



Effect of different combinations of probiotic, chicory root powder and coriander seed powder on growth, carcass, immunity, serum parameters and economics of broilers

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ABSTRACT

The trial was undertaken in a completely randomized design to evaluate the synergistic effect of probiotic (Pro), chicory root powder (CRP) and coriander seed powder (CSP) on the performance of broiler chicken. A total of 240 day-old broiler chicks were randomly allotted to six dietary treatments with 8 replicates of 5 birds in each. Treatment groups included T₁ as control, i.e. basal diet (BD) without any growth promoter and T₂ - BD + antibiotic (BMD @ 500 gm/ton). In the remaining experimental diets, T₃ - pro (@ 10 gm/100 kg) + CRP (@ 1.0%), T₄ - pro (@ 10 gm/100 kg) + CSP (@ 1.5%), T₅ - CRP (@ 1.0%) + CSP (@ 1.5%) and T₆ - pro (@ 10 gm/100 kg) + CRP (@ 1.0%) + CSP (@ 1.5%). The results revealed that supplementation of pro + CRP significantly increased the body weight gain during prestarter, starter and finisher phase and improved the feed conversion ratio during finisher phase compared to other treatment groups. Supplementation of different dietary groups did not show any significant effect on feed intake and various slaughter parameters of broilers. Whereas, the humoral immune response to ND vaccine and immune organ weights were significantly higher in all the test diets (T₃ to T₆) compared to control and antibiotic groups. Supplementation of probiotic with chicory combination group significantly reduced the serum total cholesterol and recorded higher returns over feed cost compared to other treatment groups. It can be concluded that combination of probiotic (10 g) with chicory root powder (1.0%) proved more effective than combinations of other additives.

Keywords: Body weight, Coriander seed powder, Immunity, ND titers, Probiotic, Serum cholesterol

Antibiotics are being used as growth promoters in the poultry diets all over the world. However, in recent years, there has been rising demand to reduce or abolish the use of antibiotics as growth promoters due to the detrimental human health issue of antibiotic resistance (Dibner and Richards 2005). Consumers' awareness of poultry products that do not contain antibiotic residues has increased, encouraging the use of suitable alternatives to antimicrobial compounds. Among the feed additives, probiotics, prebiotics, and enzymes have drawn more attention due to their prophylactic and growth promoting effects can be used as an alternative to antibiotics for achieving better performance (Huyghebaert *et al.* 2011).

Probiotics are single or mixed cultures of live microorganisms fed along with feed which beneficially affect the host by improving the balance of intestinal flora (Fuller 1989). Dried chicory root powder is a good source

of inulin type fructans and oligofructose chains known for having prebiotic action without any toxicity (Barbara *et al.* 2007). Symbiotics are combination of probiotics and prebiotics that improves performance of broilers (Utami and Wahyono 2019). Prebiotics serve as a nutrient source for probiotic bacterial cultures for early colonization in the intestine. The advantage of the combination is that certain substrates are available for fermentation, increasing the viability of probiotic bacteria. Therefore, the body will more fully benefit from the combination of probiotics and prebiotics (Karwan *et al.* 2016). Coriander seed powder contains 0.5%–1.0% essential oil (carvone, geraniol, limonene, borneol, camphor, elemol and linalool) having antimicrobial properties against food borne pathogen such as *Salmonella* species (Silva *et al.* 2011). In addition, it has appetizing and stimulatory effects in the digestion process by increasing production of digestive enzymes and juices, which stimulates digestion and peristaltic motion, thus improves feed efficiency (Rajeshwari and Andallu 2011). Although many previous studies showed various levels of probiotics, chicory root powder and coriander seed powder on performance of broilers, work done on effect of

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combination of the above feed additives is meager. Hence, this research trial was taken up with an aim to study the effect of different combination of the above feed additives on performance, immunity and serum parameters of broilers.

