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# Effects of Probiotics and Prebiotics on Growth Performance of Commercial Broiler

Research Article

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## ABSTRACT

This experiment was conducted to investigate the effects of probiotics and prebiotics on growth performance, carcass yield, immune parameters, and microbial content in the commercial broiler. A total of 225 one-day-old chicks of the "Lohman Meat (Indian River)" strain having 42.66 ± 0.66 g average body weight were divided into 5 experimental groups with 3 replications of 15 chicks each. The treatments were $T_0$= control (the basal diet), $T_1$= antibiotic, $T_2$= probiotic, $T_3$= prebiotic and $T_4$= probiotic + prebiotic. Weekly body weight, feed consumption, and feed conversion ratio (FCR) were recorded during the experimental period. Microbial counts were studied at the end of the experiment. The average live weight and body weight gain were significantly higher in $T_4$ treatment compared to the other groups. Improved FCR was noticed in birds fed a combined addition of probiotics and prebiotics with the basal diet. Feeding broilers with probiotics and prebiotics have significant ($P<0.05$) effects on the dressing percentage, breast, thigh, back, liver, neck, heart, and gizzard while it appeared insignificant on the intestine, spleen, and bursa ($P>0.05$). Significant ($P<0.05$) difference was observed for immune parameters i.e. White blood cell (WBC), lymphocyte, and granulocyte among the treatment groups. Treatment groups found lower *Escherichia coli* and *Salmonella* numbers than the control group. Total profit per bird in group $T_4$ was significantly higher ($P<0.05$) than in groups $T_0$, $T_1$, $T_2$, and $T_3$. From this study, it can be concluded that the combined use of commercial probiotics and prebiotics resulted in improved growth performance, carcass yields, and immunity in broiler chickens. Therefore, combined usage of the probiotic and prebiotic as antibiotic alternatives in broiler production can be recommended.

KEY WORDS combined effect, commercial broiler, growth performance, prebiotics, probiotics.

## INTRODUCTION

Enteric diseases are an important concern to the poultry industry because of lost productivity, increased mortality, and the associated contamination of poultry products for human consumption. The intestinal microbiota, epithelium, and immune system provide resistance to enteric pathogens. Increased bacterial resistance to antibiotics in humans has caused an increase in public and governmental interest in eliminating sub-therapeutic use of antibiotics in livestock. Research is focused on identifying beneficial bacterial strains and substrates along with the conditions under which they are effective bacteria in the colon" (Gibson and Roberfroid, 1995). With increasing concerns about antibiotic resistance, the ban on therapeutic antibiotic usage in Europe, and the potential for a ban in the United States, there is increasing interest in finding alternatives to antibiotics for poultry production.

Prebiotics and probiotics are two of several approaches that have the potential to reduce enteric disease in poultry and subsequent contamination of poultry products. Probiotic which means "for life" in Greek (Gibson and Fuller, 2000), has been defined as "a live microbial feed supplement which beneficially affects the host animal by improving its intestinal balance" (Fuller, 1989; Poorghasemi *et al*. 2017). Prebiotics are defined as "a nondigestible food ingredient that beneficially affects the host by selectively stimulating the growth and/or activity of one or a limited number of bacteria (*Lactobacilli* and *Bifidobacteria*) have been implicated as the causative agents for this improved health. Combinations of probiotics and prebiotics are known as symbiotics. Probiotic and prebiotic foods have been consumed for centuries, either as natural components of food or as fermented foods. Their administration has become progressively popular around the world, as seen by the growing number of foods and supplements on the market that contains billions of living microbial cells (Abd Al-Fatah, 2020; Taverniti *et al*. 2021). In addition, proposed mechanisms by which probiotics and prebiotics act include competition for substrates, production of toxic compounds that inhibit pathogens, and competition for attachment sites. Extensive research conducted with humans and rodent models has shown a reduction in pathogen colonization, alteration of microbial populations, alteration of the immune system, prevention of cancer, and reduction of triglycerides, cholesterol, and odor compounds (ammonia, skatole, indole, p-cresol, and phenol) associated with probiotic and prebiotic use (Walker and Duffy, 1998; Gibson and Fuller, 2000; Simmering and Blaut, 2001). A variety of microbial species have been used as probiotics, including species of *Bacillus*, *Bifidobacterium*, *Enterococcus*, *E. coli*, *Lactobacillus*, *Lactococcus*, *Streptococcus*, a variety of yeast species, and undefined mixed cultures. *Lactobacillus* and *Bifidobacterium* species have been used most extensively in humans, whereas species of *Bacillus*, *Enterococcus*, and *Saccharomyces* yeast have been the most common organisms used in livestock (Simon *et al*. 2001). However, there has been a recent increase in research on feeding *Lactobacillus* to livestock (Gusils *et al*. 1999; Pascual *et al*. 1999; Jin *et al*. 2000; Tellez *et al*. 2001). Hossain *et al*. (2021) reported that dietary supplementation of probiotics improves the growth performance and intestinal microbial ecosystem of broiler chicken. Hassan *et al*. (2022) concluded that supplementing broilers with probiotics and Biosol, in particular, can increase their growth performance, and improve the biochemical characteristics of the blood and transcript levels of the genes. The dominant prebiotics is fructooligosaccharide products (FOS, oligofructose, inulin). However, trans-actooligosaccharides, glucooligosaccharides, glycooligosacchriades, lactulose, lactitol, maltooligosaccharides, xylo-oligosaccharides, stachyose, raffinose, and sucrose thermal oligosaccharides have also been investigated (Monsan and Paul, 1995; Orban *et al*. 1997; Piva, 1998; Collins and Gibson, 1999). Nowadays, the efficiency of poultry to convert feed into meat plays a key role in the economics of the broiler industry in Bangladesh. Therefore, it is highly essential to improve the feed efficiency of poultry to produce meat economically, and also food safety is more seriously considered than before. On the other hand, the economy of food production is also a factor. A huge amount of antibiotics has been used to control diseases and improve performances in livestock. However, due to growing concerns about antibiotic resistance and the potential for a ban for antibiotic growth promoters in many countries in the world, there is an increasing interest in finding alternatives to antibiotics in poultry production. Probiotics and prebiotics can alter the intestinal microbiota and immune system to reduce colonization by pathogens in certain conditions. The literature on the growth performance of the commercial broiler chickens fed combined probiotics and prebiotics is still limited. We hypothesized that these products can assure as alternatives for antibiotics as pressure to eliminate growth-promoting antibiotic use increases in Bangladesh. Therefore, the proposed research work is designed to investigate the effect of probiotics and prebiotics on growth performance, carcass yield, immunity, and caecal microbial content of broiler.

