | 4.73 ± 0.460 ^c | 5.08 ± 0.400 ^b | 5.29 ± 0.520 ^b | 5.74 ± 0.330 ^a |
| Glucose (mg/dl) | 340.37 ± 14.420 ^a | 298.55 ± 12.890 ^b | 226.65 ± 13.530 ^c | 207.22 ± 15.300 ^c | 193.17 ± 12.240 ^d |

Mean values with (^{a-d}) superscript in row differ significantly at (p<0.05).T1 served as control while T2, T3, T4 and T5 were kept on drinking water containing T2: 2 g (Bc), T3: 3 g (Bc), T4: 2 g (Bs), T5: 3 g (Bs).

Values (Mean ± SD) of each experimental day in each row followed by different letters differ significantly (p<0.05).

Immune Responses

Table 5 shows the effects of treatments on immune response-related parameters. Antibody titer against NDV showed a significant (p<0.05) increase in birds, which were given drinking water supplemented with 3 g Bs/l water compared to other treatments and control. Control treatment showed a significant (p<0.05) decrease in Newcastle disease antibody titer in comparison with other treatments. DTH test indicated increased (p<0.05) values when broilers were given drinking water supplemented with 3 BS/l water compared with other treatments. Control treatment and T2 (2 g BC/l water) had lower (p<0.05) values in DTH test in comparison other treatments. Furthermore, the lymphoid organs which are related to immune response are shown in **Table 6**. According to **Table 6** the weight of the spleen did not differ significantly as a result of the dietary treatments, although bursa weights (%) were observed to be significantly (p<0.05) higher in chickens supplemented with T5 (3 g Bs/l water) compared to other treatments.

Table 5. Effect of *Boswellia* (Bc, Bs) in drink water on immune response of broilers at 35d (Mean and SE).

| Parameters | Treatments | | | | |
|--|-----------------------------|-----------------------------|-----------------------------|-----------------------------|-----------------------------|
| | T1 | T2 | T3 | T4 | T5 |
| Newcastle disease antibody titer(Log2) | 3.49 ± 270.80 ^d | 3.53 ± 282.50 ^c | 3.56 ± 313.30 ^b | 3.56 ± 307 ^b | 3.57 ± 242.1 ^a |
| DTH1test(mm) | 0.389 ± 0.0230 ^d | 0.446 ± 0.0260 ^c | 0.494 ± 0.0240 ^b | 0.502 ± 0.0310 ^b | 0.541 ± 0.0220 ^a |

Mean values with (^{a-d}) superscript in row differ significantly at (p<0.05).T1 served as control while T2, T3, T4 and T5 were kept on drinking water containing T2: 2 g (Bc), T3: 3 g (Bc), T4: 2 g (Bs), T5: 3 g (Bs).

Values (Mean ± SD) of each experimental day in each row followed by different letters differ significantly (p<0.05). ¹DTH: delayed type hypersensitivity test.

Table 6. Effect of *Boswellia* (Bc, Bs) in drink water on lymphoid organs on day 35(Mean and SE).

| Parameters | Treatments | | | | |
|------------|-----------------|-----------------|-----------------|-----------------|-----------------|
| | T1 | T2 | T3 | T4 | T5 |
| Spleen* | 0.13 ± 0.01 | 0.12 ± 0.01 | 0.11 ± 0.02 | 0.12 ± 0.01 | 0.11 ± 0.01 |
| Bursa* | 0.069 ± 0.001 c | 0.075 ± 0.003 c | 0.088 ± 0.002 b | 0.090 ± 0.001 b | 0.102 ± 0.002 a |

Mean values with (^{a-c}) superscript in row differ significantly at (p<0.05).T1 served as control while T2, T3, T4 and T5 were kept on drinking water containing T2: 2 g (Bc), T3: 3 g (Bc), T4: 2 g (Bs), T5: 3 g (Bs).

Values (Mean ± SD) of each experimental day in each row followed by different letters differ significantly (p<0.05). * Percentage of live body weight.

