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Efficacy of a bioactive botanical blend in enhancing gut integrity and productivity in broiler

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Abstract

In this 42-day research trial, 1,760 one-day-old "Cobb-430Y" broiler birds were allocated to four treatment groups, A, B, C, and D, each with 440 birds further divided into four replicates of 110 birds each to evaluate the effects of Bioactive Botanical Blend (BBB) versus a standard symbiotic on gut integrity and productivity in broilers. The BBB consisted of a blend of Piper longum, Mangifera indica, Thymus vulgaris, Ocimum sanctum, Trachyspermum ammi, Allium cepa, and Curcuma longa, while the standard symbiotic contained probiotics including Bacillus licheniformis, Bacillus coagulans, Lactobacillus acidophilus, Bacillus subtilis, and prebiotics such as mannan oligosaccharide and β-glucans. Group A served as the control receiving the basal diet. Group B was provided with BBB at 50 g/100 kg, group C received BBB at 100 g/100 kg, and group D received the standard symbiotic at 25 g/100 kg. Birds in groups C and D found non-significant (p>0.05) differences in weekly live body weight, cumulative weight gain, feed intake, and feed conversion ratio. Duodenal, jejunal, and ileal villus lengths in group C were significantly (p<0.05) increased by 4.49%, 4.40%, and 4.59%, respectively, compared with group D. The villus height to crypt depth (V/C) ratios of the duodenum and jejunum were significantly (p<0.05) higher by 9.44% and 3.44%, respectively, while duodenal crypt depth was 4.36% lower in group C compared with group D. Carcass traits such as dressing percentage (with and without giblet) and breast meat percentage in groups B, C, and D showed no significant (p>0.05) differences. Breast meat percentage in group C was 1.81% higher than in group D. Thus, the inclusion of BBB at 100 g/100 kg feed was effective in improving growth performance, carcass traits, and gut health via enhanced intestinal histomorphology. Similar effects were shown by standard symbiotic at 25 g/100 kg feed, excluding gut health.

Keywords: Bioactive botanical blend, standard symbiotic, broilers, villus height to crypt depth ratio, carcass traits, gut health

Introduction

With the global population expected to reach approximately 10 billion by the year 2050, it is estimated that food production must increase by around 70% to meet the growing demand. According to projections by the World Health Organization, global meat production will need to increase by approximately 200 million tons to reach a total requirement of 525 million tons annually to satisfy future demand [1]. To ensure safe, sustainable, nutritious and affordable supply of food, collaboration between food producers and agricultural scientists is required. Urbanization and transformation in livelihood trends will drive demand for high-quality, safe, and nutritious food. As a result, livestock sector is under constant pressure of meeting the expectation of producing quality protein while navigating challenges like industry automation, animal welfare, environmental pollution and political instability [2]. A healthy gut microbiota is crucial to ensure intestinal well-being, and disturbances, known as "dysbiosis," can arise from factors like toxins and antibiotics, leading to inflammation or infection. The intestinal epithelium serves as a protective barrier, controlling the transit of materials while safeguarding against harmful substances. Maintaining this balance is essential for overall gut function and health [3]. When the lining of small intestine allows harmful substances-such as bacteria and toxins-to enter the bloodstream, it leads to a condition named "Leaky gut". This leads to inflammation and nutrient depletion, negatively affecting metabolic functions, especially immune, metabolic, and endocrine responses.

Consequently, animal performance may be severely diminished ^[4]. After antibiotic growth promoters (AGPs) were banned, the poultry industry in Europe faced challenges like health issues, reduced performance and animal welfare concerns. To address these problems, alternatives such as prebiotics, probiotics, and plant-based additives have been introduced and are increasingly utilized in chicken and turkey production ^[5]. Researchers have suggested various promising alternatives to replicate the effects of AGP, and many of which have been already adopted in the commercial meat production. Phytogenic feed additives (PFAs) or Phyto biotics not only improve performance, meat quality and feed efficiency but also exhibit antioxidant properties, mainly due to their active components of essential oils (EOs) ^[6]. Phyto biotics also supports better nutrient absorption, possibly by boosting digestive enzymes activity such as lipase, protease, or amylase and improving gut morphology.

