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# Effect of Multi-strain Probiotic Feed Supplement on Growth Performance and Carcass Characteristics of Commercial Broiler Chickens

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## ABSTRACT

Probiotics are the best feed additives used in poultry to establish beneficial gut microflora by maintaining normal intestinal microflora by competitive exclusion antagonism. Three hundred and fifty day old commercial broiler chicks randomly allotted to 7 treatments with 10 replicates containing 5 chicks in each replicate and reared for 42 days. The treatments consist of corn-soya and fish meal-based control diet, control diet supplemented with antibiotic (Bacitracin methylene disalicylate, @500 g/ton) and probiotic at 100, 200, 400, 600 & 800 g/ ton diet. Fish meal (4%) was included in the diets as microbial challenge so as to assess the efficacy of the probiotic supplement. The body weight gain and feed conversion ratio were significantly ($P<0.05$) improved in birds fed on diets supplemented with probiotic at 200 g/ton and higher levels compared with the control. The overall feed consumption was significantly ($P<0.05$) lower in birds fed on control diet when compared to other treatment groups except the diets supplemented with probiotic 100 g/ton. There were no effects on carcass traits but the percent breast yield (%) was significantly ($P<0.05$) higher in birds fed diets with probiotics at 400, 600 and 800 g/ton. There was no significant ($P>0.05$) difference in dry matter (DM) and protein retention between the treatments. Hence, it can be concluded that probiotic at 400 g/ton may be supplemented as an alternative to antibiotic for improving performance of broiler chicken.

## HIGHLIGHTS

- In order to establish a beneficial gut microflora, one of the best feed additives that can be used is probiotics.
- The mode of action of probiotics in poultry includes maintaining normal intestinal microflora by competitive exclusion antagonism.

**Keywords:** Dry Matter, Feed conversion ratio, Fish meal, Protein retention, Probiotic

The antibiotic growth promoters were used in poultry feed to stabilize the intestinal microbial flora to improve the general performances and also to prevent some specific intestinal disorders. But in recent years, due to negative human health issue of antibiotic resistance, there is an increasing pressure to reduce or eliminate the use of antibiotics as growth promoters. In light of that situation, the feed manufacturers and the animal growers have been actively looking to an efficacious alternative to antibiotic growth promoters. These include organic acids, enzymes, probiotics and prebiotics, as feed additives in poultry production (Adil *et al*., 2011).

In order to establish a beneficial gut microflora, one of the best feed additives that can be used is probiotics (Fuller, 1989) which are viable micro-organisms that provide beneficial effects to their host by modifying the intestinal microbiota such as *Lactobacillus sp., Bifidobacterium sp., Bacillus sp.* etc. The mode of action of probiotics in

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poultry includes maintaining normal intestinal microflora by competitive exclusion antagonism, lowering the pH through acid fermentation, competing for mucosal attachment and nutrients, producing bacteriocins, stimulating the immune system associated with the gut and increasing the production of short-chain fatty acids (Sarangi *et al*., 2016). There are different types of microorganisms used as probiotics in poultry production. *Bacillus*-based probiotics are resistant to heat and tolerant to acidic conditions, can survive desiccation as well as challenging storage conditions, which makes them ideal for in-feed applications (Bader *et al*., 2012). *Bacillus subtilis* improve the growth performance of birds by secreting protease, amylase and lipase induce immunological response, improves intestinal integrity and absorption of nutrients (Khaksefidi and Ghoorchi, 2006) and also capable of producing beneficial intestinal bacteria, such as *Lactobacillus* species and improve weight gain and feed efficiency of broilers (Teo and Tan, 2007). *Bacillus spare* approved by the European Food Safety Authority (EFSA) for application in feed.

## MATERIALS AND METHODS

### Birds and management

Three-hundred-and-fifty (350) day old commercial broiler chicks will be randomly allotted to seven (7) treatments with ten (10) replicates containing five (5) chicks in each replicate. Five birds were housed in each battery brooder cell (2' × 2') with an average floor space of 82 square inches or 205 sq. cm per bird. Feed and water were offered *ad lib* and the birds were raised under identical management conditions. Birds were immunized against New castle disease (ND) with Lasota vaccine on 7th (primary) and 28th (booster) days of age and Infectious bursal disease (intermediate–Georgia strain) vaccine on 14th (primary) and 21st (booster) days of age.