## MATERIALS AND METHODS

### Location of the experiment

The research work was conducted at Sher-e-Bangla Agricultural University, Poultry Farm, Dhaka, Bangladesh with a 225-day-old chick for 28 days from 31$^{st}$ October to 27$^{th}$ November, 2020 to investigate the effect of probiotics and prebiotics on growth performance, carcass traits, immune parameters, and microbial content of broilers.

### Collection of experimental broilers

A total of 225-day old chicks of the "Lohman Meat (Indian River)" strain having 42.66 ± 0.66 g average body weight was obtained from Kazi farm limited hatchery, Gazipur, Dhaka, Bangladesh.

### Preparation of experimental house

The broiler shed was an open-sided natural house with a concrete floor. The experimental room was properly cleaned and washed by using tap water. All the equipment of the broiler house was cleaned and disinfected. The house was disinfected by n-alkyl dimethyl benzyl ammonium chloride (TimsenTM) solution before starting the experiment.

After proper drying, the house was divided into pens as per the layout of the experiment. Before placement of chicks, the house was fumigated by formalin and potassium permanganate @ 500 mL formalin and 250 g potassium permanganate (i.e. 2:1) for a 35 $m^3$ experimental area.

**Experimental materials**

The collected chicks were carried to the Sher-e-Bangla Agricultural University Poultry Farm, Dhaka, Bangladesh. They were kept in electric brooders equally for 7 days by maintaining standard brooding protocol. During brooding time only a basal diet was given, no probiotics/prebiotics was used as treatment. The chicks were supplied glucose water with vitamin C to drink for the first 3 hours to overcome dehydration and transportation stress. Subsequently, small feed particles were supplied on the newspapers to start feeding for the first 24 hours. After seven days, chicks from brooders were distributed randomly in dietary treatments. After 28 days of nursing and feeding, data were collected for the following parameters: feed intake, live weight, body weight gain, feed conversion ratio, carcass characteristics, total blood count, microbial count and profit per bird.

**Experimental treatments**

Two hundred twenty-five (225) heads of day-old broiler chicks were divided into 5 experimental groups with 3 replications of 15 chicks each. The experimental layout is presented in Table 1.

**Collection of probiotics and prebiotics**

The Syn Lac probiotics and GCW prebiotics were purchased from ACI Animal Health, Bangladesh, and Advanced Chemicals Industries Limited, Bangladesh. The probiotic was supplied with the drinking water according to the company recommended level. Prebiotics were supplied to the birds with feed according to the company recommended level.

The prebiotics was supplied 1.0% of the supplied diet. The probiotic was supplied 40 g/1000 birds in drinking water/day, 20 g/1000 birds in drinking water/day, and 40 g/1000 birds in drinking water/day at the age of 1-4 days, 8-22 days, and 22 days up respectively.

