EFFECTS OF TULSI LEAF (*Ocimum sanctum*) EXTRACT ON GROWTH PERFORMANCE AND HEMATOLOGICAL PARAMETERS IN BROILER

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Abstract

We conducted a study to evaluate the effect of tulsi leaf (*Ocimum sanctum*) extract supplementation in drinking water as a growth promoter in the broiler. A total of 50 Cobb-500 broiler chicks (day-old) were purchased from a local hatchery (CP Bangladesh Co. Ltd). After seven days of acclimatization, chicks were randomly divided into two groups:- T_0 (n=25) and T_1 (n=25). Group T_0 was kept as a control and untreated. Group T_1 was supplemented with tulsi leaf extracts 0.02% with tap water. Weekly observations were recorded for live body weight gain up to 4 weeks and hematological tests were performed at the broiler's 21st and 28th day's age. The initial body weight of groups T_0 and T_1 on 1st day of this experiment were 48.43±0.61 g and 47.77±0.22 g, respectively, and after the 28th day of the experiment, final body weights were 1386.62±18.36 g, and 1476.25±12.21 g, respectively and net profit per broiler was Tk. 18.65 and Tk. 23.09, respectively. The live body weight of the treatment group (T_1) was increased significantly ($p<0.01$) than that of the control group (T_0). The treatment group T_1 was recorded statistically significant (at 5% level) increased (6.935) live body weight than that of control group T_0. The hematological parameters total erythrocyte count (TEC), packed cell volume (PCV), and hemoglobin (Hb) estimation value of the treatment group was increased significantly ($p<0.01$), and total leukocyte count (TLC) was increased significantly ($p<0.05$). In contrast, hemoglobin (Hb) estimation showed significant difference from the control group. The results suggest that a broiler supplemented with 0.02% tulsi leaf extract could achieve better growth performance.

Keywords: Tulsi leaf extract, Growth performance, Hematological parameters, Cost-benefit analysis in broiler.

Introduction

The poultry production systems led to a marked increase in the production of poultry meat and eggs throughout the world (Ashayerizadeh *et al.,* 2009). It has triggered the discovery and widespread use of several "feed additives". The term feed additives are applied broadly to all products other than those commonly called feedstuffs, which could be added to the ration to obtain some special effects. The main objective of adding feed additives is to boost animal performance by increasing their growth rate, better feed conversion efficiency, greater livability, and lowered mortality in poultry birds. These feed additives are termed "growth promoters" and are often called non-nutrient feed additives (Singh *et al.,* 2003). Many synthetic drugs and growth promoters are supplemented to the broilers to effect rapid growth, but their use has shown many disadvantages like, high cost, adverse side-effect on the health of birds and long residual properties, etc. (Bhujbal *et al.*, 2009). Growth promoters are chemical and biological substances added to livestock food and, in this way, realize better production and financial results. European Commission banned four commonly used feed antibiotics: monensin sodium, salinomycin sodium, avilamycin, flavophospholipol (Banerjee, 1998). The banning of antibiotic growth promoter (AGP) will affect the poultry and livestock industry (Ashayerizadeh *et al.*, 2009). To minimize the loss in growth, there is a need to find an alternative to AGP. There are several non-therapeutic alternatives such as enzymes, inorganic acids, probiotics, prebiotics, and herbs (Banerjee, 1998). Since ancient times herbs and their essentials have been known for their varying degree of antimicrobial activity (Juven *et al.*, 1994). More recently, medicinal plants extracts were developed and proposed for use in food as natural antimicrobials (Hsieh *et al.*, 2001). Tulsi has attracted worldwide prominence due to its vast range of medicinal properties without showing any adverse effects.

