



Tulsi leaf and synbiotic powder in broiler diet

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## Effect of Tulsi (*Ocimum sanctum*) Leaf and Synbiotic Powder on Growth Performance, Nutrient Utilization, Carcass and Blood Biochemical Parameters in Broilers

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### ABSTRACT

A six-week feeding trial was followed by a metabolic trial using 120 broiler chicks (Cob-400) in a completely randomized block design to assess the effect of dietary supplementation with tulsi leaf and synbiotic powder as feed additives on growth performance of broilers. The four dietary treatments were designated as TC control group (basal diet), T1 supplemented with tulsi @ 0.5%, T2 supplemented with synbiotic @ 0.05%, T3 supplemented with tulsi @ 0.5% and synbiotic @ 0.05% of feed. Growth performance and carcass yields were significantly higher ( $P < 0.01$ ) in dietary treatment with supplementation of tulsi leaf and synbiotic as compared to control. Significantly higher body weight, weight gain, daily weight gain, protein efficiency ratio and performance index were observed in the T3 group supplemented with a combination of tulsi and synbiotic @ 0.5 percent and 0.05 percent, respectively. Similarly, dietary supplementation of tulsi leaf and synbiotic in combination significantly reduced serum triglyceride and cholesterol level. Whereas, dietary inclusion of tulsi leaf alone and in combination with synbiotic reduced feed intake and feed conversion ratio. However, dry matter digestibility, nitrogen balance, eviscerated weight, heart, liver, gizzard weights, total protein, albumin, globulin, haemoglobin and packed cell volume did not differ in treatment groups. The mortality rate was found to be lower in the treatment groups when compared to the control group. Based on the findings of this study, it is concluded that supplementing tulsi leaf at 0.5 percent with synbiotic powder at 0.05 percent improved the growth performance, nutrient utilization, carcass traits and blood haemato-biochemical parameters of broiler chicks without any negative effects.

**Key words:** Broiler performance, Carcass, Nutrient utilization, Synbiotic, Tulsi

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### INTRODUCTION

The poultry production systems showed a significant rise in the worldwide production of poultry meat and eggs (Armstrong, 1986), which resulted in number of "feed additives" to be discovered and widely used. In order to influence rapid growth, many synthetic drugs and growth promoters are used in broilers, but their use has shown many risks, like high cost, adverse effects on bird welfare and long residual properties etc. However, several reports have documented positive effects of herbs on the digestion process, bile salt

production and digestive enzyme activity of the intestinal mucosa and pancreas. Tulsi is known as "the mother medicine of nature" with antimicrobial, immunomodulatory, anti-stress, anti-inflammatory, anti-pyretic, anti-asthmatic, hypoglycemic, hypotensive and analgesic functions. Tulsi showed effective role in growth performance, reduced mortality, improved haemato-biochemical parameters, economic traits (Shende et al., 2020a, 2020b, 2021a), improved digestibility and nitrogen balance (Tanwar, 2018).

Synbiotics are a combination of prebiotics and

probiotics that provide synergy and net benefit to the host. A dietary synbiotic supplement improved growth performance (Min et al.; 2016; Salehimanesh et al., 2016), had antioxidant capacity (Capcarova et al., 2010; Dev et al., 2020), improved intestinal morphology and immune function (Dizazi et al., 2012; Hassanpour et al., 2013; Julendraa et al., 2020). Keeping the aforesaid facts in view, the present investigation was planned to explain the possibilities of utilization of tulsi (*Ocimum sanctum*) and synbiotic as feed additive alone or in combination on the growth performance, nutrient utilization, carcass and blood biochemical parameters of broilers.

## MATERIALS AND METHODS

### Birds and dietary treatment

An experiment was conducted for six weeks at the Poultry Farm, College of Veterinary and Animal Science, Navania, Udaipur, Rajasthan University of

Veterinary and Animal Science to determine the effect of supplementation *Ocimum sanctum* and synbiotic powder as feed additives on growth performance and nutrient utilization in broilers. In this study, 120, day old chicks were weighed individually and distributed into four different dietary treatment groups utilizing CRD. The body weight of chicks was kept similar in all groups. The diet - Tc served as control and fed as per BIS (1992), whereas diet T1 was supplemented with tulsi leaf powder @ 0.5 % whereas diet T2 was supplemented with synbiotic powder @ 0.05% and T3 was supplemented with combination of tulsi leaf powder @ 0.5 % and synbiotic powder @ 0.05 %. *Ocimum sanctum* leaf powder and synbiotic was purchased from Rishi organic Farm House, Sanganer, Jaipur, Rajasthan. Ingredient composition, proximate composition of broiler starter, finisher and are presented in Table 1. Composition of synbiotic is presented in Table 2.