MATERIALS AND METHODS

The experiment was conducted from January 2020 to February 2020 at Department of Poultry Science and Poultry Experimental Station, College of Veterinary Science, Rajendranagar, Hyderabad. To conduct the study, 240 day old commercial (Vencobb 400) broiler chicks were procured, individually weighed and wing banded. The birds were distributed randomly into 6 dietary treatments, each with 8 replicates having 5 chicks in each replicate. The chicks were reared in battery brooders under standard managemental conditions. The experimental period was from day old to 42 d of age. The birds were fed with maize and soybean meal-based diets containing 2964, 3075 and 3167 kcal ME and 22.90, 21.65 and 19.65% crude protein, respectively during prestarter (0-14d), starter (15-28d) and finisher (28-42d) phases. Treatment groups included T_1 as control, i.e. basal diet (BD) without any growth promoter and T_2 - BD + antibiotic (Bacitracin Methylene Disalicylate at 500 gm/ton – manufacturer Zoetis). In the remaining experimental diets, T_3 - probiotic (@ 10 gm/100 kg) + chicory root powder (@ 1.0%), T_4 - probiotic (@ 10 gm /100 kg) + coriander seed powder (@ 1.5%), T_5 - chicory root powder (@ 1.0%) + coriander seed powder (@ 1.5%) and T_6 - probiotic (@ 10 gm/100 kg) + chicory root powder (@ 1.0%) + coriander seed powder (@ 1.5%). Probiotic contains lyophilized and microencapsulated *Bacillus coagulans*, *Saccharomyces boulardii*, *Lactobacillus acidophilus*, *Lactobacillus delbrueckii*, *Lactobacillus plantarum*, *Streptococcus thermophilus*, *Bacillus subtilis*, *Enterococcus faecium*, *Bifidobacterium bifidum* (at the concentration of 32 billion CFU /100 g). The chemical composition of chicory root powder and coriander seed powder is given in Table 1.

Table 1. Chemical composition of Chicory root powder

Composition (%)	Chicory root powder	Coriander seed powder
Moisture	3.16	3.23
Crude protein	14.55	14.91
Fat	1.76	0.96
Ash	3.98	8.88
Crude fibre	30.01	34.21
Total carbohydrates	48.76	50.12
Inulin	46.89	-

Weekly body weight, feed intake and feed conversion ratios were calculated as feed intake per unit bodyweight gain at weekly intervals. The mortality rate was recorded throughout the experiment. On day 42, one bird from each replicate was sacrificed to estimate the various carcass parameters. The humoral immunity was estimated in birds

by measuring antibody titer to Newcastle disease (ND) vaccine (antibody production against ND virus). Broilers were vaccinated against ND by ocular route at 7th and 28th day of age with LaSota strain (ND Lasota Vac-500; Indovax Pvt.Ltd. Hyderabad, India). At 42nd day of age blood was collected, and serum was separated. Haemagglutination inhibition (HI) activity of serum was estimated, and the antibody titers (log₂) were measured by following the standard procedure (Wegmann and Smithies 1966).

On 42nd day, blood sample were collected aseptically from wing vein (one bird from each replicate) in vacutainers and kept in incubator at room temperature for serum collection. The serum is used for estimation of cholesterol, total protein, albumen globulin and antioxidant activity by using spectrophotometer with commercially available kits (ERBA diagnostic Mannheim-gmbh transasia bio-medicals Limited).

Data were analyzed for mean, standard errors, and analysis of variance using the Snedecor and Cochran (1989) method, and means were compared using Duncan test (1955) using the Statistical Package for Social Sciences (SPSS) 20.0 version software, with significance set at $P < 0.05$.

RESULTS AND DISCUSSION

Performance parameters: The data on body weight gain, feed intake (FI) and feed conversion ratio (FCR) as influenced by different dietary treatments is shown in Table 2. During prestarter (0-14 d) phase, significantly ($P < 0.05$) higher body weight gain was recorded in probiotic + chicory (T_3) and chicory + coriander (T_5) combination groups followed by probiotic + coriander (T_4) group. However, no significant ($P > 0.05$) difference was recorded among antibiotic (T_2), control (T_1), and probiotic + chicory + coriander (T_6) combination groups. During starter (15-28 d) phase, there was no significant ($P > 0.05$) difference among probiotic + coriander (T_4), probiotic + chicory (T_3), chicory + coriander (T_5) and antibiotic (T_2) groups, but they had significantly ($P < 0.05$) higher body weight gain compared to probiotic + chicory + coriander (T_6) and control (T_1) groups. During finisher (29-42 d) phase, significantly ($P < 0.05$) higher body weight gain was recorded in probiotic + chicory (T_3) group followed by probiotic + chicory + coriander (T_6), probiotic + coriander (T_4), chicory + coriander (T_5), antibiotic (T_2) and control (T_1) groups. In agreement with these results, Taherpour *et al.* (2009) indicated that supplementation of probiotic and prebiotic combination improved the body weight of broilers. Similarly, Kirkpinar *et al.* (2018) reported that probiotic + prebiotic and probiotic + enzyme combination increased body weight of broilers when compared with control. Similarly, Sanja *et al.* (2015) reported addition of synbiotics (*Enterococcus faecium* + fructooligosaccharides) improved the body weight of broilers. Supplementation of probiotics and inulin combinations significantly ($P < 0.05$) improved body weight gain in broilers was reported by Karwan *et al.* (2016). The complimentary effect of probiotic