**Experimental diets**

Starter and grower commercial Kazi broiler feeds were purchased from the local market of Bangladesh. The chemical composition of the diet is given in Table 2. Feed was supplied 4 times daily by following Lohman Meat (Indian River) Management Manual and *ad libitum* drinking water 2 times daily.

**Management procedures**

Fresh, clean, and sun-dried rice husk was used as shallow litter to absorb moisture from the fecal discharge of broiler chicken. About 250 g calcium oxide powder was mixed with rice husk in every pen as a disinfectant. The electric brooder was used to brood chicks. The brooding temperature was adjusted (below 35 ˚C) with house temperature. Electric fans were used as per necessity to save the birds from heat stress.

The brooding temperature was checked every 2 hours later by a digital thermometer to maintain the temperature of the brooder. The broiler shed was south-facing and open-sided. Due to wire-net cross ventilation was easy to remove polluted gases from the farm.

**Room temperature and relative humidity**

Daily room temperature (˚C) and humidity were recorded with a thermometer and a wet and dry bulb thermometer respectively which is presented in Table 3.

**Vaccination program**

The vaccines were applied to the experimental birds according to the vaccination schedule. One ampoule vaccine was mixed with purified water in agreement with the manufacturer's instructions. The birds were vaccinated on the proper schedule against new castle disease, infectious bronchitis and infectious bursal disease.

**Recorded parameters**

Weekly live weight, weekly feed consumption, and death of chicks to calculate mortality percent were taken during the study. FCR was calculated from the final live weight and total feed consumption per bird in each replication. After slaughter carcass weight and gizzard, liver, spleen, bursa, intestine, and heart were measured from each broiler chicken. The dressing yield was calculated for each replication to find out the dressing percentage.

**Immune parameter**

At the end of the experiment, a blood sample was collected randomly from each replication of every treatment. 2mL blood was collected from a wing vein with a syringe in a vacutainer. Vacutainer contains EDTA solution which prevents blood coagulants. A few hours after collection the blood sample was tested by Auto Blood Analyzer in the laboratory.

**Estimation of *Escherichia coli* (E. coli) and *Salmonella* population in broiler caecum**

At the end of the experiment, 15 birds of each treatment group were slaughtered for extraction of caecal contents.

**Table 1** Layout of the experiment

| Treatments | Arrangement of treatments | Replications | | | Total number of birds |
|---|---|---|---|---|---|
| | | 1 | 2 | 3 | |
| $T_0$ | Basal feed | 15 birds | 15 birds | 15 birds | 45 |
| $T_1$ | Basal feed + antibiotic | 15 birds | 15 birds | 15 birds | 45 |
| $T_2$ | Basal feed + probiotic | 15 birds | 15 birds | 15 birds | 45 |
| $T_3$ | Basal feed + prebiotic | 15 birds | 15 birds | 15 birds | 45 |
| $T_4$ | Basal feed + probiotic + prebiotic | 15 birds | 15 birds | 15 birds | 45 |
| **Grand total** | | 75 | 75 | 75 | **225** |

$T_0$: control (the basal diet); $T_1$: the basal diet + antibiotic; $T_2$: probiotic; $T_3$: prebiotic and $T_4$: probiotic + prebiotic.

**Table 2** Chemical composition of the basal diet (starter and grower)

| Parameter | Starter diet | Grower diet |
|---|---|---|
| Protein | 21.0 % | 19.0 % |
| Fat | 6.0% | 6.0% |
| Fiber | 5.0% | 5.0% |
| Ash | 8.0% | 8.0% |
| Lysine | 1.20% | 1.10% |
| Methionine | 0.49% | 0.47% |
| Cysteine | 0.40% | 0.39% |
| Tryptophan | 0.19% | 0.18% |
| Threonine | 0.79% | 0.75% |
| Arginine | 1.26% | 1.18% |

**Table 3** Average temperature and humidity

| Week | Date | Average temperature (˚C) | Average humidity (%) |
|---|---|---|---|
| **1st** | 31.10.2020-06.11.2020 | 33.90 | 53.14 |
| **2nd** | 07.11.2020-13.11.2020 | 30.27 | 53.00 |
| **3rd** | 14.11.2020-20.11.2020 | 30.03 | 56.72 |
| **4th** | 21.11.2020-27.11.2020 | 28.94 | 58.71 |

Eosin methylene blue (EMB) and *Salmonella shigella* (SS) agar were purchased from the local market (HIMEDIA company) of Bangladesh. The caecal content from each sample was diluted and EMB and SS agar were used to culture the *E. coli* and *Salmonella* bacteria respectively. Then, the petri dishes were sent to the bacterial growth chamber for 24 hours at 37 ˚C. The population of bacteria in each agar was estimated as CFU $g^{-1}$ (colony-forming unit).