### Experimental design

A growth trial was conducted in randomized block design, comprising of seven dietary treatments where in $T_1$ was basal diet + 4% fish meal, $T_2$ - Basal diet + 0.05% Antibiotic (Bacitracin methylene disalicylate, BMD) +4% fish meal, $T_3$ - basal diet + 100 billion cfu/ton of Probiotic + 4% fish meal, $T_4$ - basal diet + 200 billion cfu/ton of Probiotic + 4% fish meal, $T_5$ - basal diet + 400 billion cfu/ton of Probiotic + 4% fish meal, $T_6$ - basal diet + 600 billion cfu/ton of Probiotic + 4% fish meal, T 7- basal diet + 800 billion cfu/ton of Probiotic + 4% fish meal.

**Table 1:** Ingredient composition of basal diets (in kgs) fed to the commercial broilers from 0-42 d.

| Ingredient | Pre-starter (0-14d) | Starter (15-28d) | Finisher (29-42d) |
|---|---|---|---|
| Maize | 52.5 | 54.0 | 55.92 |
| Soybean meal | 37.2 | 33.69 | 30.13 |
| Fish meal | 4 | 4 | 4 |
| Vegetable oil | 3.0 | 5.30 | 6.99 |
| Di-calcium phosphate | 1.2 | 1.26 | 1.29 |
| Limestone | 0.9 | 1.05 | 0.70 |
| Salt | 0.2 | 0.10 | 0.09 |
| DL-Methionine | 0.3 | 0.21 | 0.24 |
| L-Lysine HCl | 0.2 | 0.10 | 0.22 |
| Trace Mineral Mixture | 0.10 | 0.10 | 0.10 |
| Vitamin $AB_2D_3K$ | 0.01 | 0.01 | 0.01 |
| Vitamin B-Complex | 0.01 | 0.01 | 0.01 |
| Choline chloride (60%) | 0.10 | 0.10 | 0.10 |
| Toxin binder | 0.10 | 0.10 | 0.10 |
| Total | 100 | 100 | 100 |
| **Nutrient composition** | | | |
| ME( kcal/kg) | 3000 | 3125 | 3250 |
| Crude protein (%) | 22.5 | 21.0 | 19.5 |
| Lysine (%) | 1.42 | 1.25 | 1.14 |
| Methionine (%) | 0.61 | 0.53 | 0.54 |
| Calcium (%) | 1.00 | 0.92 | 0.88 |
| Available phosphorous (%) | 0.45 | 0.42 | 0.40 |
| Sodium | 0.16 | 0.11 | 0.10 |
| Chloride | 0.18 | 0.18 | 0.18 |

*Vitamin premix provided per kg diet: Vitamin A 200000 IU, Vitamin D3 3000IU, Vitamin E 10mg, Vitamin K 2mg, Riboflavin 25mg, Vitamin B1 1mg, Vitamin B6 2mg, Vitamin B12 40mg, and Niacin 15mg; *Trace mineral provided per kg diet: Manganese 120mg, Zinc 80mg, Iron 25mg, Copper 10mg, Iodine 1mg and selenium 0.1 mg.

### Collection of data

The data of body weight was recorded on individual birds as well as on weekly basis, while the cumulative feed consumption of each replicate was recorded at weekly intervals up to 42 days of age. Mortality was recorded throughout experimental period for calculating the percent livability. The feed conversion ratio (FCR) was calculated (feed intake/body weight gain) considering mortality, as and when it occurred to maintain accuracy in the data collection by weighing back the feed on the day of mortality in that particular group.

**Carcass parameters**

Carcass parameters were studied at the end of the experiment (42$^{nd}$ day). One bird from each replicate was randomly selected, starved over night with free access to water, weighed and sacrificed by cervical dislocation on the next day. Dressing percentage, breast yield, relative weights of organs like liver, heart, gizzard, abdominal fat, giblet weight were recorded.

**Nutrient Retention (DM, CP)**

At the end (42 days) of the experiment, a metabolic trial of 3 days duration was conducted on one bird from each replicate. The birds were fasted for 14 hours (7.00 PM to 9.00 AM) to ensure the emptying of gastrointestinal tract. Next day morning by 9.00 AM they were offered with weighed quantities of respective diet. A plastic sheet (pre-weighed) was spread over the litter collection tray. On the subsequent days at 9.00 AM, the litter was collected quantitatively, dried at 80°C for 20 hours and weighed to record the faecal output on dry matter basis. Simultaneously the leftover feed was withdrawn from feeders at 9.00 AM to record the residual weight of the feed to arrive at the feed intake in each replicate group. Representative samples of feed offered and leftover feed were collected and dried at 100±5°C for 8 to 10 h to estimate the dry matter intake. The three day pooled dried samples of feed offered, residue and excreta samples were analyzed for protein.