Table 2. Synergistic effect of probiotic, chicory root powder and coriander powder on phase wise performance of broiler chicken

Trt	Diet	Body weight gain (g)			Feed intake (g)			Feed conversion ratio (FCR)		
		Pre-starter (0-14 d)	Starter (15-28 d)	Finisher (29-42 d)	Pre-starter (0-14 d)	Starter (15-28 d)	Finisher (29-42 d)	Pre-starter (0-14 d)	Starter (15-28 d)	Finisher (29-42 d)
T ₁	Control	216 ^c	789 ^c	969 ^d	232	1119	2182	1.08	1.42	2.25 ^d
T ₂	Antibiotic	215 ^c	850 ^{ab}	1041 ^c	214	1173	2179	1.00	1.38	2.09 ^c
T ₃	Probiotic + Chicory	229 ^a	860 ^{ab}	1096 ^a	227	1163	2163	0.99	1.35	1.98 ^a
T ₄	Probiotic + Coriander	223 ^b	864 ^a	1063 ^{bc}	223	1169	2172	1.00	1.35	2.04 ^{bc}
T ₅	Chicory + Coriander	232 ^a	859 ^{ab}	1053 ^c	221	1167	2173	0.95	1.36	2.06 ^{bc}
T ₆	Probiotic + Chicory + Coriander	216 ^c	843 ^b	1081 ^{ab}	214	1158	2172	0.99	1.37	2.01 ^{ab}
SEM		1.139	4.330	6.657	2.847	9.710	9.975	0.0131	0.0107	0.0108
N		8	8	8	8	8	8	8	8	8
P-value		0.001	0.001	0.001	0.358	0.652	0.701	0.170	0.453	0.001

Values bearing different superscripts within a column are significantly ($P < 0.05$) different. Trt, Treatment.

and chicory powder on body weight gain as observed in the present study might be due to better microbial environment in gut by competitive exclusion and antagonism (Nurmi and Rantala 1973). The fermentation activity of inulin inhibits the growth of *Escherichia coli* and *Salmonella*, selectively stimulate the growth of *Bifidobacteria* and *Lactobacillus* development in the gut (Yusrizal and Chen 2003). Contrary to above results, Hofacre *et al.* (2003) and Al-Khalaifa *et al.* (2019) did not find positive effect on body weight in broilers fed with prebiotic + probiotic combination at 28 d] of age.

The ANOVA revealed that there were no significant ($P > 0.05$) differences in FI among different dietary treatments during pre-starter (0-14 d), starter (15-28 d), finisher phase (29-42d). In line with the findings of the present study, Ashayerizadeh *et al.* (2009) and Al-Khalaifa *et al.* (2019) reported that supplementation of probiotic + prebiotic combination did not have significant ($P > 0.05$) on FI of broilers. Similar results were also reported by Kirkpinar *et al.* (2018) and Utami and Wahyono (2019). These variations may be due to environmental factors and levels of the additives used in the experiment.

Statistical analysis on feed conversion ratio (FCR) values revealed that there was no significant ($P > 0.05$) difference among different dietary treatments during prestarter (0-14 d) and starter (15-28 d) phases. However, during finisher (29-42 d) phase, significantly ($P < 0.05$) better feed conversion ratio was recorded in probiotic + chicory (T₃) combination group compared to other treatments. However, no significant ($P > 0.05$) difference was observed among probiotic + chicory + coriander (T₆) group, antibiotic (T₂), probiotic + coriander (T₄) and chicory + coriander (T₅) groups but they had better FCR than control group (T₁). The increased FCR in probiotic + chicory (T₃) combination group might be due to symbiotic relation between chicory inulin and probiotic. Chicory root powder inulin serves as a source of nutrient for the probiotic bacterial cultures for early establishing in the gut. Similar results were reported by Szakacs *et al.* (2015) and Sanja *et al.* (2015) who stated that probiotic + prebiotic combination

improved feed efficiency in broilers. Ashayerizadeh *et al.* (2009) reported that addition of antibiotic, probiotic + prebiotic combination improved FCR compared to control. Improved feed efficiency with probiotic 0.4% + prebiotic 0.2% was also reported by Utami and Wahyono (2019). However, in contrary to our findings, Kirkpinar *et al.* (2018) and Al-Khalaifa *et al.* (2019) did not find positive effect of probiotic and prebiotic combination on FCR of broilers.