The mechanism of action of most Phyto biotics is not yet fully unravelled. Nevertheless, numerous studies have reported the growth-promoting, antimicrobial, antioxidative, immunomodulatory effects in animals. Piperine from Piper longum and mangiferin from Mangifera indica both exhibit strong antioxidant and gut-supportive properties, enhancing nutrient absorption and reducing oxidative stress [7]. Piperine improves feed efficiency by increasing the absorptive surface of the intestine, while mango leaf extract boosts antioxidant enzyme activity and reduces stress markers, contributing to better overall health and performance in broilers [8]. Thymol-rich Thymus vulgaris and Ocimum sanctum (Tulsi) share potent antimicrobial, anti-inflammatory, and gut-enhancing effects [9]. Thymus vulgaris improves intestinal morphology and microbial balance. Tulsi, especially in combination with clove, significantly enhances villus height, final body weight, and carcass yield, positioning both as effective natural alternatives to AGPs [10].

Ajwain (Trachyspermum ammi) and bentonite both contribute to improved gut health and feed conversion [11]. Ajwain, due to its high thymol content, enhances villus height and reduces harmful microbes like E. coli [12]. At the same time, bentonite enhances nutrient retention, pellet quality, and intestinal villus architecture, thereby supporting improved digestion and performance in broilers [13]. Onion (Allium cepa) and turmeric (Curcuma longa) are recognised as natural growth promoters because of their antioxidant, antimicrobial, and anti-inflammatory effects. Onion enhances Lactobacillus populations and reduces E. coli, while turmeric supplementation boosts antioxidant status, reduces liver stress, and improves broiler productivity, making them ideal for AGP-free feeding strategies [14, 15]. In this study, we evaluate the efficacy of a phytogenic blend-comprising Piper longum, Mangifera indica, Thymus vulgaris, Ocimum sanctum, Trachyspermum ammi, Bentonite, Allium cepa, and Curcuma longa—acknowledged for their synergistic antimicrobial, antiinflammatory, antioxidant, and gut health-promoting properties. This combination is compared against symbiotics, which integrate prebiotics and probiotics to improve gut flora balance and nutrient absorption. While symbiotics are widely recognized for enhancing immunity and performance, their viability is often affected by heat, storage conditions, and strain-specific limitations. Phytogenic additives, by contrast, are heat-stable and bioactive across multiple physiological systems. Our objective is to determine whether this multifunctional phytogenic blend can offer a superior or equivalent alternative to conventional symbiotics in broiler production.

Materials and Methods Housing and Vaccination

This trial was caried out at Dr. B.V. Rao Institute of Poultry Management and Technology (IPMT), Pune, Maharashtra, India.

One-day-old "Cobb 430Y" broiler chicks of mixed sexes (n = 1760) were obtained from Venkateshwara Hatcheries Pvt. Ltd. Pune, Maharashtra, India. The birds were housed in non-environmentally controlled sheds. Birds were maintained in separate pens with 2 sq. ft. of area per bird. Rice husk was used as the bedding material, and 2-3 inches of rice husk litter on a cement floor was evenly distributed. After the arrival of the chicks, they were weighed and randomly distributed into separate pens in a completely randomized design.

For the first five days, they were provided with Venky's Aminovit Gold through drinking water. Starting from the day the birds arrived; their live body weight (LBW), body weight gain, feed consumption and feed conversion ratio was recorded at the end of each week.

The birds were vaccinated with the Super LaSotaTM vaccine against Newcastle disease on day 7, followed by a booster dose on day 21. The vaccination for Infectious Bursal Disease (IBD-intermediate strain) was given on day 14 and its booster on day 28. Both the vaccines were administered intranasally, and their boosters were dosed through oral consumption in drinking water.

Bioactive Botanical Blend (BBB)

A phytogenic mixture, hereafter referred to as BBB, was used in this experiment. This mixture was composed of *Piper longum*, *Mangifera indica*, *Thymus vulgaris*, *Ocimum sanctum*, *Trachyspermum ammi*, *Allium cepa and Curcuma longa*. The raw materials were sieved through vibratory shifter to remove unwanted material, then blended and mixed homogenously before administering in feed.