## STATISTICAL ANALYSIS

The data was analyzed using General Linear Model procedure of Statistical Package for Social Sciences (SPSS) 15$^{th}$ version and means were compared using Duncan's multiple range test (Duncan, 1955) and significance was considered at $P<0.05$.

## RESULTS AND DISCUSSION

**Body Weight gain**

The results showed that over all body weight gain (0-42d) was significantly ($P< 0.05$) higher in birds fed on diets supplemented with graded levels of probiotic compared to the birds fed on control diet and comparable to diets containing antibiotic (Table 2). The above findings are in consistent with Khaksefidi and Groorchi (2006); Patel *et al.* (2015); Bai *et al.* (2017); Shirisha *et al.* (2017); Nguyen *et al.* (2019) who reported that supplementation of Bacillus strains significantly increased the body weight gain in broilers. The improvement in body weight gain in probiotic supplemented groups may be a result of beneficial effect of bacillus strains on intestinal microflora (Bozkurt *et al.*, 2009, Panda *et al.*, 2006) improved digestion and utilization of nutrients (Park *et al.*, 2018) and help in metabolism of minerals and synthesis of vitamins (Biotin, Vit-B1, B2, B12 and K) (Upadhayay and Vishwa, 2014).

**Feed Intake**

In pre-starter, starter phases, feed consumption was significantly ($P<0.05$) lower at control when compared to other treatment groups. The overall feed consumption was significantly ($P<0.05$) higher in birds supplemented with graded levels of probiotics when compared to control except the diets supplemented with Probiotic 100g/ton (Table 2). The above findings are in agreement with Knap *et al.* (2011), Youssef *et al.* (2017), Bai *et al.* (2017) who reported increased feed intake in birds supplemented with probiotic containing *Bacillus strains*. The improved feed intake following addition of multi strains of *bacillus* may be due to their beneficial effects on the gut flora (Steiner, 2006), improved nutrient digestibility and increased activity of intestinal enzymes (Jeong and Kim, 2014).

**Feed Conversion Ratio**

Over all FCR was significantly ($P<0.05$) better in birds fed with antibiotic 500 g/ton, probiotic at 400 g/ton & followed by probiotic (200, 600, 800, 100 g/ton) supplementation when compared to birds fed with control diet (Table 2). These findings support the previous reports of Knap *et al.* (2011), Park *et al.* (2014), Youssef *et al.* (2017), Bai *et al.* (2017) who reported that supplementation of probiotic

**Table 2:** Effect of dietary inclusion of multi strain probiotic at graded levels to diets containing fish meal on the production performance of broiler chicken during overall (0-42d) period

| Diets | Levels (g/ton) | Production performances | | |
|---|---|---|---|---|
| | | Body weight gain (g) | Feed Intake (g/bird) | FCR |
| Control | 0 | 1,748.25$^{c}$ | 3,000.35$^{c}$ | 1.74$^{a}$ |
| Antibiotic | 500 | 2,122.23$^{a}$ | 3,292.76$^{a}$ | 1.55$^{b}$ |
| Probiotic | 100 | 1,913.70$^{b}$ | 3,072.55$^{bc}$ | 1.62$^{b}$ |
| Probiotic | 200 | 1,987.14$^{ab}$ | 3,183.03$^{ab}$ | 1.60$^{b}$ |
| Probiotic | 400 | 2,121.27$^{a}$ | 3,334.97$^{a}$ | 1.57$^{b}$ |
| Probiotic | 600 | 2,026.46$^{ab}$ | 3,229.78$^{ab}$ | 1.60$^{b}$ |
| Probiotic | 800 | 2,080.19$^{ab}$ | 3,335.68$^{a}$ | 1.61$^{b}$ |
| N Value | | 10 | 10 | 10 |
| P Value | | 0.001 | 0.001 | 0.025 |
| SEM | | 23.63 | 25.028 | 0.015 |

Means bearing different superscripts within a column are significantly (P<0.05) different.

