

# Influence of Probiotics supplementation on Growth and Haemato-biochemical Parameters in Growing Cattle

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**How to cite this paper:** Yasmin, F., Alam, J., Kabir, E., Maruf, A., Islam, A., & Hossain, M. (2021). **Influence of Probiotics Supplementation on Growth and Haemato-biochemical Parameters in Growing Cattle.** *International Journal of Livestock Research*, 11(6), 36-42. <https://dx.doi.org/10.5455/ijlr.20210401123825>

**Received** : Apr 01, 2021  
**Accepted** : May 03, 2021  
**Published** : Jun 30, 2021

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## Abstract

*The study was carried out to evaluate the effects of dietary probiotic supplementation on growth performance and haemato-biochemical parameters of growing cattle. A total of 12 crossbred growing male cattle were randomly divided into three equal groups. In control group (T1), cattle were fed basal diet. In treatment groups T2 and T3, cattle were fed the same basal diet but supplemented with bacterial probiotic, and combination of bacterial plus yeast probiotic, respectively. The trial lasted for 90 days. Probiotic supplementation resulted in significant ( $P<0.05$ ) improvement in body weight and average daily body weight gain over control group. In addition, dietary inclusion of probiotics resulted in significant ( $P<0.05$ ) increase in Hb, RBCs, PCV, MCV and WBCs values. Concentrations of serum glucose, triglyceride and LDL were significantly ( $P<0.05$ ) lowered in probiotic treated groups T2 and T3 than in control T1. It was concluded that probiotics supplementation could be used to improve the growth and haemato-biochemical parameters of growing cattle.*

**Keywords:** Biochemistry, Growing Cattle, Growth Performance, Haematology, Probiotic

## Introduction

Livestock enterprise is one of the fastest growing agricultural subsectors having many benefits, but this has created a major public health issues and that is sub-therapeutic use of antibiotics as growth promoters in livestock feed. Its use in livestock feed has various problems, e.g., development of microbial populations antibiotic resistance in animals and humans (Fey *et al.*, 2000; Budino *et al.*, 2005) along with antibiotic residues transfer into the food chain (Chen *et al.*, 2005). Due to their ban in many countries (Abd El-Tawab *et al.*, 2016) along with public health concern about the use of antibiotics, search for suitable alternatives has been intensified. Probiotics are one such alternative considered as an emerging, safe and viable for increasing the performance of farm animals (Alayande *et al.*, 2020).

Probiotics are viable microorganisms and when administered in sufficient amounts, can modify the gastrointestinal microbiota of the host (Rook and Burnet, 2005) in a way resulting in better health and productivity. The most commonly used probiotic microorganisms belong to bacteria (lactic acid and non-lactic acid bacteria), yeasts (dairy strains) or fungi (Tripathi *et al.*, 2008). Probiotics have multiple beneficial effects like improved rumen microbial ecology (Musa *et al.*, 2009), nutrient digestibility (Krehbiel *et al.*, 2003; Abd El-Ghani, 2004), nutrient absorption and feed conversion rate (Antunovic *et al.*, 2006; Whitley *et al.*, 2009), haematological parameters (Dar *et al.*, 2017) and decreased cholesterol (Fayed *et al.*, 2005) leading to improved performance in farm animals. However, scanty information is available concerning the effect of probiotic supplementation on growing cattle under Bangladeshi condition. In view of this, the present investigation was carried out to study the effect of supplementation of probiotic on growth performance including hematological and biochemical variables in growing cattle.

## Materials and Methods

### Animals and Management

The experiment was carried out at the Animal Farm of Sher-e-Bangla Agricultural University in Dhaka, Bangladesh, with twelve crossbred growing male cattle of around 16-18 months of age. Animals were randomly divided into three groups (4 in each) in terms of their live weight. The control group T<sub>1</sub> was fed a basal diet without any probiotic supplement, whereas the experimental treated groups (group T<sub>2</sub> and T<sub>3</sub>) were fed the same basal diet but group T<sub>2</sub> was supplemented with probiotic containing multiple bacterial cultures of 100 ml/animal/day and group T<sub>3</sub> was supplemented with probiotic containing combination of bacterial and yeast culture of 100 ml/animal/day. They were housed under similar management conditions in separate rows on the concrete-floored, well-ventilated and properly managed shed. The three groups were fed with *ad libitum* same basal diet composed of forages and commercially available concentrate ingredients. Forages (native green grasses) were supplied *ad libitum*, and the concentrate feed mixture (wheat bran, 27%; rice polish, 25%; maize crust, 15%; sesame oil cake, 20%; molasses, 10%; mineral mixture, 2%; common salt, 1%) feeding was limited to 3.0 kg per day per animal throughout the trial. They had free access to fresh clean drinking water. The trial was conducted for duration of 90 days.