Mortality: Mortality was not influenced by supplementation of different dietary groups at 42 d of age. However, mortality rate was within the acceptable range.

Carcass parameters: The dietary treatments did not to show any significant ($P > 0.05$) difference in slaughter parameters at 42 d of age (Table 3). In agreement with the results of this study, several researchers demonstrated that addition of probiotics (Ray *et al.* 2019), chicory root powder (Praveen *et al.* 2017) and coriander seed powder (Naeemasa *et al.* 2015 and Reddy *et al.* 2019) did not result in any significant ($P > 0.05$) effect on slaughter parameters of broilers. On the contrary, few studies indicated significant ($P < 0.05$) effect on slaughter parameters of broilers with probiotics (Panda *et al.* 1999), chicory powder (Izadi *et al.* 2013) and coriander seed powder (Taha *et al.* 2019).

Immunity: Significant difference ($P < 0.05$) was observed in the relative weights of bursa and spleen at 42 d of age (Table 4). Probiotic + coriander combination (T₄) had higher bursa and spleen weight among all the treatments. No significant ($P > 0.05$) difference was observed among probiotic + chicory (T₃), chicory + coriander (T₅) and probiotic + chicory + coriander (T₆) groups but they had higher bursa and spleen weight compared to control (T₁) and antibiotic (T₂) groups indicating better immunity in all the test groups. However, there was no significant ($P > 0.05$) difference in thymus weight among different dietary groups. However, supplementation of all the dietary treatments (T₃ to T₆) significantly ($P < 0.05$) increased the antibody response to ND vaccine (HI activity) compared to control and antibiotic groups (Table 5). The increased antibody production against NDV in probiotics, coriander

Table 3. Synergistic effect of probiotic, chicory root powder and coriander powder on slaughter parameters (% live weight) of broilers

Trt	Diet	Eviscerated yield	Liver	Heart	Gizzard	Giblet yield	Ready to cook yield	Abdominal fat
T ₁	Control	60.37	2.37	0.55	1.91	4.83	65.20	1.42
T ₂	Antibiotic	60.38	2.31	0.52	1.98	4.82	65.18	1.32
T ₃	Probiotic + Chicory	61.85	2.33	0.60	1.95	4.88	66.73	1.23
T ₄	Probiotic + Coriander	60.35	2.46	0.59	2.02	5.07	65.42	1.35
T ₅	Chicory + Coriander	61.90	2.14	0.56	1.94	4.64	66.54	1.28
T ₆	Probiotic + Chicory + Coriander	62.10	2.13	0.54	2.01	4.69	66.78	1.21
SEM		0.158	0.041	0.010	0.026	0.062	0.544	0.038
N		8	8	8	8	8	8	8
P-value		0.814	0.112	0.125	0.821	0.399	0.897	0.622

Values bearing different superscripts within a column are significantly ($P < 0.05$) different. Trt, Treatment.

Table 4. Synergistic effect of probiotic, chicory root powder and coriander powder on Immune organ weights (% live weight) and NDV titers ($\log_2$) of broiler chicken

Trt	Diet	Bursa	Spleen	Thymus	NDV titers ($\log_2$)
T ₁	Control	0.076 ^d	0.112 ^c	0.812	2.85 ^c
T ₂	Antibiotic	0.080 ^c	0.105 ^c	0.828	3.06 ^b
T ₃	Probiotic + Chicory	0.092 ^b	0.139 ^b	0.907	3.26 ^a
T ₄	Probiotic + Coriander	0.105 ^a	0.176 ^a	0.947	3.39 ^a
T ₅	Chicory + Coriander	0.089 ^{bc}	0.133 ^b	0.879	3.27 ^a
T ₆	Probiotic + Chicory + Coriander	0.089 ^{bc}	0.131 ^b	0.858	3.42 ^a
SEM		0.0020	0.0039	0.0168	0.349
N		8	8	8	8
P-value		0.001	0.001	0.183	0.002

Values bearing different superscripts within a column are significantly ($P < 0.05$) different. Trt, Treatment.

powder and chicory root powder might be due to enhanced macrophage activity and increased local antibodies (IgA) at mucosal surfaces (Huang *et al.* 2004). The weight of spleen, thymus and bursa is directly proportional to immune response in broilers (Kabir *et al.* 2004). The higher lymphoid organs weight (bursa, spleen and thymus) also supports the increased immune response in

test groups. Probiotic bacteria are capable of enhancing both specific and nonspecific immune responses by activating macrophages, increasing cytokine production by intraepithelial lymphocytes, and increasing the levels of immunoglobulins (Huang *et al.*, 2004).