**Table 3:** Effect of dietary inclusion of multi strain probiotic at graded levels to diets containing fish meal on variables and organ weights (% live weights) at 42 days of age

| Diets | Levels (g/ton) | Dressed yield (%) | Giblet (g) | Abdominal fat(g) | % Breast yield |
|---|---|---|---|---|---|
| Control | 0 | 69.09 | 4.84 | 1.32 | 16.63$^{b}$ |
| Antibiotic | 500 | 71.61 | 4.85 | 1.04 | 18.68$^{a}$ |
| Probiotic | 100 | 70.55 | 4.56 | 1.09 | 17.27$^{b}$ |
| Probiotic | 200 | 70.63 | 4.80 | 1.29 | 17.07$^{b}$ |
| Probiotic | 400 | 71.60 | 4.82 | 1.26 | 19.59$^{a}$ |
| Probiotic | 600 | 70.85 | 4.57 | 1.20 | 19.09$^{a}$ |
| Probiotic | 800 | 70.91 | 4.73 | 1.21 | 19.02$^{a}$ |
| N | | 10 | 10 | 10 | 10 |
| P Value | | 0.327 | 0.827 | 0.384 | 0.001 |
| SEM | | 0.357 | 0.066 | 0.036 | 0.176 |

Means bearing different superscripts within a column are significantly (P<0.05) different.

containing *bacillus strains* improved the FCR. Awad *et al.* (2010) reported that probiotic increased villi height, which provided more surface area for efficient absorption of nutrients, there by leading increase nutrient absorption and improving the feed conversion in broilers.

### Livability

Livability and mortality was not influenced by the treatments. The above findings are in agreement with Upendra and Yatiraj (2002), Anjum *et al.* (2005), Reis *et al.* (2017). Patel *et al.* (2015) and Karouglu and Durdag (2005) who reported reduced mortality in probiotic supplemented group compared to control in broilers. Reduced mortality in probiotic supplemented group might be due to better immunity and high antibacterial activity thereby reducing enteric diseases and improving health of host (Collins and Gibson, 1999).

### Carcass Characteristics

There was no significant (P>0.05) difference in percent dressed yield, giblet and abdominal fat among the treatments. The breast yield significantly (P<0.05) higher in birds fed diets with probiotic when compared to control. These findings corroborate with Yadav *et al*. (2019) who reported that *Bacillus species* supplemented group had significantly higher breast yield than the control group. The increase in percent breast yield might be due to increase in nitrogen retention as *B. subtilis* positively affect the ileal CP digestibility (Reis *et al.*, 2017). Patel *et al.* (2015) and Abudabos *et al.* (2013) observed no significant difference in carcass yield due to supplementation of probiotics.

### Nutrient Retention (DM, CP)

There was no significant (P>0.05) difference in DM and protein retention between the treatments. However, percent DM and CP retention were numerically higher in birds fed with probiotic 400 g/ton compared to the birds fed on diet supplemented with antibiotic 500 g/ton and control. The above findings are in agreement with Patel *et al.* (2016) who observed no significant difference in nutrient retention (DM and CP) in broilers when supplemented with probiotic. On contrary Reis *et al.* (2017) and Nguyen *et al.* (2019) reported that the dietary supplementation of probiotic (*Bacillus subtilis*, strain DSM 17299) @ 500g/ton of feed improved DM retention in broilers.

**Table 4:** Effect of dietary inclusion of multi-strain probiotic at graded levels to diets containing fish meal on Dry matter% and Crude protein % (Nutrient retention (%)) of broiler chicken (0-6 wks of age)

| Diets | Levels (g/ton) | DM % | Crude Protein % |
|---|---|---|---|
| Control | 0 | 64.97 | 80.80 |
| Antibiotic | 500 | 68.59 | 81.19 |
| Probiotic | 100 | 68.02 | 82.40 |
| Probiotic | 200 | 68.05 | 82.44 |
| Probiotic | 400 | 69.56 | 85.91 |
| Probiotic | 600 | 68.85 | 84.43 |
| Probiotic | 800 | 68.80 | 84.40 |
| P Value | | 0.250 | 0.431 |
| SEM | | 0.680 | 0.985 |

Means bearing different superscripts within a column are significantly (P<0.05) different.

## CONCLUSION

Supplementation of probiotics at 400 g/ton improved the body weight gain and feed intake feed conversion ratio and breast yield % when compared to control and comparable to antibiotic. Performed better compared to diet supplemented with 100 and 200 g/ton.

Based on the overall results, it is concluded that probiotic at 400 g/ton may be supplemented as an alternative to antibiotic for improving performance in broiler chicken.

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