### Statistical analysis

Total data were compiled, tabulated, and analyzed by the objectives of the study. Excel analysis by applying one-way ANOVA using Statistical Package for Social Sciences (SPSS, 2011). Differences between means were tested using Duncan's multiple comparison test, least significant difference (LSD), and significance was set at $P < 0.05$.

## RESULTS AND DISCUSSION

The present study was performed to investigate the effect of feeding probiotics and prebiotics on the growth performance of commercial broilers.

The production performances of broiler chicken were evaluated by average live weight, average live weight gain, average feed consumption (FC), feed conversion ratio (FCR), average body weight gain, weekly body weight gain, survivability, immunity parameter, and microbiological count in the caecum of birds.

Carcass characteristics were taken by dressing percentage (DP), carcass weight, and relative weight of giblet organs. The results of this research with discussion are given below.

The data presented in Table 4 showed the effect of probiotics and prebiotics on the growth performance of broiler.

**Table 4** Effects of probiotics and prebiotics on growth performances of broiler chicken

| Treatments | Average live weight (g/bird) | Average BWG (g/bird) | Average FC (g/bird) | Final FCR | Dressing (%) | Survivability (%) |
|---|---|---|---|---|---|---|
| | Mean±SE | Mean±SE | Mean±SE | Mean±SE | Mean±SE | Mean±SE |
| $T_0$ | 1806.67±2.81$^{b}$ | 1764.01±2.84$^{b}$ | 2183.33±1.63 | 1.21±0.012 | 66.63±0.33$^{b}$ | 95.56±0.83 |
| $T_1$ | 1848.33±1.25$^{a}$ | 1805.67±1.27$^{a}$ | 2241.57±0.70 | 1.21±0.017 | 69.28±0.79$^{bc}$ | 100.00±0.00 |
| $T_2$ | 1825.00±0.82$^{b}$ | 1782.34±0.83$^{b}$ | 2248.33±0.49 | 1.21±0.025 | 73.95±1.57$^{abc}$ | 100.00±0.00 |
| $T_3$ | 1828.33±1.48$^{b}$ | 1785.67±1.50$^{b}$ | 2186.67±1.08 | 1.21±0.021 | 73.08±0.19$^{ac}$ | 100.00±0.00 |
| $T_4$ | 1875.00±2.36$^{a}$ | 1832.34±2.39$^{a}$ | 2220.8±0.39 | 1.19±0.010 | 79.27±0.77$^{a}$ | 100.00±0.00 |
| Level of significance | * | * | NS | NS | * | NS |

$T_0$: control (the basal diet); $T_1$: the basal diet + antibiotic; $T_2$: probiotic; $T_3$: prebiotic and $T_4$: probiotic + prebiotic.
BWG: body weight gain; FC: feed consumption and FCR: feed conversion ratio.
The means within the same column with at least one common letter, do not have significant difference (P>0.05).
* (P<0.05).
SE: standard error.
NS: non significant.

The relative average live weight (g) of broiler chickens at the end of the 4$^{th}$ week in the dietary group $T_0$, $T_1$, $T_2$, $T_3$, and $T_4$ were 1806.67 ± 2.81, 1848.33 ± 1.25, 1825.00 ± 0.82, 1828.33 ± 1.48, and 1875.00 ± 2.36 respectively (Table 4).

There was a significant (P<0.05) difference between the $T_4$ and control as well as the others. Moreover, there was no significant difference between the antibiotics group and the combined probiotic and prebiotic treated group. The highest live weight was found in the $T_4$ (1875.00±2.36) and the lowest result was in the $T_0$ (1806.67±2.81) group. Rehman *et al*. (2020) reported that supplementation of prebiotics or probiotics can improve the growth performance of broilers. Prebiotics are used as substrates for survival and multiplication of probiotics in a lower gut region that act as symbiotic (Hanamanta *et al*. 2011). An appropriate combination of both components in a single product should ensure a superior effect, compared to the activity of the probiotic or prebiotic alone. Therefore, the present study revealed that the addition of probiotics and prebiotics with basal diet separately or combined increases the overall live weight in the broiler.

The data presented in Table 4 also showed the effect of feeding probiotics and prebiotics on total body weight gain (gram per broiler chicken) broiler. From the table, it is clear that there are differences in total body weight gain among the treatments.

The relative total body weight gain (g) of broiler chickens in the dietary group $T_0$, $T_1$, $T_2$, $T_3$, and $T_4$ were 1764.01 ± 2.84, 1805.67 ± 1.27, 1782.34 ± 0.83, 1785.67 ± 1.50, and 1832.34 ± 2.39 respectively. The highest result was found in the $T_4$ (1832.34±2.39) and the lowest result was in the $T_0$ (1764.01±2.84) group.