Tulsi also promotes bird's growth and feed efficiency because of their antibacterial properties (WHO, 1997). A decoction of tulsi leaf is a popular remedy for cold. Tulsi leaves also check for vomiting and have been used as

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anthelmintic (Bhargawa *et al.,* 1996). Tulsi also possesses antifungal activity against *Aspergillus niger*. Aqueous tulsi extract is effective in patients suffering from viral encephalitis (Nath *et al.*, 2012). Aqueous decoction of the whole plant lowers the blood sugar (glucose) level and is said to control Diabetes mellitus (Khanna and Bhatia, 2003).The investigation was, therefore, designed to study the hematological effects of tulsi leaf extract in the broiler, to establish the effect of tulsi leaf extract as a growth promoter and its safety in broiler chickens. The main objective of adding feed additives is to boost animal performance by increasing their growth rate, better-feed conversion efficiency, greater livability, and lowered mortality in poultry birds.

Materials and Methods

Study area

The present research work was undertaken with the collaboration of a local poultry farm and the Department of Pharmacology and Toxicology, Faculty of Veterinary, Animal and Biomedical Sciences, the Sylhet Agricultural University, Sylhet, Bangladesh.

Study period

This experiment was carried out from July through December 2018 at the Department of Pharmacology and Toxicology, Sylhet Agricultural University, Sylhet.

Selection of experimental chicks

Fifty commercials, unsexed, straight run day old, Cobb-500 broiler chicks from a single batch were purchased from a commercial hatchery, Habiganj, Sylhet. These chicks were weighed individually and distributed into two groups viz T_0 and T_1: the control group and the treatment group. Each group consists of 25 chicks.

Management

Housing and equipment

Before the start of the experiment, the poultry house, waterers and feeders were cleaned, washed, disinfected, and kept ready. The poultry house was disinfected with the help of lime-stone before the arrival of the chicks. The room where the experiment was conducted divided into compartments according to the need of the experiment and keeping in mind the standard floor space required for broilers, waterers, and feeders. All the chicks were reared on a deep litter system from day- old to 28 days of age. Paddy husk was used as litter material.

Brooding

The brooding temperature was maintained by using electric bulbs. The adequate temperature was provided by adjusting the wattage and height of electric bulbs as per atmospheric temperature and distribution of birds in the compartment.

Vaccination and medication

All the broiler chicks were immunized against Marek's disease Ranikhet (New Castle disease), and IB (Infectious Bronchitis Disease) at the hatchery level. Chicks were vaccinated against Ranikhet (New Castle disease) and Gumboro (Infectious Bursal disease) at the 7^{th} and 14^{th} days of age. On the arrival of chicks on the farm, ground maize and glucose mixed water were provided *adlibitum*. The vitamin B and antibiotic supplement were continued for about 4-5 days, along with drinking water.

Preparation of tulsi leaf (Ocimum sanctum) extract

Mature and disease-free tulsi leaves were collected from the local garden. Then I went to the crop botany department with some samples, and after some examination, they finally certified the plant as tulsi. After washing, the fresh leaves were cut into small pieces with simple scissors and sun-dried, then 20 g mixed with added fresh water made up to 1

liter. Then boiled made up of 1 litter and stored in a refrigerator at 4˚C to preserve the active ingredients of juice (Khatun *et al*., 2013).

Weekly Live Body Weight (g)

Body weight of the individual experimental chick was recorded using a digital weighing balance at the day-old and after that at weekly intervals till five weeks of age. The scale with 0.01g accuracy was used in the digital weighing balance.

Weekly Gain in Body Weight (g)

Broiler chicks were, weighed individually at weekly intervals up to five weeks of age, and weekly body weight gain was calculated by subtracting the live body weight of the previous week from that of the current week and recorded in g.
A weekly gain in Body Weight (g) = Current Week Weight (g)-Previous Week Weight (g)

Weekly Feed Consumption (g)

Adlibitum feed was offered daily, and treatment wise record was maintained. At the end of every week, the leftover feed was measured, and the feed consumed during the week was calculated.

Weekly Feed Consumption= Feed offered during the week – Feed leftover at the end of the week.

Feed Conversion Ratio (FCR)

Adlibitum feed was offered daily, and treatment wise record was maintained. At the end of every week, the leftover feed was measured, and the feed consumed during the week was calculated.