Experimental Design

A total of 1760 broiler birds were assigned to four experimental groups such as A, B, C, and D. Each group consisted of 440 birds, divided into 4 replicates with 110 birds in each replicate.

The replicates were arranged to ensure uniform distribution across available housing spaces, following the layout and management practices of the trial site. The duration of the research trial was 42 days.

The treatment groups were as follows (Table 1):

Group A: Control group receiving basal diet

Group B: Basal diet + 50 g BBB/100 kg of feed

Group C: Basal diet + 100 g BBB/100 kg of feed

Group D: Basal diet + 25 g Standard symbiotic/100 kg of feed

The standard symbiotic used in the experiment was a synergistic blend of probiotics consisting of *Bacillus coagulants*, *Bacillus licheniformis*, *Bacillus subtilis*, *Lactobacillus acidophilus* (Total count not less than 4 billion CFU/g) and prebiotics (Mannan oligosaccharide and β -Glucans).

Table 1: Dietary treatment groups of experimental birds

	A	В	C	D
ſ	Control broiler diet	Inclusion of Bioactive Botanical	Inclusion of Bioactive Botanical Blend	Inclusion of Standard Symbiotic at
	without inclusion of Bioactive	Blend at dose of 50 g/100 kg of		dose of 25 g/100 kg of finished
	Botanical Blend	finished feed	at dose of 100 g/100 kg of finished feed	feed

Feed and water

Mash type feed was prepared in-house at IPMT for the trial. Water was available *ad libitum* for all the birds. The feeding regimen was as follows: Pre-starter-weeks 1 and 2, Starter-weeks 3 and 4,

Finisher-weeks 5 and 6. The proximate analysis of the feed was carried out at Envirocare Labs Pvt. Ltd., Thane, MH, India 400604 and the results are presented in Table 2.

Table 2: Proximate compositions of pre-starter, starter and finisher diets

Chemical Parameters	Pre-starter	Starter	Finisher
ME (Kcal/kg)	3021	3097	3204
Loss on drying%	13.5	12.6	11.1
Crude protein%	22.4	21.7	18.7
Total ash%	7.2	6	6.7
Ether extract%	2.7	4.7	8
Soluble dietary fibre%	Below limit of quantification	Below limit of quantification	1.6
Insoluble dietary fibre%	18	15.1	13.9

Evaluation of zootechnical characteristics

In each replicate, 20 birds were randomly selected for average body weight recording. Average weekly feed intake was calculated by subtracting leftover feed from total feed offered. The average weekly gain in body weight was calculated by subtracting the live body weight of previous week from that of current week and recorded in grams. Weekly feed conversion ratio (FCR) was calculated as:

FCR= Total feed consumed in a week (in grams)
Weight gained in a week (in grams)

Carcass Traits

On 42nd day of the trial, six birds per replicate were randomly selected for carcass trait evaluation and slaughtered using halal method. Before slaughtering, birds were subjected to a three-hour feed withdrawal and their live body weights were recorded. Post-slaughter, birds were completely bled and defeathered, including removal of the skin. Dressed weight (with and without giblets), weight of breast meat and total giblet were measured. All carcass trait parameters including dressing percentage (with and without giblets), breast meat yield, and relative giblet weights were calculated as a percentage of live body weight for comparison among groups. The dressing percentage was calculated by using the formula.

Dressing % (with or without giblet) = $\frac{\text{Weight of dreesed chicken with or without giblet (g)} \times 100}{\text{Live body weight (g)}}$

Intestinal Histomorphology Parameters

From the slaughtered birds (n=96), a 2 cm segment from the midpoint of the duodenum, jejunum and ileum were collected aseptically and fixed in 10% neutral buffered formalin for 48 hours. Tissues were processed using standard histological methods: washed, dehydrated in graded ethanol, cleared in xylene, embedded in paraffin, sectioned at 5 µm, and stained with

haematoxylin and eosin ^[16]. Sections were examined under a light microscope (Nikon E100) at 40x magnification. Six intact villi per section were randomly selected to measure villus height (from tip to villus-crypt junction) and crypt depth (from base to opening). The villus height to crypt depth (V/C) ratio was then calculated.