### Probiotic Preparation

Probiotic bacteria and yeast (*Saccharomyces cerevisiae*) were isolated and characterized from milk and milk products, and maize, respectively. The isolation and identification of above-mentioned bacteria and yeast was done earlier in Environmental Biotechnology Laboratory of Sher-e-Bangla Agricultural University (SAU), Dhaka, Bangladesh. Bacterial probiotic were made from four strains *Lactobacillus gallinarum* JCM 2011(T), *Streptococcus infantarius* subsp. coli HDP90246 (T), *Streptococcus salivarius* subsp. thermophilus ATCC 19258(T), and *Streptococcus equinus* ATCC 9812(T). Yeast probiotic was made from *Saccharomyces cerevisiae*\_1. 90 ml distilled water, 10 ml molasses, 10 g skimmed milk and 5 g dextrose were mixed for every 100 ml mixture and autoclaved according to the procedure Harrigan (1998) and then cooled to room temperature and mixed the bacterial culture 1ml/100 ml mixture and incubated them for 24 hours. Then, spread plate technique was used to count bacterial colony and 2×10<sup>8</sup> CFU found in per 100 ml of probiotic mixture. For yeast and bacterial mixed probiotics 0.5 g live yeast was mixed per 100 ml of bacteria containing probiotics mixture and incubated for 24 hours.

### Live Weight Measurement

The body weight of the cattle was recorded with an electronic weighing balance at the start and end of the experimental feeding, and regularly at weekly intervals until the study was completed.

## Haematological and Biochemical Analyses

At the end of the feeding trial (on 90<sup>th</sup> day), the blood samples (12 ml/ cattle) from jugular vein were collected and 2ml from each were taken into EDTA containing tubes for analysis of total blood count (CBC) and haemoglobin level. Blood samples (10 ml) were collected in sterilized test tubes for total serum cholesterol measurement, serum glucose measurement and lipid profile. All the blood and serum sample were analyzed in ACI Animal Health Diagnostic Laboratory, Gulshan, Dhaka, Bangladesh.

### Statistical Analysis

Statistical analysis was done using SPSS 24.0 for Windows software.  $P < 0.05$  was considered to be statistically significant.

## Result and Discussion

### Growth Performance

There was significant ( $P < 0.05$ ) effect of dietary probiotics on final body weight and average daily body weight gain (Table 1). The final body weight and average daily gain of cattle were higher ( $P < 0.05$ ) in probiotic treated  $T_2$  and  $T_3$  groups than in  $T_1$  control group. However, the probiotic supplemented groups were statistically at par ( $P > 0.05$ ) with each other. The result of the present study was in agreement with the reports of Timmerman *et al.* (2005) on body weight gain due to supplementation of probiotics in calves. Ghazanfar *et al.* (2015) also reported improved growth rate with a probiotic containing *Saccharomyces cerevisiae* given to growing dairy heifers. A possible positive effect of probiotic on body weight gain might be due to more cellulolytic activity resulting in improved fibre degradation, increased microbial protein synthesis leading to more amino acid supply post-ruminally (Erasmus *et al.*, 1992). Further, better body weight gain in probiotic supplemented animals may also be associated with increased consumption and efficient utilization of feed (Antunović *et al.*, 2005). Krehbiel *et al.* (2003) reported increased body weight gain and feed conversion rate in probiotics supplemented beef cattle. It was also reported that dietary inclusion of *Saccharomyces cerevisiae* leads to a 9.5 and 7.8% increase in body weight gain in calves and growing adult cattle, respectively (Fiems, 1993).