Weekly Feed Consumption= Feed offered during the week – Feed leftover at the end of the week.

Hematological parameter

To conduct hematological studies, ten birds from each group were randomly selected for blood collection. Blood samples were collected through the jugular vein and wing vein in sterile EDTA glass vials on the 21st and 28th days of the experiment, and hematology was done within 6 hours of sample collection. Hematological parameters included hemoglobin concentration (Hb), total erythrocyte count (TEC), total leucocyte count (TLC), and Packed Cell Volume (PCV) was estimated by (Nambiar, 1960; Natt and Hervic, 1954; Doumas, 1978).

Cost benefits analysis of broiler production

The economics of rearing broiler chicks for the complete experimental period was calculated by taking into consideration the cost of chicks, cost of feed consumed by birds, supplementation cost, and miscellaneous expenditures. Net profit per bird was calculated from the sale of birds in the market on a live body weight basis. Similarly, net profit (Tk) per kg body weight was calculated for all treatment groups.

Data analysis

The data pertaining to all the parameters were subjected to statistical analysis by applying a t-test through SPSS software and Duncan's multiple range test (DMRT, 1955).

Results & Discussion

The present study was undertaken with the effects of tulsi leaf extract supplementation (*Ocimum sanctum*) on the growth performance of broilers. The parameters such as growth performance, hematological profile, carcass yield, and economics of broiler were studied.

Growth performance

The growth performance of experimental broilers was evaluated based on average weekly body weight and average weight gain, feed consumption, and FCR.

Average weekly live body weight (g)

The average weekly body weights (g) per bird were recorded from 0 days to the 5th week of age in the control group and treatment group and are presented in Table 1.

Table1: Effects of tulsi leaf extract on weekly growth performance (g) per bird

Week	Control (T_0)Mean ± SD	Treatment (T_1)Mean ± SD	P-value	Significance level
0	48.43±0.61	47.77±0.22	0.188	NS
1	185.17±3.33	205.80±2.86	0.532	NS
2	518.9±4.96	545.87±5.31	0.016	**
3	1019.20±12.26	1064.47±11.61	0.745	**
4	1386.62±5.47	1476.25±3.79	0.000	**

From table 1, it is observed that the average body weight of the day-old chicks was 48.43±0.61, and 47.77±0.22 in the control group and treatment group, respectively. The difference between the control group and the treatments was non-significant. The result revealed that in 1st week, there was no significant difference between the control and the treatment group. The body weight gain of broilers at the end of 1st week was significantly less. Treatment was started on the 8th day. So at the end of 2nd week was indicated significantly higher body weight in the treatment group which was supplemented with the 0.02% tulsi leaf extract. Similarly, the average live body weight of the 3rd week indicated significantly ($P<0.01$) higher body weight at the end of the 4th week the average live weight of the birds indicated significantly ($P<0.01$) showed the same trend of findings Mazhar-Iiahi et al. (2007). Lanjewar et al. (2008) who have reported a significant increase in the live body weight of broilers fed on tulsi leaf extract at 0.02% in the water on live weights of broilers. Similar findings were reported by Reddy et al. (2008); Mode et al. (2009); Patankar (2009), and Thange (2009).

Average weekly body weight gain (g)

The averages weekly body weight gains (g) were recorded from day-old to five weeks of age of broilers for different groups and are presented in table 2. It was observed from the data that there was no significant difference in body weight gain at the end of 1st week between the control group and the treatment group. However at the end of 2nd week, the treatment group supplemented with 0.02% tulsi leaf extract showed significantly ($P<0.05$) higher body weight gain followed by the control group.