Statistical analysis

All data was presented as mean \pm standard error (SE). Treatment effects were evaluated using one-way analysis of variance (ANOVA) ^[17], followed by Tukey's post hoc test ^[18] for multiple comparisons. Differences were considered statistically significant at p<0.05. All analyses were conducted using GraphPad Prism software (version 10.3.0).

Results

Growth performance parameters Average live body weight (g)

Table 3 shows the effect of a bioactive botanical blend and a standard symbiotic on the average LBW of broilers. No significant (p<0.05) difference was seen during the first two weeks in average LBW, among the four experimental groups. In the third and fourth weeks, groups C and D found significantly higher (p<0.05) live body weights in comparison with the control group A and group B, while both these groups (C and D) found non-significant (p>0.05) difference. During the weeks 3, 5 and 6 group B showed significantly (p<0.05) higher LBW in comparison with control group A. In week 5, group C showed the highest (p<0.05) LBW in comparison with all three groups. In week 6, group C found a significantly (p<0.05) higher LBW than the control group A and group B, but showed non-significant (p>0.05) difference in comparison with group D. The average cumulative LBW (g) at the sixth week increased by 4.96%, 7.91%, and 6.26% in groups B, C, and D, respectively, in comparison with the control group A. Group C exhibited a 1.55% higher value compared to group D.

Table 3: Effect of bioactive botanical blend and standard symbiotic on average weekly live body weight (g) in broilers

Wash		Experimental groups				
Week	A	В	C	D		
Day old	43.47±0.18	43.19±0.22	43.25±0.27	42.87±0.9		
1	198.25±0.85	204.75±4.21	207.25±1.80	206.75±3.09		
2	498.50±7.85	507.25±4.40	515.00±5.40	510.00±4.56		
3	908.75°±4.23	937.50 ^b ±12.07	987.50°±9.89	984.25a±1.89		
4	1421.75 ^b ±14.21	1432.25 ^b ±15.09	1536.00a±11.75	1515.00a±11.73		
5	1983.25 ^d ±10.06	2018.50°±3.43	2106.00°a±2.97	2078.75b±3.20		
6	2587.50°±9.58	2715.75 ^b ±25.35	2792.25a±2.78	2749.50ab±26.17		

Average cumulative body weight gain (g)

Table 4 presents the effect of a bioactive botanical blend and standard symbiotic on the average cumulative BWG in broilers. During the first two weeks, no significant (p>0.05) difference was observed across the four experimental groups. During the third and fourth weeks, groups C and D found significantly higher (p<0.05) cumulative BWG in comparison with groups A and B. However, groups C and D showed non-significant (p>0.05) difference. In weeks 3, 5, and 6, group B exhibited significantly higher

cumulative BWG against the control group A. In week 5, group C showed the highest (p<0.05) cumulative BWG compared to all three groups: A, B, and D. In week 6, group C showed a significant difference (p<0.05) compared to control group A and group B, but showed similar (p<0.05) BWG with group D. The average cumulative live body weight gain at the sixth week was higher by 5.05%, 8.06%, and 6.39% in groups B, C, and D, respectively, relative to the control group A. Group C exhibited a 1.57% higher value compared to group D.

Table 4: Effect of bioactive botanical blend and standard symbiotic on average cumulative weekly live body weight gain (g) in broilers

Week	Experimental groups				
vveek	A	В	C	D	
1	1 154.78±0.88 161.57±4.42		164.00±2.03	163.88±3.05	
2	455.03±8.01	464.07±4.49	471.75±5.63	467.13±4.62	
3	865.28°±4.35	894.32 ^b ±12.23	944.25 ^a ±10.10	941.38 ^a ±1.87	
4	1378.28 ^b ±14.22	1389.07 ^b ±14.98	1492.75 ^a ±11.88	1472.13 ^a ±11.65	
5	1939.78 ^d ±9.89	1975.32°±3.37	2062.75°a±2.88	2035.88 ^b ±3.29	
6	2544.03°±9.60	2672.57 ^b ±25.23	2749.00°a±2.70	2706.63ab±26.23	