**Table 1:** Effects of probiotics in growth performance of growing cattle

Parameters	T <sub>1</sub> group	T <sub>2</sub> group	T <sub>3</sub> group	Level of significance
Initial body weight (kg)	146.31±7.15	131.14±10.3	164.41±12.1	NS
Final body weight (kg)	189.88 <sup>b</sup> ±9.76	183.95 <sup>a</sup> ±9.1	218.93 <sup>a</sup> ±10.67	*
ADG, kg	0.48 <sup>b</sup> ±27	0.59 <sup>a</sup> ±28	0.60 <sup>a</sup> ±18	*

Data were presented as mean ± standard error. Means with uncommon superscripts at the same row differ significantly; \*,  $P < 0.05$ ; NS, non-significant.  $T_1$ = Control group no probiotics,  $T_2$ = probiotic fed group (bacteria),  $T_3$ = probiotic fed group (bacteria + yeast), ADG, average daily gain.

### Haematological Parameters

Compared with the  $T_1$  control group, significantly ( $P < 0.05$ ) increase of Hb, PCV, RBC count and MCV were found in probiotic supplemented  $T_2$  and  $T_3$  groups (Table 2). The results of present study were consistent with the Ghazanfer *et al.* (2015) who found that Hb, and RBC count were higher ( $P < 0.05$ ) in the probiotic administered groups than heifers in control group. Al-Saiady *et al.* (2010) and Dar *et al.* (2017) reported that the dietary inclusion of probiotics is advantageous in improving haemoglobin content and packed cell volume in crossbred calves. Morill *et al.* (1995) observed significantly different values of mean corpuscular volume (MCV) in probiotic fed calves. Probiotic supplemented lambs showed a significant ( $P < 0.05$ ) increase of Hb, PCV, and RBC count than the control (El-Sayed and Mousa, 2020). These results would be explained as the supplementation of probiotics improved absorption of iron salt from the small intestine; also, probiotics were found to produce vitamin B that facilitated the blood-cell forming processes (Kander, 2004).

**Table 2:** Effects of probiotics on haematological parameters of growing cattle

Haematological variables (units)	T <sub>1</sub> group	T <sub>2</sub> group	T <sub>3</sub> group	Level of significance
Hb (g/dL)	10.30 <sup>b</sup> ±0.15	15.40 <sup>a</sup> ±0.23	14.85 <sup>a</sup> ±0.21	*
PCV (%)	32.79 <sup>b</sup> ±2.36	47.08 <sup>a</sup> ±.50	43.93 <sup>a</sup> ±.93	*
RBC (million/mm <sup>3</sup> )	4.3700 <sup>b</sup> ±0.20	5.4833 <sup>ab</sup> ±0.18	4.9300 <sup>a</sup> ±0.04	*
MCV (fL)	88.31 <sup>b</sup> ±0.84	91.14 <sup>ab</sup> ±0.67	92.25 <sup>a</sup> ±0.75	*
WBC (per mm <sup>3</sup> )	11450 <sup>b</sup> ±229.12	12133.33 <sup>b</sup> ±440.95	13750 <sup>a</sup> ±250	*
Platelets (per/μL)	243630±1833.09	202000±14106.73	239500±9500	NS

Hb hemoglobin, PCV packed cell volume, RBC erythrocyte count, MCV mean corpuscular volume, WBC total leukocyte count; Data were presented as mean ± standard deviation. Means with uncommon superscripts at the same row differ significantly; \*,  $P < 0.05$ ; NS, non-significant. T<sub>1</sub>= Control group no probiotics, T<sub>2</sub>= probiotic fed group (bacteria), T<sub>3</sub>= probiotic fed group (bacteria + yeast).

There was a significant ( $P < 0.05$ ) increase in WBC count for supplemented cattle compared with control. These results were in accordance with the results of Al-Siaidy *et al.* (2010) who reported significantly higher white blood cell count in calves fed with bacteria probiotic than control group. The same was also reported by Ghazanfer *et al.* (2015) who found that WBC count was higher in heifers on probiotics than those fed diets without probiotics. Increase number of WBC might be involved in the production of more immune cells (Gaggia *et al.*, 2010) that play an important role in defending the biological system against different diseases. However, there were no remarkable changes in levels of platelets count in experimental groups compared with those in the control group.