Table 2: Effects of tulsi leaf extract on weekly body weight gain (g)

Week	Control (T_0)	Treatment (T_1)	P-value	Significance level
1	137.24±3.42	160.51±1.76	0.0521	**
2	333.50±6.46	339.80±7.58	0.0251	*
3	500.30±13.35	518.60±9.92	0.0532	NS
4	367.42±3.53	412.05±2.91	0.0009	***

The weekly body weight gain in the 1st and 3rd weeks did not show any significant difference between the control groups and treatment. But at the 2nd and 4th week, all the treatment groups showed significantly (P<0.05) highest body weight gain compared to the control. Similar findings were observed by Primprikar (1994), and Mode *et al.* (2009), who reported a significant increase in weekly body weight gain with the supplementation of *Ocimum sanctum* (Tulsi) leaves powder. The findings of Lanjewar *et al.* (2008) corroborate with the present findings who reported a significant increase (P<0.01) in weekly weight gain by supplementation of tulsi leaf powder (TLP) 0.5% and 1% in the diet of broilers. Similar results were reported by Patankar (2009), who included TLP in the diet of broilers during the summer season as a herbal antioxidant. The work of Rao *et al.* (1999); Kumar *et al.* (2000); Pande (2000) also revealed a similar effect of zee stress on body weight gain in broilers. The data revealed that the treatment group supplemented with tulsi leaf extract showed significantly (P<0.01) higher body weight gain. This indicated that there was a positive influence of tulsi leaf extract. This may be attributed to growth stimulating properties of *Ocimum Sanctum* which causes improved nutrient utilization from the diet. These findings are in agreement with Mazhar-Iiahi *et al.* (2007) who reported a significant increase in body weight gain in the broiler supplementation with 2% tulsi leaf extract.

Average weekly feed consumption (g)

The average weekly feed consumption (g) was recorded from day-old to four weeks of age of broilers for different groups are presented in table 3. The result revealed that there was no significant difference in weekly feed consumption between the groups at the end of the first week. The weekly feed consumption ranged from 166.86 in the control group to 185.22 g in the treatment group supplemented with 0.02% tulsi leaf extract. At the end of second-week, values ranged from 435.06 g in the control group to 393.11 g in the treatment group, which was also supplemented with 0.02% tulsi leaf extract.

Table 3: Effects of tulsi leaf extract on weekly feed consumption (g) per bird

Week	Group		P-value	Significance level
	Control (T_0)	Treatment (T_1)		
1	166.86±1.6	185.22±3	0.726	NS
2	435.06±1.79	393.11±2.72	0.023	*
3	682.27±10.53	762.56±5.86	0.053	NS
4	726.40±5.08	725.86±9.85	0.026	*
Cumulative Feed Consumption	2010.59±11.59	2066.75±27.63	-	-

At the end of 1st week, the difference between the control group and the treatment group was non- significant. As well as at the end of 2nd week, the difference between the control and the treatment groups was also non-significant. At the end of 3rd week, the average weekly feed consumption was significantly (P<0.05) higher in the treatment group supplemented with 0.02% tulsi leaf extract. At the end of the 4th week, there was no significant difference in birds' average weekly feed consumption of birds in different groups. The present study's findings agree with Lanjewar *et al.* (2008), who observed no significant difference in feed intake of birds who received 0.05 and 1% tulsi leaf powder in their diet. Similar observations were noted by Thange (2009) reported a non-significant effect on feed consumption of the birds fed on tulsi leaf powder 5 g/kg of the feed. However, Patankar (2009) reported significantly lower feed consumption in tulsi leaf powder-supplemented broiler groups, which contrasts with present findings.

Average weekly Feed Conversion Ratio (FCR)

The average weekly FCR was recorded from the first to six weeks of age of broilers from different groups and is presented in Table 4. The average weekly FCR during the 1st, 2nd, 3rd and 4th week differ non-significantly between the two groups. This indicated that there was no influence of tulsi leaf extract on average weekly FCR. The cumulative FCR at the end of the 4th week was 1.19 and 1.15 in the control and treatment groups, respectively. The data revealed that there was no significant difference in cumulative FCR between different treatment groups. The result indicated no

influence of *Ocimum sanctum* on cumulative FCR in birds. Mazhar-Iiahi *et al.* (2007) observed a non-significant effect in FCR in the group supplemented with tulsi leaf extract. Patankar (2009) observed similar findings, who observed a non-significant effect in FCR in the group supplemented with tulsi leaf powder. However, Lanjewar *et al.* (2008) observed a significantly improved FCR ($P<0.01$) in a group supplemented with 1% tulsi leaf powder. Similar findings were observed by Thange (2009). Wheeler (1994); Rao *et al.* (1999); Pande (2000), and Kumar *et al. (*2004) observed increased FCR in the group supplemented with 1% tulsi leaf extract.