Dietary supplementation of MOS improved weight gain (P<0.01) and feed conversion ratio (FCR) (Rehman *et al*. 2020). Hossain *et al*. (2021) recommended that dietary supplementation of probiotic 50g BL/Metric ton feed may have a benefit to promote growth performance of broilers. Kabir *et al*. (2004) conducted a 6-week growth performance study with broilers and found that live weight gain and carcass yields were significantly higher in broilers fed probiotic supplementation.

The results highlighted that the combination of probiotics and prebiotics improves the average body weight gain in the commercial broiler.

Data presented in Table 4 showed that the effect of different treatments on final feed consumption (gram per broiler chicken) was not significant (P>0.05). The mean of total feed consumption of broiler chicks at the end of the 4$^{th}$ week in the dietary group $T_0$, $T_1$, $T_2$, $T_3$, and $T_4$ were 2183.33 ± 1.63, 2241.57 ± 0.70, 2248.33 ± 0.49, 2186.07 ± 1.08, and 2220.80 ± 0.39 respectively. The highest average feed consumption was found in the $T_4$ (2220.80±0.39) and the lowest result was in the $T_0$ (2183.33±1.63) group. It is similar to the findings that the addition of probiotics improves feed consumption in layers and broilers (Nahashon *et al*. 1994).

Results of feed intake in this study were in line with Abdel-Raheem and Abd-Allah (2011) who observed that feed intake was improved by the supplementation of probiotics and prebiotics. The supplementation of probiotics decreased gastric emptying time, which leads to higher feed intake (Rahman *et al*. 2018; Abdel-Raheem and Abd-Allah, 2011). Therefore, this study suggests that the addition of probiotics and prebiotics in feed improves feed intake of broiler.

Table 4 showed that feed conversion ratio (FCR) was not significant (P>0.05) among the treatment groups. However, feed conversion ratio (FCR) in the dietary groups $T_0$, $T_1$, $T_2$, $T_3$, and $T_4$ were 1.21, 1.21, 1.21, and 1.19 respectively. Dietary supplementation of MOS improved weight gain (P=0.01) and feed conversion ratio (FCR) (P=0.03) during the overall period (Rehman *et al*. 2020). The combined group of probiotics and prebiotics showed better FCR than the control and antibiotic group (Table 4). Likewise, Nikpiran *et al*. (2013) and Li *et al*. (2014) found improved FCR by probiotics and prebiotics. Fallah *et al*. (2014) determined that FCR was improved by symbiotics in broiler and ostrich chicks. Improved FCR might be due to maintaining normal microbiota and better ileal digestibility by the addition of probiotics and prebiotics (Rahman *et al*. 2018). Therefore, this study revealed that the addition of probiotics and prebiotics in feed with broiler improves the FCR. In this study, the dressing percentage at $T_4$ (79.27±0.77) group was significant (P<0.05) compared with the other treatment groups $T_0$ (66.63±0.33), $T_1$ (69.28±0.79), $T_2$ (73.95±1.57), and $T_3$ (73.08±0.19) (Table 4). Moreover, the $T_2$ (probiotics) and $T_3$ (prebiotics) groups showed a better dressing percentage than the $T_0$ (control) and $T_1$ (antibiotic) groups. Alam and Ferdoushi (2018) also reported the significant dressing percentage in the probiotics added groups. Results of dressing percentage in this study were in line with other researchers who reported that the dressing percentage was increased by the addition of symbiotics (Abdel-Raheem and Abd-Allah, 2011; Saiyed *et al*. 2015). Therefore, the present study suggests that the combined addition of probiotics and prebiotics can increase the dressing percentage of the broiler. The survivability rate showed in Table 4. Survivability rate was higher for the probiotics, prebiotics and the combined probiotics and prebiotics treated group (100±0.00) than the control group (95.56±0.83) but there was no significant (P>0.05) difference amongst them. The overall survivability (0-4 weeks) during the experimental period was higher in the treatment groups. O'Dea *et al*. (2006) reported that there were no significant differences in broiler mortality between the probiotic treatment groups in any of the trials. In contrast to our findings, mortality percentage was decreased by symbiotic supplementation (Pelicano *et al*. 2005). So, the possible cause of survivability might be due to the development of immunity amongst the treatment groups than the control. Data presented in Table 5 showed that the carcass weight in the different treatment groups is better than the control group. The results revealed that the treatments had significant effects (P<0.05) in dressed breast, back, and thigh in the $T_4$ treatment than the other treatments.