Table 1. Feed ingredients and chemical composition of basal diet for broiler chickens (%)

Ingredients	Broiler starter ration (0-21 day)	Broiler finisher ration (22-42 day)
Maize	51.7	54.75
Soybean deoiled cake	33.0	30.0
Deoiled rice bran	10.0	10.0
Raw rice bran	2.0	1.5
Vegetable oil	2.0	2.5
L-Lysine	0.15	0.15
DL-Methionine	0.15	0.1
*Premix	1.0	1.0
Analyzed chemical composition(%)		
Dry matter	92.34	93.98
Crude protein	22.50	21.20
Ether extract	4.50	5.40
Crude fibre	4.40	4.80
Total ash	10.80	9.60
Nitrogen free extract	57.80	59.00
Calcium	1.42	1.26
Phosphorus(Av.)	0.71	0.53
ME (Kcal/kg of feed)	3040	3106

\*Each kg of premix contains: Vitamin A 7, 00, 000 IU; Vitamin D3 70000 IU; Vitamin E 250 mg; Vitamin B3 1 g; Zn 9.6 g; S 7.2 g; Cu 4.2 g; 6000 mg; Fe 1500 mg; Mg 1500 mg; I 325 mg; Co 150 mg; K 100 mg; Cr 75 mg; Se 7.5 mg; Na 6 mg; Ca 191 g; P 95 g.

Table 2. Ingredient composition of synbiotic used in the experimental trial

Ingredients	Active constituents	Concentration
Prebiotic	Mannon - oligosachharide	14-16%
Probiotics	<i>Lactobacillus acidophilus</i>	09 CFU/g
	<i>Lactobacillus bulgaricus</i>	
	<i>Lactobacillus plantarum</i>	
	<i>Streptococcus faecium</i>	
	<i>Bifidobacterium bifidus</i>	

### Feeding and management of birds

During the course of the study, identical standard management practices concerning brooding, feeding, watering and control of diseases etc. were followed for each group. As bedding material, clean and dry wheat straw was used. Commercial broiler starter and finisher rations were provided to broiler chicks from day 1-21 and day 22-42, respectively. Ad libitum feeding was ensured throughout the feeding trial. At the time of procurement, all the day-old chicks were vaccinated against Marek's disease. The chicks were also inoculated by eye drop at the age of seventh against New Castle Disease / Ranikhet Disease and booster dose was given on twenty first day of age in drinking water. Chicks were inoculated against Infectious Bursal Disease on the fourteenth day of maturity. Strict and exhaustive sterile measures and care were taken to prevent infection or disease from occurring by any scroungers into the poultry house. Individual chicks were weighed at the start of the experiment and then at weekly intervals for 6 weeks in the morning hours before feeding. The body weights were recorded using a digital weighing balance, to calculate weight gain from the difference in body weight attained at the end and start of the relevant period. Average daily gain (ADG) was calculated by dividing total body weight gain from number of days. The average feed consumption of each treatment group was recorded. The disparity between the weight of feed supplied and the remaining feed left on a dry matter basis was also used to calculate cumulative overall feed consumption over the period of the experiment. The feed conversion ratio was calculated by dividing the cumulative feed intake by the total body weight gain of birds. A performance index was calculated by dividing the weight gained by the feed conversion efficiency and protein efficiency ratio was obtained by dividing the average weight gained by the protein consumed. Birds under the experiment were supervised daily for if any death or ailment during daily routine poultry farm operations to calculate

the mortality rate. The dead birds were then subjected to routine post-mortem examination.

### Metabolic Trial

The metabolism trial was conducted on six birds per treatment group at the end of the 6th week; the birds were randomly selected and shifted to individual metabolic cages. Polythene sheet of appropriate size was spread over the dropping trays for collection of mixed excreta and the birds were individually fed with respective treatment diets. The adaptation period lasted about 2 days, followed by a collection period of 5 days. During the collection period, data on the amount of feed offered, left over, and excreta voided were collected in order to determine nutrient utilization. After deducting the leftover feed from the feed offered, the daily feed intake was calculated. Representative feed samples were taken from the bulk, ground, and stored. For the purpose of nitrogen (N) determination, samples of excreta were taken and preserved in 20 ml of 25% H<sub>2</sub>SO<sub>4</sub> in wide-mouth glass stoppered bottles and kept in refrigerator. Whereas, for determination of the dry matter content, samples were kept in previously weighed clean petri dishes and dried in hot air oven at 850°C till constant weight was obtained. The dried material obtained was subsequently ground and secured for further analysis. The nitrogen content of feed and excreta was determined by Kjeldahl using Kel plus automatic nitrogen analysis equipment. The percentage of nitrogen retention under different treatments was calculated by the per cent difference in intake and output of the nutrient. Treatment wise digestibility of dry matter of diet was calculated by using the equation: weight of dry matter consumed minus weight of excreta voided on dry matter basis divided by weight of dry feed consumed.