DISCUSSION

Effect of *Boswellia* (BC, BS) on Broiler Performance

Boswellia carterii and *Boswellia serrata* positively affected body weight, weight gain, feed intake and E.B.I, especially among broilers that received 3 g Bs/l in drinking water through the experimental period. These findings may be attributed to the presence of boswellic acid which contains phenolic acid and is known to improve enhance protein and energy by decreasing microbial competition in the host for nutrients of subclinical infections and secretion of immune mediators and also by a decrease in the production of ammonia, as revealed in studies by Chevrier et al. [22] and Dibner and Buttin [30]. In this context [31] indicated that presence *Boswellia* in a digestible system is considered as a stimulant, gas repellent, an catalyst for appetite, and it increases the flow the digestive juices and thus leads to improvement process of digestion and absorption because it stimulates the secretion of pancreatic enzymes. These results are in harmony with [32] who reported that rats diabetic treated with BC for 30 days led to significant increase in body weight, liver glycogen (P>0.01). Contrary results by Pooja et al. [33] found that rats treated with a high dose (1000 mg BS/kg B. wt. /day) had a considerable reduction in body weight gain. In the current study, FCR had an important (p<0.05) effect on control in comparison with the other treatments. These results are inconsistent with Tabatabaei et al. [34] who reported that FCR in birds treated with 0.5% BS was higher than broilers fed in control during the grower period, but the best FCR was in birds treated with 0.15 BS for the total period whereas, the differences were not statistically significant. *Boswellia* effect on broilers' performance observed in this study is in line with previous findings that reported dietary supplementation with extracts of some plants enhanced growth performance and feed efficiency in broilers [35-37]. In addition, Khalaji et al. [38] suggested that plant extracts could enhance nutrient absorption due to greater villi length and crypt depth.

Effect of *Boswellia* (BC, BS) on Serum Biochemical Profiles

Serum biochemical and heamatological profiles could be beneficial measures for the assessment of the safety or monitoring of harmful effects of boswellia on broilers health. Determining the impact of these boswellia (BC, BS) on the concentration of total protein, globuline, albumin, cholesterol and glucose can be achieved by evaluating the blood of the test subjects [39]. The effect of inclusion of 2, 3% BC and 2, 3% BS in drinking water for 6 weeks on broiler chickens was a significant rise in the serum total protein concentration in T5 in comparison with control (**Table 4**), while the rest of the treated groups also showed significant increase in comparison with the control. These outcomes are opposed to those of Pooja et al. [33] who indicated that rats treated with *Boswellia serrata* at three different doses 100, 500 and 1000 mg/kg B.wt./day exhibited no changes in total protein in comparison with the control group animals. Also, Sakuntala et al. [40] indicated that using 100, 500 and 1000 mg/kg respectively of alcoholic extract of *Boswellia ovalifoliolata* orally for 28 days did not result in any significant difference in total protein between the treated and control groups. However, the results in the present study, similar to those of Zhang [41] who discovered that the concentration of total protein in serum of ginger-supplemented 5 g/kg of broiler diet was higher at 21 days and 42 days of age in comparison with that of control broilers. Thus, it can be suggested that elevated total serum protein level could be attributed to high level of protein and other nutrients in boswellia. This could be in line with the findings of Hoffman [42], who indicated that serum protein levels can be affected by nutritional influences. Notably the apparent higher total protein in serum blood for chickens treated with boswellia, may also have been responsible for an improved rate of BW and BWG [43].

The level of serum globulin showed significant (p<0.05) increase in groups that received 3 g Bs/l in drinking water compared to control, whereas, the other treatments with boswellia had also shown increasing significance compared with the control. The level of globulin was increased by adding experimental additives, which may indicate enhanced immunity for the birds. These results agree with those of AL-Homidan [44], who stated that the chicks fed *Zingiber officinale* at 2% for 7 weeks of the diet, were observed to have a higher value in the serum globulin concentration in comparison with the control broilers. The researcher attributed this increase of the serum globulin to the immunostimulant effect of *Zingiber officinale*. However, further studies on immunological parameters in boswellia for broilers are needed [45].