Average cumulative feed intake (g)

Table 5 presents the effect of the bioactive botanical blend and standard symbiotic on the average cumulative feed intake in broilers. During the first two weeks, there was no significant difference (p>0.05) in feed intake among the four experimental groups. However, in weeks 3 and 4, groups C and D exhibited significantly higher (p<0.05) feed intake compared to groups A and B, with no significant difference (p>0.05) observed between

groups C and D. In week 3, group B showed significantly higher (p<0.05) feed intake than group A, whereas no significant difference (p>0.05) was noted between the two groups in weeks 4 and 5. In week 5, group C recorded the highest feed intake, which was significantly higher (p<0.05) compared to the other three groups. By week 6, no significant difference (p>0.05) in cumulative feed intake was observed among the experimental groups.

Table 5: Effect of bioactive botanical blend and standard symbiotic on average cumulative weekly feed consumption (g) in broilers

Week	Experimental groups				
vv eek	A	В	С	D	
1	1 172.88±1.81 173.35±1.22		174.69±1.54	173.88±1.95	
2	558.28±4.82	551.58±1.25	550.95±2.18	551.94±1.81	
3	1131.06°±10.67	1156.90 ^b ±8.56	1203.06 ^a ±7.98	1198.41 ^a ±2.53	
4	1946.14 ^b ±13.05	1954.64 ^b ±9.57	2046.87 ^a ±13.33	2032.09 ^a ±5.30	
5	3123.94°±16.13	3125.90°±7.61	3251.48 ^a ±4.96	3192.27 ^b ±10.07	
6	4425.96±44.83	4467.95±30.78	4555.58±12.06	4511.50±24.70	

Average cumulative weekly feed conversion ratio (FCR)

Table 6 shows the effect of bioactive botanical blend and standard symbiotic on average cumulative feed conversion ratio in broilers. In week 1 and week 3 there was no significant (p>0.05) difference in FCR among four different experimental groups. In week 2 groups B, C and D were showed no significant (p>0.05) difference, whereas only groups C and D showed significant (p<0.05) difference compared with control group A. In week 4 groups C and

D showed significantly (p<0.05) improved FCR compared with groups A and B. In week 5, there was no significant (p>0.05) difference between groups B, C and D, however these groups showed significant (p<0.05) difference compared with control group A. In week 6, four different experimental groups exhibited no significant (p>0.05) difference in FCR. At the sixth week, the average cumulative FCR improved by 3.91%, 4.77%, and 4.20% in groups B, C, and D, respectively, compared to the control group A.

Table 6: Effect of bioactive botanical blend and standard symbiotic on average cumulative weekly feed conversion ratio (FCR) in broilers

Week	Experimental groups				
VV CCK	A	В	C	D	
1	1.117±0.01	1.073±0.03	1.065±0.01	1.061±0.02	
2	1.227 ^a ±0.02	1.189 ^{ab} ±0.01	1.168 ^b ±0.01	1.182 ^b ±0.01	
3	1.307±0.01	1.294±0.01	1.274±0.01	1.273±0.00	
4	1.412 ^a ±0.01	1.407°a±0.01	1.371 ^b ±0.00	1.380 ^b ±0.01	
5 1.610 ^a ±0.0		1.582 ^b ±0.01	1.576 ^b ±0.00	1.568 ^b ±0.01	
6	1.740±0.01	1.672±0.01	1.657±0.00	1.667±0.13	

Intestinal histomorphology parameters

Table 7 presents the effect of bioactive botanical blend and standard symbiotic on intestinal histomorphology parameters in broilers. The villi length and V/C ratio of duodenum, jejunum and ileum found significant (p<0.05) across the four different treatment groups. Group C, exhibited highest (p<0.05) villi length and V/C ratio followed by groups D, B and A. For duodenal villi length, group C, which was supplemented with BBB at 100g/100 kg of feed, showed increases of 9.95%, 5.18%, and 4.49% compared to groups A, B, and D, respectively. For jejunal villi length, it showed increases of 10.98%, 5.39%, and 4.40% over groups A, B, and D, respectively. For ileal villi length, group C showed higher (p<0.05) values by 12.83%, 5.19%, and 4.59% in comparison with groups A, B, and D, respectively. Group C exhibited a higher (p<0.05) duodenal V/C ratio by 39.03%, 15.19%, and 9.24% compared to