Table 4: Effects of tulsi leaf extract on weekly feed conversion ratio (g/day)

Week	Group		P-value	Significance level
	Control (T_0)	Treatment (T_1)		
1	0.9±0.06	0.9±0.03	0.234	NS
2	1.16±0.01	1.06±0.16	0.053	NS
3	1.26±0.03	1.26±0.02	0.745	NS
4	1.45±0.08	1.40±0.19	0.341	NS
Cumulative feed consumption	1.19±0.09	1.15±0.07	-	-

Hemoglobin (gm%)

The Hb (gm%) values on the 21st day among different treatment groups ranged from 10.45 g/dL in the control group on the 21st day to 11.1 g/dL in the treatment group at the 21st day. The data revealed (Table 5) revealed that the treatment group fed on 0.02% *Ocimum sanctum* has numerically higher Hb % compared to the control group. There was a significant ($p<0.01$) difference which indicated that the supplementation of *Ocimum sanctum* significantly affects the Hb% on the 21st day of the broiler. The data presented in table 5 indicated that the Hb (gm%) at the 28th day age of broilers ranged from 10.6 g/dL in the control group to 11.65 g/dl in the treatment groups. However, there was no significant difference between the control and treatment groups, indicating no influence of Ocimum on Hb (gm%) on the 28th day of experimentation. The findings of Hb (gm%) are in agreement with Gupta and Charan (2007), who reported a normal range of Hb (gm%) in the groups supplemented with *Ocimum sanctum* in the broiler diet.

Total Erythrocyte count (million/cumm^3)

The total erythrocyte count (Table 5) among the different treatment groups ranged from 2.37 million/cumm^3 to 2.58 million/cu mm in the control group to the treatment group. There was no significant difference in the TEC at the 21st day of age. The TEC on the 28th day ranged from 2.53 million/cu-mm in the control group to 2.795 million/cu mm treatment group. The result of TEC showed a significant difference ($p<0.01$) with the treatment of *Ocimum sanctum*, which indicated that the supplementation with *Ocimum sanctum* has a significant effect on Total Erythrocyte Count. The present findings follow the work of Bobade (2006), who reported a significant increase in TEC in broiler fed on *Ocimum sanctum*. Gupta and Charan (2008) reported a normal range of TEC in the group supplemented diet without Vitamin E and Se.

Total Leukocyte count (thousand/cumm^3)

The total leukocyte count (Table 5) among the treatment groups ranged from 25.02 thousand/cumm^3 to 28.90 thousand/cu mm in the control group to the treatment group. There was a significant ($p<0.01$) difference in the TLC at the 21st day of age, indicating that the use of tulsi leaf extract as a supplemented food significantly affected TLC. The TLC at the age of the 28th day ranged from 18.35 thousand/**cu** mm to 47.2 thousand/cumm^3 in the control group to the treatment group, which has no significant difference. This data indicates that the effect of tulsi leaf extract at the age of 28th has no significant effect on TLC.