### Proximate analysis

The proximate analysis was carried out at Department of Animal Nutrition, College of

Veterinary and Animal Science, Navania, Udaipur for a starter and finisher feed and excreta of the metabolic trial (AOAC, 2016). The total nitrogen content of feed and excreta was determined following standard Kjeldahl's method using Kel plus nitrogen analyzer (Pelican equipment).

### Carcass characteristic and organometry

The birds used for metabolism trial were further used at the end of 6th week for study of the effect of different treatments on carcass characteristic that included dressed weight percent, edible yield percent and eviscerated weight percent, giblet per cent etc. For that, one representative bird from each replicate was weighed and slaughtered after the completion of the five-day trial. Selected birds had live weight almost similar to the mean live weight of the population concerned. The birds were weighed immediately before slaughtering. The slaughtering was done by severing the jugular vein and 5-6 minutes bleeding time was allowed for each bird. The weight of organs, liver, heart and gizzard was taken separately by using a digital weighing balance. The dressed birds were eviscerated by giving a median cut in the abdomen and removing the crop, gullet, trachea and viscera. The lungs were scrapped off. Heart, liver, pancreas, spleen and gizzard were separated from GI tract. Eviscerated weight was calculated by subtracting the weight of viscera except for giblet from carcass yield. The weight of giblets such as heart, liver and gizzard were taken and express as percent of pre-slaughter weight.

### Hematological and serological parameters

On 42<sup>nd</sup> day of the experiment for hematological and serum biochemical parameters studies, about 3ml of blood samples were collected from wing vein of 2 representative birds of each replicate. For the estimation of hemoglobin (Hb) and packed cell volume (PCV), using automatic hematology analyzer, half of the blood was transferred into sterilized EDTA containing tubes. The remaining blood sample was transferred to non EDTA tubes for preparation of serum. Subsequently, serum was separated through centrifugation of sample at 3000 rpm for 15 minutes and stored at - 20°C until further analysis. These samples were analyzed by automatic haemo analyzer, using commercial test kits as per protocol procedure for total protein, serum albumin, serum cholesterol and triglyceride. Serum globulin content was determined from total protein levels by subtracting the serum albumin concentrations.

### Statistical analysis

Data collected during the current research work was subjected to statistical analysis by implementing standard methods of variance analysis as defined by statistical package for social science (SPSS 2018), version 20. Significance of mean differences (F-values) was calculated by Dunken's multiple-range test as updated by Kramer (1956).

## RESULTS AND DISCUSSION

### Growth Performance

The results of growth performance are summarized in Table 3.

Table 3. Effect of tulsi and synbiotic on growth performance of broilers

Growth performance Parameters	Treatment groups				SEM	P value
	TC	T1	T2	T3		
BW	21781 <sup>a</sup>	2261 <sup>b</sup>	2300 <sup>bc</sup>	2346 <sup>c</sup>	14.68	0.0010
WG	2139 <sup>a</sup>	2222 <sup>b</sup>	2261 <sup>bc</sup>	2306 <sup>c</sup>	14.75	0.0011
DWG	50.9 <sup>a</sup>	52.9 <sup>b</sup>	53.8 <sup>bc</sup>	54.9 <sup>c</sup>	0.35	0.0011
FI	4542 <sup>b</sup>	3950 <sup>a</sup>	4565 <sup>b</sup>	3998 <sup>a</sup>	60.15	0.0005
FCR	2.12 <sup>c</sup>	1.79 <sup>a</sup>	2.04 <sup>b</sup>	1.80 <sup>a</sup>	0.01	0.0000
PI	1011 <sup>a</sup>	1245 <sup>c</sup>	1108 <sup>b</sup>	1288 <sup>d</sup>	5.96	0.0000
PER	2.23 <sup>a</sup>	2.66 <sup>c</sup>	2.35 <sup>b</sup>	2.73 <sup>d</sup>	0.02	0.0000