However, there is no significant difference in wing and drumstick within the treatment groups. However, in the treatment $T_4$ group the carcass weight is better than the other treatment groups. Alam and Ferdousshi (2018) found the lower percent of abdominal fat and the higher percent of the dressed carcass, breast, and thigh were observed in experimental probiotic(s) groups. Probiotics supplementation has a significant effect on carcass yield, live weight gain, immune response, and prominent cut-up meat parts (Soomro *et al*. 2019).

Carcass characteristics were improved by the addition of prebiotic in broiler diet which might be related to inhibition of colonization of intestinal pathogens and improved utilization of nutrients (protein and energy) in diet (Toghyani *et al*. 2011). Probiotics can well dwell in the digestive system with help of prebiotics as with this they can well tolerate an anaerobic environment, for example, low oxygen, low pH, and temperature.

Data presented in Table 6 showed the relative weight of internal organs (liver, heart, neck, gizzard, intestine, spleen, and bursa) of broilers fed a diet containing probiotic, prebiotic, probiotic + prebiotic, control and antibiotic added group.

The result showed a significant difference (P<0.05) among the different groups. The $T_4$ (probiotic+prebiotic), $T_3$ (prebiotic), and $T_2$ (probiotic) treated group showed better results than the control group. It was also observed that there was no significant difference among the groups for the immune organ intestine, spleen, and bursa but in all cases, the $T_4$ group showed improved results. Feeding broilers with probiotics have significant effects (P<0.05) on dressed carcass weight, abdominal fat, breast, thigh, and liver while it appeared insignificant on gizzard (P>0.05) (Alam and Ferdoushi, 2018). The liver and heart weights were decreased by the supplementation of probiotics and prebiotics in the Japanese quail diet (Nikpiran *et al*. 2013). Breast, gizzard, and thigh yield were increased by symbiotics and MOS (Santin *et al*. 2001). In probiotic and prebiotic treated groups, the weight of the internal organ is higher than in the control group. Therefore, this is due to the positive effect of probiotics and prebiotics on the carcass trait of chicken.

The immune parameter mainly WBC, lymphocyte, and granulocyte was counted and the data has presented in Table 7. The WBC, Lymphocyte, and Granulocyte were statistically significant (P>0.05) among different treatments. The highest granulocyte was in control (12.38±0.11), indicating low immunity in the control group. The lowest WBC (38.50±16.58), lymphocyte (34.60±15.13), and granulocyte (2.60±1.00) were found in $T_2$.

**Table 5** Effects of probiotics and prebiotics on carcass characteristics of broiler chickens

| Treatments | Breast (g) | Thigh (g) | Wing (g) | Drumstick (g) | Back (g) |
|---|---|---|---|---|---|
| | Mean±SE | Mean±SE | Mean±SE | Mean±SE | Mean±SE |
| $T_0$ | 399.75±1.03$^{b}$ | 101.65±2.77$^{ab}$ | 137.76±2.70 | 79.51±0.64 | 216.84±5.97$^{bc}$ |
| $T_1$ | 475.87±3.11$^{abd}$ | 89.27±1.09$^{ab}$ | 89.27±0.36 | 72.01±1.59 | 200.26±2.30$^{bc}$ |
| $T_2$ | 446.40±1.96$^{bc}$ | 80.96±1.0 7$^{b}$ | 124.92±0.91 | 72.08±0.81 | 233.53±0.33$^{bc}$ |
| $T_3$ | 470.59±1.14$^{d}$ | 90.90±0.71$^{ab}$ | 125.54±1.50 | 75.46±0.79 | 220.99±1.32$^{ab}$ |
| $T_4$ | 520.17±1.32$^{a}$ | 102.98±0.73$^{a}$ | 112.28±1.63 | 82.93±0.41 | 280.70±2.77$^{a}$ |
| Level of significance | * | * | NS | NS | * |

$T_0$: control (the basal diet); $T_1$: the basal diet + antibiotic; $T_2$: probiotic; $T_3$: prebiotic and $T_4$: probiotic + prebiotic.
The means within the same column with at least one common letter, do not have significant difference (P>0.05).
* (P<0.05).
SE: standard error.
NS: non significant.