Table 5: Effects of tulsi leaf extract on hematological parameters in broiler

Haematological Profile	**Group**		**P-value**	***Significant level***
	Control (T_0)	Treatment (T_1)		
Haemoglobin (gm%)	10.45±0.25	11.1±0.55	0.021	*
TLC(thousand/cumm^3)	25.02±6.62	28.90±2.85	0.000	**
TEC (million/cumm^3)	2.37±0.15	2.58±1.92	0.276	*
PCV (%)	31.85±5.21	37.05±2.69	0..504	*

Packed cell volume (%)

The Packed Cell Volume (Table 5) among the different treatment groups ranged from 31.85 % to 37.05 % in the control group to the treatment group. There was a non-significant difference in the PCV at the 21st day of age, indicating that the use of tulsi leaf extract as a supplemented food has no significant effect on PCV. The PCV at the age of 28th days ranged from 33.3 % to 37.45 % in the control group to the treatment group; however, the value had no significant effect. So, tulsi leaf extract has significant effect on PCV.

Table 6: Relative giblet weight (heart, gizzard, liver, spleen and pancreas) of broilers supplemented with tulsi leaf extract from 2-4 weeks of age.

Variable	Group		P-value	Significance level
	T_0	T_1		
Relative heart weight	0.356±0.025	0.412±0.065	0.001	***
Relative gizzard weight	1.324±0.023	1.256±0.012	0.605	NS
Relative liver weight	2.241±0.034	2.305±0.032	0.002	**
Relative spleen weight	.114±0.011	0.123±0.032	0.011	**

At the age of 28th days post mortem was performed. Relative giblet weight was taken, especially heart, gizzard, liver, spleen, and pancreas. The data revealed that relative heart weight, liver weight, and spleen weight have significant values that mean tulsi leaf positively affects giblet weight. But in the case of relative gizzard, weight data showed a non-significant value (Table 6).

Cost-benefits analysis of broiler production

Cost-benefit analysis of broiler production under trial was worked out by considering the prices of inputs prevalent at the time of the experiment in the market. Input consists of the cost of day- old chicks, feed, tulsi leaf extract, vaccine, and other expenses. The broilers were sold Tk. 120 per kg on a live weight basis. Other expenses like labour, electricity and miscellaneous were also considered uniform for the treatment and control groups. From the result presented in table 7, it is seen that the cost of feed per kg after the addition of tulsi leaf extract remains same. Cause the tulsi plants are a natural source. So, the feed cost for the control and treatment groups are the same. The results show that maximum income was obtained from the treatment group in which birds were supplemented with tulsi leaf extract 2ml/L drinking water.

Table 7: Cost-benefit analysis of broiler production per bird in experimental treatment groups

Sr. No.	Particulars	Group	
		T_0	T_1
1	Chick Cost (Tk)	28	28
2	Average feed consumed (kg)/ chicks	2.010	2.066
3	Feed price/kg (Tk)	46	46
4	Cost of herbal growth promoters (Tk)	00	00
5	Feed cost (Tk)	92.46	95.036
6	Miscellaneous (Tk)	20	20

7	Total cost/ broiler (Tk)	140.46	143.036
8	Average live weight (kg)	1.386	1.476
9	Sale price/kg live wt.	120	120
10	Sale price/broiler (Tk)	166.32	177.12
11	Net profit/broiler (Tk)	25.86	34.084
12	Net Profit/Kg weight	18.65	23.09

The findings of the present study are following the Lanjewar *et al.* (2008) who studied the effect of dietary supplementation of tulsi on the performance of the broilers and observed the highest net profit in the group supplemented with tulsi leaf extract than the un-supplemented group. Patankar (2009) studied the comparative efficacy of herbal antioxidants in ameliorating summer stress in broilers and observed comparatively lower net profit in the treatment groups supplemented with herbal antioxidants due to the higher cost of supplementation and increased in feed consumption in herbal supplemented group.

From the findings of the present study, it can be concluded that supplementation with tulsi leaf extract with drinking water causes a significant increase in live body weight and improvement in weekly weight gain and feed efficiency as compared to that of the control group of broiler. Thus tulsi leaf extract supplementation in the broiler rations may be useful for the safe, economic, and efficient production of broiler, and this formulation could be used as an alternative to commercial growth promoters. However, further studies are essential to assess the impact of these medicinal plants on the quality of broiler meat and immune status to ensure the safety of human consumption.

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