groups A, B, and D, respectively. Similarly, the jejunal V/C ratio in group C was elevated by 21.71%, 8.52%, and 3.44% relative to groups A, B, and D. In the ileum, group C showed an increase in V/C ratio by 30.52%, 3.65%, and 0.47% in comparison with groups A, B, and D, respectively. Duodenal crypt depth of birds from different experimental groups displayed significant (p<0.05) difference. The crypt depth of duodenum of group C was 20.87%, 8.69% and 4.36% lower in comparison with groups A, B and D, respectively. Group C exhibited significantly (p<0.05) lower duodenal crypt depth in comparison with other three different experimental groups. Group D exhibited significantly (p<0.05) lower jejunal crypt depth compared with other three groups, succeeded by groups C, B and A. Ileal crypt depth was lower (p<0.05) in group D, trailed by groups B, C and A.

Table 7. Effect of bioactive botanical blend and standard symbiotic on average histomorphology parameters (µm) of small intestine in broilers

Histomorphology	Experimental groups			
parameters	A	В	С	D
Duodenal villi length	1787.02 ^d ±3.89	1868.19°±0.34	1964.88 ^a ±0.42	1880.53 ^b ±0.22
Jejunal villi length	1684.83 ^d ±0.12	1774.30°±0.20	1869.92 ^a ±0.38	1791.05 ^b ±0.30
Ileum villi length	1065.34 ^d ±0.36	1142.80°±0.24	1202.06 ^a ±0.68	1149.34 ^b ±0.49
Duodenal crypt depth	280.09°±0.17	242.64 ^b ±0.07	221.55d±0.35	231.64°±0.35
Jejunal crypt depth	243.78°±0.52	228.89 ^b ±0.48	222.44°±0.27	220.37 ^d ±0.20
Ileal crypt depth	163.47 ^a ±0.47	139.14°±0.22	141.23 ^b ±0.33	135.68 ^d ±0.19
Duodenal V/C ratio	6.38 ^d ±0.01	$7.70^{\circ} \pm 0.00$	8.87a±0.01	8.12 ^b ±0.01
Jejunal V/C ratio	6.91 ^d ±0.01	7.75°±0.02	8.41a±0.01	8.13 ^b ±0.01
Ileal V/C ratio	$6.52^{d} \pm 0.02$	8.21°±0.01	8.51a±0.02	8.47 ^b ±0.01

Carcass traits

Table 8 presents the effect of bioactive botanical blend and standard symbiotic on average carcass traits in broilers. Dressed weight, both with and without giblets, was significantly higher (p<0.05) in groups B, C, and D in comparison with control group A. Furthermore, groups C and D exhibited greater (p<0.05) dressed weight with giblets than group B. Regarding breast meat weight, all treatment groups B, C, and D found a significant increase (p<0.05) in comparison with the control group A. Non-significant (p>0.05) differences were found across the groups for giblet weight and dressing percentage with giblets. However, dressing percentage without giblets was significantly (p<0.05) higher in groups B and C against the control group A, while group D did not

differ significantly (p>0.05) from the control. Dressing percentage without giblet was higher by 3.05% and 3.00% in groups B and C, respectively, in comparison with control group A. For breast meat weight percentage groups C and group D showed significant (p<0.05) difference compared with control group A, whereas group B showed no significant (p>0.05) difference. Breast meat weight percentage increased by 8.34% and 6.42% in groups C and D, respectively, in contrast to the control group A. Breast meat weight percentage in group C was 1.81% higher compared to group D. The giblet weight percentage in the control group A was significantly (p<0.05) higher in comparison with groups B and D (P<0.05), but did not differ significantly (p>0.05) from group C.