The means bearing different superscript in a row differ significantly TC - Basal diet (Control); T1- Basal diet supplemented with 0.5% tulsi; T2- Basal diet supplemented with 0.05% synbiotic; T3- Basal diet supplemented with 0.5% tulsi and 0.05% synbiotic. BW- Body weight, WG- Weight gain, DWG-Daily weight gain, FI- Feed intake, FCR- Feed conversion ratio, PI- Performance Index, PER- Protein efficiency ratio

There was a significant improvement ( $P < 0.01$ ) in BW, BWG, DWG, FI, FCR, PI and PER at the end of experimental period with supplementation of tulsi and synbiotic alone and in combination @ 0.5 and 0.05%, respectively in the diets as compared to control group. However, the best results were observed in group supplemented with tulsi leaf and synbiotic in combined form @ 0.5 and 0.05%, respectively. Bird performance and feed utilization efficiency are linked with quality and quantity of microbial load presents in the gut of broilers (Islam, 2008). Synbiotics are either valuable microorganisms or substrates that promote the growth of these microorganisms, which can be appropriately harnessed by food manufacturers and hold significant assurance for the health care industry. According to the current study, some researchers discovered a positive effect of tulsi leaf and synbiotic. Lanjewar et al. (2008), Swathi et al. (2012), Bhosale et al. (2015), Hasan et al. (2016) and Biswas et al. (2017) reported highly significant improvement in growth performance recorded as a result of tulsi leaf supplementation. Herbs have antimicrobial activity thus favor better nutrient absorption or the stimulation of the immune system (Wenk, 2003). These results well corroborate with, Bozkurt et al. (2009), Abdel et al. (2012), and Abdel et al. (2017) who recorded significant improvement in growth performance due to synbiotic supplementation. Awad et al. (2009) reported significant increase in daily body weight gain in synbiotic supplemented group as compared to control and probiotic supplemented group. Tanwar (2018) and Yadav (2018) observed significant effect on PER and PI due to supplementation of Tulsi leaf powder in broilers as compared to control. The performance of synbiotic supplemented groups in terms of PER was higher than control group well corroborate with Ashayerizadeh et al. (2011) and

Dev et al. (2020). Nagar et al. (2021) observed significant improvement in PER and PI of broilers when supplemented with combination of tulsi and synbiotic. Results of present study regarding feed intake due to tulsi leaf supplementation are in accordance with Singh et al. (2014) who reported significant ( $P < 0.05$ ) decrease in feed consumption due to supplementation of tulsi leaf @ 0.5 & 1% of ration than those of control group. Das et al. (2016) noticed increase in feed consumption as a result of inclusion of synbiotic in the diet of broilers.

### **Nutrient Metabolizability**

The metabolizability of DM and nitrogen balance was found non-significant among the treatment group with supplementation of tulsi leaf and synbiotic alone and in combination. Nagar (2021) who observed non-significant effect of synbiotic on dry matter metabolizability of broilers in contrast to the present study. Apata (2008) observed significant effect on dry matter metabolizability due to supplementation of probiotic culture *Lactobacillus bulgaricus*.

### **Percent mortality**

Out of total of 120 chicks reared in the present study, 8 chicks died during entire experimental period. Thus, total mortality was 6.67%. The mortality of chicks varied from 3.33 to 10 per cent among different treatment groups. Singh et al. (2014), Tanwar (2018) also reported reduced mortality percent with supplementation of tulsi leaf powder. Similarly, Abdel et al. (2012) and Popovic et al. (2015) also observed reduced mortality due to synbiotic supplementation.

### **Carcass traits**

The data of carcass traits for various treatment groups have been presented in Table 4.

Table 4. Effect of tulsi and synbiotic on carcass traits of broilers

Carcass Parameters	TC	T1	T2	T3	SEM	P value
Dressing (%)	75.4 <sup>a</sup>	76.8 <sup>a</sup>	78.8 <sup>b</sup>	78.4 <sup>b</sup>	0.59	0.0251
Eviscerated weight (%)	63.6	63.1	59.7	61.0	0.96	0.0874
Heart wt. (%)	0.54	0.55	0.52	0.52	0.02	0.5980
Liver wt. (%)	1.93	2.07	1.58	1.77	0.22	0.4844
Gizzard wt. (%)	1.73	1.70	1.66	1.77	0.06	0.6308

The means bearing different superscript in a row differ significantly ( $P < 0.05$ )

TC - Basal diet (Control); T1- Basal diet supplemented with 0.5% tulsi; T2- Basal diet supplemented with 0.05% synbiotic; T3- Basal diet supplemented with 0.5% tulsi and 0.05% synbiotic.