## Biochemical Measurements

### Effect on Blood Glucose Level

Blood glucose level were significantly decreased ( $P < 0.05$ ) in the probiotic supplemented T<sub>2</sub> and T<sub>3</sub> groups compared with the T<sub>1</sub> control group (Table 3). Probiotics supplementation can lead to decreased blood concentrations of glucose as the result of improvement in fibre digestion, which leads to increased acetic acid and reduction of propionic acid production in the rumen (Antunović *et al.*, 2005; Bruno *et al.*, 2009).

### Effect on Blood Cholesterol Level

The blood cholesterol concentration was lower for probiotic treated T<sub>2</sub> and T<sub>3</sub> groups compared to T<sub>1</sub> control, but this difference was not statistically significant (Table 3). Lower cholesterol values might be due to the production of short-chain fatty acids upon fermentation by probiotics as described by De Preter *et al.* (2007) in humans. The results are in accordance with the results of Al-Saiady *et al.* (2010) who also reported a significant decrease in serum cholesterol levels in probiotic administered calves. The administration of probiotics is beneficiary in improving lipid profiles along with the reduction of blood cholesterol (Ooi and Liong, 2010).

**Table 3:** Effect of probiotics on blood biochemical profile of growing cattle

Biochemistry variables (units)	T <sub>1</sub> group	T <sub>2</sub> group	T <sub>3</sub> group	Level of significance
Glucose (mmol/L)	3.41 <sup>a</sup> ±0.22	2.76 <sup>ab</sup> ±0.08	2.29 <sup>b</sup> ±0.10	*
Cholesterol (mmol/L)	5.09±0.07	4.55±0.16	4.74±0.18	NS
Triglyceride (mg/dL)	68.33 <sup>a</sup> ±0.88	63.01 <sup>b</sup> ±1.67	58.15 <sup>c</sup> ±0.35	*
LDL (mg/dL)	150.76 <sup>a</sup> ±1.13	141.43 <sup>a</sup> ±4.12	124.31 <sup>b</sup> ±4.78	*
HDL (mg/dL)	39.29±0.5 5	40.50±0.76	39.64±0.14	NS

Data were presented as mean ± standard deviation. Means with uncommon superscripts at the same row differ significantly; \*,  $P < 0.05$ ; NS, non-significant. T<sub>1</sub>= Control group no probiotics, T<sub>2</sub>= probiotic fed group (bacteria), T<sub>3</sub>= probiotic fed group (bacteria + yeast).

### Effect on Triglyceride

Probiotic supplementation induced a significant reduction of plasma triglyceride levels compared with control cattle

(Table 3). Lower levels of blood triglyceride may be either due to decrease in intestinal lipid absorption or increased lipid catabolism. Such a reduction was reported by Ayad *et al.* (2013) in cows supplemented with yeast and also found in probiotic-supplemented kids or lambs (Abas *et al.*, 2007; Baiomy, 2011).

### ***Effect of Probiotic on HDL and LDL***

Probiotic has significant effect on low density lipoproteins (LDL) concentration in cattle and lowest LDL concentration  $124.31^{b\pm 4.78}$  mg/dL was found in probiotic added T<sub>3</sub> group. The addition of probiotic to the diet leads to decrease in LDL concentration in kids or lambs (Abas *et al.*, 2007; Baiomy, 2011). There was no significant effect of probiotic on the serum high density lipoproteins (HDL) concentration among all the investigated cattle. The results of the present study are in agreement with Panda *et al.* (1995) and Noori *et al.* (2016) who found that the addition of probiotics had no significant effect on serum HDL concentration.

### **Conclusion**

Supplementation of probiotic(s) containing bacteria or bacteria plus yeast to growing cattle increased body weight gain. Furthermore, the dietary inclusion of this probiotic had also improving effect on hematological and serum biochemical variables. Hence, probiotic could be used as a useful supplement to improve body growth and health status of growing cattle.

### **Acknowledgements**

This research was funded by the Ministry of Education, Bangladesh.

### **Conflict of Interests**

There is no conflict of interest.

### **Publisher Disclaimer**

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