**Table 6** Effects of probiotics and prebiotics on internal organs of broiler chickens

| Treatments | Liver (g/bird) | Heart (g/bird) | Neck (g/bird) | Gizzard (g/bird) | Giblet (g/bird) | Intestine (g/bird) | Spleen (g/bird) | Bursa (g/bird) |
|---|---|---|---|---|---|---|---|---|
| | Mean±SE | Mean±SE | Mean±SE | Mean±SE | Mean±SE | Mean±SE | Mean±SE | Mean±SE |
| $T_0$ | 44.97±0.72$^{b}$ | 9.31±0.55$^{b}$ | 43.11±6.29$^{a}$ | 20.64±0.04$^{b}$ | 118.03± 1.06$^{b}$ | 119.13±0.37 | 2.06±0.27 | 2.47±0.70 |
| $T_1$ | 44.53±1.13$^{abc}$ | 9.15±0.27$^{b}$ | 40.30±6.07$^{a}$ | 36.85±1.24$^{a}$ | 130.83±0.76$^{ab}$ | 130.11±1.20 | 2.00±0.30 | 2.16±0.30 |
| $T_2$ | 45.60±0.52$^{bc}$ | 10.51±0.56$^{b}$ | 32.51±5.51$^{ab}$ | 41.17±0.36$^{ab}$ | 129.79±0.36$^{ab}$ | 118.52±1.30 | 2.10±0.48 | 2.17±0.46 |
| $T_3$ | 51.25±1.24$^{abc}$ | 11.60±0.40$^{ab}$ | 36.37±5.78$^{b}$ | 40.12±0.80$^{a}$ | 139.34±0.85$^{ab}$ | 116.99±0.52 | 2.45±0.51 | 2.74±0.70 |
| $T_4$ | 56.85±0.56$^{a}$ | 12.23±0.25$^{a}$ | 44.98±6.77$^{a}$ | 35.19±2.25$^{ab}$ | 149.25±1.30$^{a}$ | 128.14±2.24 | 3.38±0.42 | 3.05±0.44 |
| Level of significance | * | * | * | * | * | * | * | * |

$T_0$: control (the basal diet); $T_1$: the basal diet + antibiotic; $T_2$: probiotic; $T_3$: prebiotic and $T_4$: probiotic + prebiotic.
The means within the same column with at least one common letter, do not have significant difference (P>0.05).
* (P<0.05).
SE: standard error.
NS: non significant.

**Table 7** Effects of probiotics and prebiotics on immune parameters of broiler chickens

| Treatments | WBC (x$10^9$/L) | Lymphocyte (x$10^9$/L) | Granulocyte (x$10^9$/L) |
|---|---|---|---|
| | Mean±SE | Mean±SE | Mean±SE |
| $T_0$ | 15.47±0.22$^{a}$ | 2.82±0.59$^{abc}$ | 12.38±0.11$^{a}$ |
| $T_1$ | 8.07±0.45$^{b}$ | 2.56±0.39$^{a}$ | 4.64±0.49 |
| $T_2$ | 10.4±0.17$^{c}$ | 3.32±0.23$^{a}$ | 6.90±0.15 |
| $T_3$ | 11.83±0.28$^{c}$ | 2.97±0.29$^{b}$ | 8.70±0.16$^{d}$ |
| $T_4$ | 5.70±0.19$^{d}$ | 2.13±0.17$^{c}$ | 3.45±0.11$^{e}$ |
| Level of significance | * | * | * |

$T_0$: control (the basal diet); $T_1$: the basal diet + antibiotic; $T_2$: probiotic; $T_3$: prebiotic and $T_4$: probiotic + prebiotic.
The means within the same column with at least one common letter, do not have significant difference (P>0.05).
* (P<0.05).
SE: standard error.
NS: non significant.

The prebiotics are used as substrates for survival and multiplication of probiotics in a lower gut region that act as symbiotic (Hanamanta *et al*. 2011). Prebiotics are promising in controlling pathogens such as *Escherichia coli* and *Salmonella* and stimulate the growth of *Lactobacilli* and *Bifidobacteria*. Probiotics modify the intestinal ecosystem by supplying digestion enzymes, reducing pH, and increasing the activity of enzymes in the gastrointestinal tract (Kabir, 2009; Abd El-Hack *et al*. 2020). Probiotics and prebiotics were supplemented with a poultry diet to prevent diseases (Elgeddawy *et al*. 2020).

The use of probiotics and prebiotics in poultry feed can improve the immune status of broiler chickens.

The number of *E. coli* and *Salmonella* in the caecum of the birds was counted and the data has presented in Table 8. The number of *E. coli* and *Salmonella* in the caecum of birds was statistically non-significant among the treatments.

The highest *E. coli* colony was found in the control ($T_0$) group (8.80±0.29), indicating low immunity in the control group. The lowest *E. coli* colony was found in $T_1$ (8.21±0.27), $T_2$ (8.23±0.31), $T_3$ (8.3±0.28), and $T_4$ (8.3±0.55).