Serbio-chemicals: Among all the treatment groups, probiotic + chicory combination group recorded significantly ($P < 0.05$) lower cholesterol level at 42 days of age. However, serum cholesterol levels in other groups were intermediate and comparable with antibiotic group and lower than control group (Table 5). In agreement with the above results, Ashayerizadeh *et al.* (2009) observed decreased serum cholesterol and triglycerides concentration with probiotic + prebiotic combination in broilers. Similarly, Taherpour *et al.* (2009) reported that probiotic + prebiotic and prebiotic + butyric acid combination lowered the serum cholesterol compared to control. Similarly, Yousfi *et al.* (2017) reported that supplementation of chicory leaf extract + probiotic combination significantly ($P < 0.05$) decreased serum HDL and cholesterol compared to control and antibiotic. They also stated that the decreased cholesterol level might be due to the properties of chicory leaf extract to stimulate lactic acid producing bacteria which secretes hydrolase enzyme and that in turn converts bile salts into deconjugated bile acids and ultimately resulted in the reduced serum cholesterol level.

Supplementation of all the dietary groups did not show any significant ($P > 0.05$) effect on total protein, albumin,

Table 5. Synergistic effect of probiotic, chicory root powder and coriander powder on serum biochemical parameters of broilers at 42 d of age

Trt	Diet	Serum total cholesterol (mg/dL)	Total Protein (g/dL)	Albumin (g/dL)	Globulin (g/dL)	A/G ratio
T ₁	Control	220 ^c	4.94	2.04	2.05	1.02
T ₂	Antibiotic	189 ^b	4.97	2.09	2.03	1.03
T ₃	Probiotic + Chicory	137 ^a	5.04	2.11	2.08	1.06
T ₄	Probiotic + Coriander	179 ^b	4.94	2.18	1.91	1.16
T ₅	Chicory + Coriander	181 ^b	5.03	2.11	2.06	1.08
T ₆	Probiotic + Chicory + Coriander	182 ^b	4.91	2.10	1.97	1.09
SEM		4.57	0.029	0.027	0.041	0.034
N		8	8	8	8	8
P-value		0.001	0.769	0.803	0.849	0.878

Values bearing different superscripts within a column are significantly ($P < 0.05$) different. Trt, Treatment.

globulin and albumin/globulin ratio (A/G ratio). Similarly, Ashayerizadeh *et al.* (2009) did not find any significant ($P>0.05$) difference in serum total protein, albumin and globulin with probiotic + prebiotic supplementation in broilers. Alam and Ferdaushi (2018) reported that supplementation of probiotic did not have significant ($P>0.05$) on serum total protein, albumin, globulin, and A/G ratio.

Economics: Significantly ($P<0.05$) higher returns were recorded in probiotic + chicory (T_3) followed by probiotic + coriander (T_4), chicory + coriander (T_5) and probiotic + chicory + coriander (T_6) groups compared to control (T_1) and antibiotic (T_2) groups (Supplementary Table 1 and 2). The highest profit per bird was recorded with probiotic + chicory (₹11.88 / bird) followed by probiotic + coriander (₹8.60/bird), chicory+coriander (₹6.55/bird) and probiotic + chicory + coriander (₹6.36 / bird) groups. Similarly, Rout *et al.* (2016) reported that supplementation of broilers with probiotic, organic acid alone and combination increased the net return per kg live weight gain compared to control. Osfar *et al.* (2019) recorded highest income over feed cost in prebiotic supplemented diet compared to the control. Increase in the net profit of the broilers fed with probiotic + chicory (T_3) combination group might be attributed to symbiotic effect of probiotic and chicory root powder results in improved weight gain and better FCR leading to more returns over feed cost.

The results showed that combination of probiotic (10 g) with chicory root powder (1.0%) proved more effective than combinations of other additives in terms of body weight gain, FCR, lowered serum cholesterol and better returns over feed cost. However, supplementation of different feed additives did not show any influence on feed intake, carcass, total protein, albumin and globulin levels of broilers. Therefore, probiotic and chicory root powder combination can be used in place of antibiotic growth promoter in broilers.

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