Table 8: Effect of bioactive botanical blend and standard symbiotic on average carcass traits (g) in broilers

Carcass traits	Experimental groups				
Carcass traits	A	В	C	D	
Live weight	2644.00°±39.46	2964.96 ^b ±26.87	3272.04 ^a ±65.54	3164.00°±24.48	
Dressed weight with giblet	1908.08°±30.89	2167.00b±21.38	2396.00°a±40.78	2301.25°a±24.85	
Dressed weight without giblet	1718.04°±30.94	1984.54 ^b ±23.32	2188.54a±38.13	2104.67 ^{ab} ±20.49	
Breast meat weight	565.46°±7.58	660.96 ^b ±10.83	757.25 ^a ±21.74	717.96 ^{ab} ±12.80	
Giblet weight	190.04±8.67	182.46±5.00	207.46±4.76	196.58±7.32	
Dressing percentage with giblet	72.13±0.12	73.08±0.45	73.23±0.44	72.74±0.29	
Dressing percentage without giblet	64.95 ^b ±0.40	66.93°a±0.36	66.90°±0.49	66.54 ^{ab} ±0.33	
Breast meat weight%	21.34 ^b ±0.16	22.30ab±0.28	23.12a±0.24	22.71a±0.30	
Giblet weight%	7.18 ^a ±0.32	6.15 ^b ±0.21	$6.33^{ab}\pm0.08$	6.20 ^b ±0.19	

Discussion

Growth performance parameters

During whole experimental period, birds supplemented with BBB at dose of 100g/100kg of broiler feed performed similar (p<0.05) with group provided with standard symbiotics at inclusion of 25g/100kg of feed. Both these groups showed significantly (p<0.05) higher LBW in comparison with the group supplemented with 50 g BBB/100 kg of feed and the control group. It might occur due to maximum nutrient absorption by the small intestinal villi. Furthermore, all three treatment groups, including those receiving 50 g and 100 g of BBB as well as 25 g of standard symbiotic per 100 kg of feed, demonstrated significantly (p<0.05) improved total BWG in comparison with the control group. At sixth week of experimental period, all the four treatment groups found non-significant (p>0.05) for cumulative FI and FCR.

Our findings were in accordance with those, who showed that due to the supplementation of Curcuma longa powder at dose of around 4 or 6 g/kg of feed significantly (p<0.05) increased LBW and total BWG against the control group. Similarly, nonsignificant (p>0.05) difference was seen in FI and FCR during whole experimental period [19]. Conversely, other researchers noted that inclusion of curcumin at a level of 200 mg/kg of feed resulted in a significantly (p<0.05) greater FCR in comparison with the control group during the time periods of 15 to 28 days and 1 to 28 days of age [20]. In accordance with Islam et al., (2023) [21], who demonstrated that supplementation with mixture of 1% clove powder and 3% Ocimum sanctum in drinking water significantly (p<0.05) enhanced final LBW and BWG in broilers. In divergence, due to the provision of onion extracts at concentration of 5, 10, 15, or 20 g per kg of broiler feed improved the growth performance [22]. On the contrary, supply of piperine at doses of 60, 120, and 180 mg per kg of feed exhibited non-significant (p>0.05) effect on FI, BWG, or FCR [23]. In accordance with our results, Rahman et al., (2017) [24] observed a significant (p<0.05) difference in LBW due to the supplementation of onion powder at doses of 1.5, 2, and 2.5 g per kg of feed. In contrast, FI was more (p<0.05) in comparison with the control group.

Intestinal histomorphology parameters

Efficient nutrient absorption is closely associated with increase in height of villi and reduction in depth of crypt of an intestine. Thus, availability of nutrients is enhanced by thyme essential oil by positively influencing the intestinal villus structure, including its height, width, and crypt depth [25]. Longer villi in the small intestine provide more surface area for the absorption of nutrients [26]. In small intestine the occurrence of morphological changes such as the enhancement in the villus height and ratio of V/C and reduction in crypt depth may occur due to the extracts that are present in BBB and also due to standard symbiotic contents. The decrease in crypt depth suggests a reduction in epithelial cell turnover; it is linked to enhanced intestinal efficiency and decreased metabolic energy demand for mucosal regeneration [27]. In the present study dietary inclusion with a BBB at 100 g/100 kg of feed significantly (p<0.05) improved the length of villus and ratio of V/C in the duodenum, jejunum, and ileum in comparison with control group, along with the groups receiving 25 g of standard symbiotic/100 kg or 50 g of BBB/100 kg of feed. Duodenal crypt depth was significantly reduced with 100 g BBB/100 kg of feed, while jejunal and ileal crypt depths were lower with 25 g standard symbiotic/100 kg of feed compared to other groups.