As presented in **Table 4**, the level of albumin indicates a significant (p<0.05) increase in the control group compared to the other treatments which showed significant decrease. These results may be indication of normal liver function from supplementation

of boswellia compare to the control group. The findings of this study are in agreement with those of Ademola et al. [46] who reported that broilers that received different levels of garlic and ginger (5, 10,15 g/kg 10,15 g/kg diet) showed a decrease in albumin concentration in treated groups compared to chicks used as control. Similar results were produced by Mehdi et al. [47], who noticed that birds fed 4 g/kg cinnamon were not affected by the dietary treatments in their serum albumin. Likewise, Ali et al. [48], discovered that a diet of 0.2% *Cuminum cyminum* had no effect on chicks' plasma albumin concentration.

The level of total cholesterol showed a considerable increase in the control group while in comparison the lowest value was in the group that received 3 g Bs/l in drinking water. The other treated groups showed significant decrease in comparison with control. Our results were in agreement with those of Pandey et al. [49] who reported that the supplementation of *Boswellia serrata* gum resins extract 15 mg/100 g body wt for 90 days led to a reduction in serum cholesterol and increased HDL in rats. Similar results were obtained by Hazim et al. [50] who observed that birds treated with *Boswellia carterii* at levels of 0.5, 0.75 and 1 g/l in drinking water had significant ($p < 0.05$) decrease in cholesterol, triglycerides and LDL. This suggests the possibility that *Boswellia serrata* supplementation restores β -cells function for insulin secretion, and that insulin helps to reduce serum lipid profiles [51]. Furthermore, Altman, et al. [14] mentioned that *Boswellia carterii* may have direct protective effect on β cells through its antioxidant action.

The blood-glucose levels showed significant ($p < 0.05$) increase in the control group whereas the lowest value was in the group that received 3 g Bs/l water in drinking water in comparison with the control. However, the remaining treated groups showed significant decrease in comparison with control. Similarly, Hazim et al. [50], reported that drinking water of broiler chickens supplemented with different levels of *Boswellia carterii* powder caused significant decrease in blood plasma concentrations of glucose, at levels 0.5, 0.75 and 1 g/l. However, 0.75, 1 g/l water supplementation led to lower values of blood plasma concentrations of glucose. Ahangarpour et al. [52] also reported that 6 weeks supplementation of *Boswellia serrata* to type 2 diabetic patients, produced a very significant decrease in fasting blood glucose and an increase in insulin level. Similarly, aqueous extract of *Boswellia glabra* (*B. glabra* Roxb. (Also known as *B. serrata*) increased the synthesis of secretory granules in the beta-cell, and led to an increase in pancreatic enzyme resulting in reduced blood-sugar level [53].

Effect of Boswellia (BC, BS) on Hematological Parameters

From the results of hematological parameters, the indication is that red blood cells, packed cells volume and Hb have higher value ($p < 0.05$) in T5 (3 g Bs/l water) than T4 and T3 compared with the control group (T1). These indicate that with the administration of *Boswellia serrata*, animals showed an improvement in major haematological parameters. It may be concluded that the *Boswellia serrata* has a positive effect on the health of animals. The results of this study concur with those of Sturikie et al. [54] who reported that improvements in digestion and absorption are accompanied by an increase of red blood cells because they are responsible for transporting food to the various cells of the body. However, our results are contrary to those of Pooja et al. [33] who observed that rats treated with *Boswellia serrata* with three different doses of 100, 500 and 1000 mg/kg/day orally did not show any changes in hematological parameters like WBC count, RBC count, hematocrit, and platelet count when compared with the control group with the exception of hemoglobin, which showed slightly higher values in the high-dose group. Likewise, Sakuntala et al. [40] found that rats treated with doses of 100, 500 and 1000 mg/kg respectively in alcoholic extract of *Boswellia ovalifoliolata* orally for 28 days showed no significant difference compared to the treated groups and control group in terms of the hematological parameters [54].