**Table 8** Effects of feeding probiotics and prebiotics on microflora (log 10 CFU/g) in the caecum of broiler

| Treatments | No. of *E. coli* colony (CFU/g) | No. of *Salmonella* colony (CFU/g) |
|---|---|---|
| | Mean±SE | Mean±SE |
| $T_0$ | 8.80±0.29 | 9.17±0.04 |
| $T_1$ | 8.21±0.27 | 8.77±0.22 |
| $T_2$ | 8.23±0.31 | 8.73±0.23 |
| $T_3$ | 8.3±0.28 | 8.70±0.23 |
| $T_4$ | 8.3±0.55 | 8.70±0.22 |
| Level of significance | NS | NS |

$T_0$: control (the basal diet); $T_1$: the basal diet + antibiotic; $T_2$: probiotic; $T_3$: prebiotic and $T_4$: probiotic + prebiotic.
The means within the same column with at least one common letter, do not have significant difference (P>0.05).
SE: standard error.
NS: non significant.

**Table 9** Effects of probiotics and prebiotics on economic aspects of broiler chicken farming

| Parameters | Treatments | | | | | Level of significance |
|---|---|---|---|---|---|---|
| | $T_0$ (Mean±SE) | $T_1$ (Mean±SE) | $T_2$ (Mean±SE) | $T_3$ (Mean±SE) | $T_4$ (Mean±SE) | |
| Feed cost per bird (USD) | 0.81±0.29 | 0.83±0.13 | 0.84±0.09 | 0.85±0.48 | 0.83±0.07 | NS |
| Chick cost (USD) | 0.37 | 0.37 | 0.37 | 0.37 | 0.37 | NS |
| Cost of probiotics and prebiotics | 0.00 | 0.005 | 0.006 | 0.006 | 0.012 | NS |
| Common expenditure per bird (USD) | 0.053 | 0.053 | 0.053 | 0.053 | 0.053 | NS |
| Total expenditure per bird (USD) | 1.24±0.24 | 1.26±0.10 | 1.27±0.07 | 1.28±0.39 | 1.26±0.06 | NS |
| Receipt per bird when sold (1.51 USD/kg live weight) | 2.73±1.01 | 2.79±0.45 | 2.76±0.29 | 2.76±0.53 | 2.83±0.85 | NS |
| Profit per bird (USD) | $1.49±1.18^{b}$ | $1.53±0.53^{ab}$ | $1.49±0.36^{b}$ | $1.48±0.96^{b}$ | $1.57±1.11^{a}$ | * |

$T_0$: control (the basal diet); $T_1$: the basal diet + antibiotic; $T_2$: probiotic; $T_3$: prebiotic and $T_4$: probiotic + prebiotic.
The means within the same row with at least one common letter, do not have significant difference (P>0.05).
* (P<0.05).
SE: standard error.
NS: non significant.

The highest *Salmonella* colony was found in the control ($T_0$) group (9.17±0.04). The lowest *Salmonella* colony was found in $T_1$ (8.73±0.23), $T_2$ (8.73±0.23), $T_3$ (8.70±0.23), and $T_4$ (8.70±0.22) which indicates the addition of probiotics and prebiotics with feed reduces the number of pathogenic bacteria in the intestine and improves the gut health. Gut microflora has significant effects on host nutrition, health, and growth performance of chickens (Barrow, 1992) by interacting with nutrient utilization and the development of the gut system of the host. Probiotics greatly affect the intestinal microbiota. They work against *Salmonella* to prevent birds from infection and have beneficial effects on performance (Santin *et al*. 2001). The health effect of symbiotics is probably associated with the individual combination of a probiotic and prebiotic (De Vrese and Schrezenmeir, 2008). Therefore, the present study reveals that combined usage of probiotics and prebiotics in broiler diet ameliorates the health status.

The result of the economic analysis revealed that the combined probiotic and prebiotic treated group ($T_4$) had significantly (P<0.05) better profit than the control group and the other groups except for the antibiotic group (Table 9). Total expenditure per bird was slightly high in treated groups than in control but was statistically insignificant (P>0.05).

The addition of probiotics and prebiotics not only improves the growth performance in commercial broilers but also cost-effective. Therefore, it can be suggested that the addition of probiotics and prebiotics combined with feed may be cost-effective and alternative to the antibiotic in broiler production.

## CONCLUSION

The research work was conducted to investigate the combined effect of probiotics and prebiotics on growth performance, carcass traits, immune parameters, and microbial count of commercial broilers. From this study, it could be concluded that addition of probiotic and prebiotic performed positively more or similar to antibiotic separately and significant (P<0.05) performance was observed when the probiotic and prebiotic are added combinedly than control and antibiotics on growth performance, carcass characteristics immune parameters and microbial load of broiler chickens. Therefore, combined usage of these probiotics and prebiotics and feeding as an antibiotic alternative in broiler production can be recommended to avoid the human health hazard.

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