Our findings aligned with Guo et al., (2023) [20], who showed that curcumin supplementation at a dose of 200 mg/kg of feed significantly improved V/C ratio in comparison with control group. Similarly, Rehman et al., (2017) indicated that the incorporation of onion powder significantly increased villus height; by contrast, the control group exhibited significantly lower small intestinal crypt depth compared to the supplemented groups [24]. Likewise, Ghazanfari et al., (2023) [28] found that increasing the concentration of ajwain essential oil to 300 ppm significantly (p<0.05) enhanced the ratio of V/C in the jejunum of broiler chickens. Our findings aligned with Islam et al., (2023) [21] in reporting increased (p<0.05) duodenal, jejunal and ileal height of villus with 1% clove and 3% Ocimum sanctum. However, contrary to our findings, they observed non-significant (p>0.05) difference in the V/C ratio. Likewise, Cardoso et al., (2012) [23] reported a significant (p<0.05) improvement in the crypt depth of duodenum and ileum following the provision of piperine at concentrations of 60, 120, and 180 mg per kg of feed, in comparison with the control group. Improvements (p<0.05) in jejunal and ileal height of villus were also recorded. In line with Omar et al., (2020) [29], inclusion of onion extracts rich in phenolic compounds at doses of 2 g and 3

g/kg of feed significantly improved villus height and width in the small intestine.

Carcass traits

BBB improved the dressing percentage without giblets, but no effect was seen on dressing percentage with giblets. BBB and standard symbiotic both showed improvement in breast meat weight. The enhancement in breast muscle weight could be linked to curcumin, the principal bioactive compound in Curcuma longa (turmeric powder), which stimulates the digestive system and enhances nutrient utilization in poultry [30]. Our findings aligned with earlier studies that demonstrating a significant (p<0.05)increase in breast meat weight in broilers provided with turmeric powder at 7 g/kg of the feed [31]. Varying with our results, thyme essential oil at the doses of 0.025%, 0.075% and 0.125% as well as a prebiotic at a dose of 0.075% had no effect (p>0.05) on breast meat weight in comparison with control group [32]. In contrast, dressed weight displayed no significant (p>0.05) from the provision of Ocimum sanctum powder at doses of 0.5%, 1% and 1.5% in quail [33].

Our findings were in accordance with previous studies indicating that mango saponin supplementation at doses of 0.14% and 0.28% had no effect on dressing percentage; however, our results were inconsistent with earlier findings that reported no effect on breast meat weight [34]. Likewise, Rathor et al., (2025) [35] reported that 1.5% Ajwain supplementation in broiler diet resulted in significantly (p<0.01) higher yield of carcass and eviscerated parts when compared against the control group. Similarly, turmeric powder had no effect on carcass yield% [19]. In contrast, the dressing percentage was improved by the inclusion of piperine at doses of 50 and 100 mg/kg of feed [36]. Inconsistent with our findings, other researchers found that the provision of onion extracts had no variation in weight of carcass [22]. In line with Urusan and Bolukbasi (2017) [37], who noted a significant (p<0.05) difference in weight of carcass, but not in carcass yield, following the incorporation of powdered form of Curcuma longa at doses of 2, 4, 6, and 8 g/kg of feed.

Conclusion

It is evident from the present research trial that, the incorporation of BBB at dose of $100\,\mathrm{g}/100\,\mathrm{kg}$ of feed was as effective as the standard symbiotic (25 g/100 kg) in improving growth and carcass traits. However, the BBB at dose of $100\mathrm{g}/100\,\mathrm{kg}$ feed, resulted in improved intestinal histomorphology of experimental birds as compared to the standard symbiotic. This improvement in gut structure indicates a positive impact of the BBB on overall gut health, potentially supporting better nutrient absorption and immune function.

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