Effect of Boswellia (BC, BS) on Immune Response

In immune response, boswellia affected the antibody titer against NDV, delayed type hypersensitivity (DTH), and relative weight of lymphoid organs (bursa) compared with control. It is unfortunate that to our best there have been no other available reports regarding the effects of boswellia (BC, BS) on birds' immunity. However, Sharma et al. [55] reported that rats treated with BAs at 25-100 mg/kg/day for 21 days showed increased body weight, total leukocyte counts, and primary and secondary antibody titers. Treatment with doses beyond 50 mg/kg/day, caused reduction of polymorphonuclear (PMN) and an increase in lymphocyte population. However, Chevrier et al. [22] found that *Boswellia carterii* dissolved in sesame oil in concentrations of 10 μ g/ml to 50 μ g/ml is immunomodulatory rather than immunosuppressive, as these concentrations simultaneously inhibit TH1 cytokine production and promote TH2 cytokine production. In the present study antibody titer against NDV and DTH tests was of higher value ($p < 0.05$) in treatment T5 (3 g Bs/l water) and followed, in effectiveness by the other treated groups when broilers were offered drinking water supplemented with boswellia compared to control. Bursa weights were observed to be of higher value ($p < 0.05$) for chickens which were offered drinking water supplemented with 3 gBs/l water and followed in effect (2 g Bs/l water) (3 g BC/l water) compared to control. As boswellia has been reported to have antifungal [56], antimicrobial [57], Antioxidant Activities [58], Anti-Inflammatory [59] and immunomodulatory [22], broilers have more resistance against diseases with larger bursa of fabricius [60]. Broilers have higher synthesis of immunoglobulins with larger bursa of fabricius [61] and higher antibody titers with birds having larger bursas [62]. Our results concur with those of Saeed et al. [63] who used different levels of chicory leaf extracts in drinking water and exhibited significant effect ($P < 0.05$) on Newcastle disease (ND) titers. Similarly, birds treated with different levels of chicory extract were observed to have higher antibody titers. Aqueous extract of ginger rhizome mixed in water gives better performance as an immune stimulant against ND [64]. In the humoral defense system, a mixture of BAs at lower doses enhanced secondary antibody titers and in the cellular defense boswellic acids increase lymphocyte proliferation [65]. In contrast, *Boswellia serrata*

extract (BES) and constituents of BES reduce the cellular activity of the immune system by inhibiting the activation, proliferation and differentiation of B- and T- lymphocytes (IL-1, IL-2, IL-4, IL-6), tissue destruction (IL-1), action of NK-cells (IL-12), fever (IL-1, IL-6) and antibody production (IL-6) ^[66].

The delayed type hypersensitivity (DTH) test is a way to evaluate cell-mediated immunity *in vivo*. The study observed that DTH test was improved by boswellia (BS, BC) treatments, especially T5 (3g BS/l). Our finding is similar to that of Gayathri et al. ^[67] who indicated reported that *Boswellia serrata* extract exhibited anti-inflammatory in human peripheral-blood mononuclear cells (PBMCs) and mouse macrophages by inhibiting tumor necrosis factor-alpha (TNF-alpha), interleukin-1beta (IL-1beta) Banno et al. ^[68] also reported that boswellic acid extracts from *Boswellia carterii* have been shown to inhibit the production of inflammatory mediators, reduce the production of leukotrienes through its inhibitory action on lipoxigenase.

CONCLUSION

In conclusion, drinking water supplementation with different levels of boswellia (Bc,Bs) resulted in significant improvement to most of the rearing efficiency included within this study. Therefore, T5 (3 g Bs/l water) followed by other treatments with boswellia can be used as an efficient feed additive for the enhancement of general productivity traits and health status of birds.

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