The statistical analysis of data shown in revealed significant ( $P < 0.05$ ) effect of supplementation of tulsi alone and in combination on dressed weight but non-significant on eviscerated weight. On observing the data, highest dressed weight was recorded in T3 which showed non-significant difference with T2 group and contrarily, lowest was observed in TC group which was non-significantly differed with T1 group. Statistical analysis of data showed non-significant effect among the treatment groups on liver, heart and gizzard weight. Results of the present study regarding non-significant effect on carcass yield due to tulsi leaf well corroborate with the findings of Vasantha kumar et al. (2013) who reported non-significant effect of tulsi on dressing

percentage of broiler with incorporation of tulsi leaf powder in broiler feed. Awad et al. (2009) observed significant effect on carcass yield due to synbiotic supplementation. Results of the present study regarding eviscerated weight percentage (%) well corroborated with findings of Prajapat (2016) who reported non-significant effect of incorporation of tulsi leaf powder on eviscerated weight percentage (%) of broiler with those of control chicks.

#### Haemato-biochemical parameters

The results of haemato-biochemical parameter for various treatment groups have been presented in Table 5.

Table 5. Effect of tulsi and synbiotic on Haemato-biochemical parameters of broilers

Treatment Groups	Parameters						
	Hb gm%	PCV %	Protein (g/dl)	Albumin (g/dl)	Globulin (g/dl)	Triglyceride (mg/dl)	Cholesterol (mg/dl)
TC	8.97	25.27	3.46	2.20	1.27	42.50 <sup>c</sup>	157.15 <sup>c</sup>
T1	9.14	27.80	3.44	2.22	1.22	42.26 <sup>b</sup>	156.32 <sup>b</sup>
T2	9.40	27.57	3.64	2.24	1.42	42.11 <sup>b</sup>	156.11 <sup>ab</sup>
T3	9.44	29.87	3.79	2.27	1.52	41.80 <sup>a</sup>	155.48 <sup>a</sup>
SEM	0.13	1.09	0.11	0.07	0.09	0.05	0.21
P value	0.123	0.117	0.169	0.899	0.156	0.001	0.008

The means bearing different superscript in a column differ significantly ( $P < 0.01$ )

TC - Basal diet (Control); T1- Basal diet supplemented with 0.5% tulsi; T2 - Basal diet supplemented with 0.05% synbiotic; T3 - Basal diet supplemented with 0.5% tulsi and 0.05% synbiotic.

The statistical analysis revealed highly significant ( $P < 0.05$ ) effect of feeding of tulsi and synbiotic in different treatment groups on triglyceride and cholesterol level. Serum triglyceride and cholesterol level was significantly reduced in treatment groups as compared to control groups. However, non-significant effect was observed for haemoglobin, packed cell

volume, total protein, albumin and globulin value among the treatment groups.

The results obtained from study in text are in line with the findings of Salah et al. (2018) who recorded similar effect on cholesterol due to supplementation of synbiotic. Similarly, Lanjewar et al. (2008) and Shende et al. (2020a) also reported significant reduction in

serum triglyceride and cholesterol level due to tulsi leaf powder. Sharifi et al. (2011) recorded significant reduction in serum triglyceride level due to supplementation of synbiotic powder. The results obtained from study are in line with the findings of Beski et al. (2015) who recorded non-significant effect on serum total protein due to supplementation of synbiotic. That apart, results obtained in the present study are in accordance with the findings of Gupta and Charan (2007) who observed non-significant effect of tulsi leaf powder on Hb and PCV value of the broilers. Likewise, the results obtained in the present study on PCV level of broilers well corroborate with findings of Shende et al. (2021b) who recorded non-significant effect of synbiotic on Hb and PCV levels of broilers.

### CONCLUSION

Supplementation of tulsi leaf and synbiotic powder as feed additives at 0.5% and 0.05%, respectively improved growth performance of broilers. Similarly, data from this study indicated that supplementation of combination of tulsi leaf and synbiotic powder @ 0.5% and 0.05%, respectively resulted in improved carcass yield, improved nutrient utilization and haemato-biochemical parameter. It could be concluded that supplementation of tulsi @ 0.5% and synbiotic @ 0.05% is very effective and could be a viable suggestion option for rising broiler production.

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