Effects of feeding probiotic *Lactobacillus spp.* on growth performances, scours and *Escherichia coli* shedding in dairy calves

S.M. Amanullah[1]*, R. Bateen[2], K.S. Huque[1], M.R. Alam[2], M.S. Alam[1], R.N. Subarna[1], M.M. Hossain[3] and K.M.R. Islam[3]

[1]Bangladesh Livestock Research Institute, Savar, Dhaka-1341, [2]Department of Animal Science, Bangladesh Agricultural University, Mymensingh-2202, [3]Department of Livestock Services, Ministry of Fisheries and Livestock, Bangladesh

Abstract

A feeding trial was conducted on 21 crossbred (Local×Friesian, Local×Sahiwal, Sahiwal×Friesian) calves of around one week age for a period of 70 days with the objective of evaluating effects of probiotic on growth performances of calves. Calves were selected and distributed equally into three groups maintaining equal sex ratio in each group, namely the farm practice (FP), control and probiotic groups. The calves were fed on milk as 12% of their live weight up to 14 days and then 10% milk of their live weight up to 70 days of age and wheat bran was supplied to the calves from 50th day at the rate of 250g/calf. The calves of probiotic group werefedaprobiotic mixture containing lactic acid bacteria (LAB) at 0.5 g /d. The data of feed intake and growth for all groups were recorded up to 35 days and then up to 70 days for probiotic and control groups. The calves under probiotic group were achieved 82.8% and 74.5% higher gain ($P<0.05$) than that of FP and control group, respectively and feed conversion ratio was observed significantly ($P<0.05$) better in probiotic group (1.83) than that in FP (3.41) and the control group (3.50) up to 35 days of trial. However, no significant differences were found in average live weight gain and feed conversion ratio between probiotic and the control groups (1.99 vs 2.07, respectively) up to 70 days of the trial. Average counts of *E. coli* were lower in probiotic group than that in the control and FP. The incidence of diarrhoea in probiotic fed group was found 3 and 13 times lower compared to FP and the control group, respectively.

(**Key words:** Probiotic, *Lactobacillus*, growth performance, dairy calves.)

Introduction

Calf morbidity and mortality are of great concern of farmers as these represents an irrefutable and irrevocable financial loss to the livestock industry. Calf mortality up to 12 months of age has been reported as 9% under rural (Debnath *et al*., 1990) and 13.4% under farm condition (Debnath *et al*., 1995) in Bangladesh, which is mostly associated with diarrhoea and respiratory diseases. Severe calf losses (60.55%) due to digestive problems were reported in some part of the country, of which 34.8% mortality is due to diarrhoea (Samad *et al*., 2002). Moreover, those calves survived from diarrhoea failed to achieve their normal growth and productivity at their adult age. In addition, suckling calves are also suffered from reduced disease resistance and malnutrition due to inadequate colostrum intake and suckling (Samad *et al*., 2001). In Bangladesh, 63.25% of newborn calves under rural conditions are deprived from first colostrum of their mother (Samad *et al.*, 2001). Hossain *et al*. (2014) reported average 5.6% calf mortality over 12 years in Central Cattle Breeding and Dairy Farm (CCBDF) with a range of 1.05 to 11.58% and about

*Corresponding author: sm_aman03@yahoo.com

70% of total mortality was reported upto 12 months of age.

The use of antibiotics as therapeutic and/or preventive measures is a usual practice to reduce calf losses and to increase disease resistance in livestock farms. Extensive and prolonged use of antibiotics may impair the intestinal flora ecosystem by gaining resistance to the antibiotics and increase susceptibility of calves to some pathogenic organisms, and consequently, increase the risk for diarrhoea and malabsorption in intestines. More recently, growing concern over the use of antibiotics and other growth stimulants in animal feeds causes the potential risk of antibiotic residues appearing in meat and milk. The need for a food supply that is perceived as safe by consumers has prompted livestock producers to explore alternative strategies to enhance the overall health conditions and performances of their herd or flock.

Probiotics, defined as "live microorganisms which, when administered in adequate amounts, confer a health benefit on the host" (FAO/WHO, 2001), have become a major topic of research over the past decades. The work on probiotic for cattle has increased in recent years and positive effects have been found for feed intake, weight gain, milk yield and quality, early weaning, decrease of scouring and faecal coliform count and reduced demand for antibiotic treatment (Kilmer, 2005; Yoon and Stern, 1995). Nocek *et al*. (2002) have demonstrated that probiotic supplementation containing yeast and *Enterococcus faecium* could increase daily dry matter intake (DMI) and milk production during the postpartum period. Feeding probiotic consisted of *B. subtilis* was found to reduce scours in dairy calves (Higginbotham and Robinson, 2005) and have a positive effect on feed efficiency during 1 to 4 weeks of age and on immediate post-weaning gain (Jenny *et al*., 1991). Due to the raised concerns regarding *E. coli* contamination of foods and the widespread distribution of it in beef cattle, many scientists (Brashears *et al*., 2003) focused on using probiotic as a feed supplement in controlling the faecal shedding of *E. coli*.

The mechanisms of probiotic actions i.e. reduction of faecal mutagenic enzymes (Pedrosa *et al*., 1995), adherence to epithelial cells (Ocana and Nader-Macias, 2001; Reid et al., 1993), stimulation of macrophages (Kirjavainen *et al*., 1999; Tejada-Simon and Pestka, 1999), production of bacteriocins (De-Vuysta *et al*., 2004; Itoh *et al*., 1995) and reduction of enteric infections by pathogens (Younts-Dahl *et al.*, 2005; Coconnier *et al*., 2000) were suggested. Nevertheless, the suggested mechanisms are largely unclear and the data on the effect of feeding probiotic on animal growth performance and meat quality so far are minimal.

The Animal Production Research Division (APRD) of Bangladesh Livestock Research Institute (BLRI) through a series of works developed *Lactobacillus (LAB)* cultures for feeding calves. In a preliminary study, it was observed that feeding the cultures reduced the concentration of *E coli* and *Salmonella* (Amanullah *et al*., 2008) in calf faeces. The above trial was conducted with low number of calves and a shorter trial period, which were the major limitations of the preliminary study. This investigation warrant further study with sufficient number of week old calves. Therefore, this study was undertaken

with the objectives i) to determine the effect of feeding LAB on growth performances of calves and ii) to determine the efficacy of LAB feeding on calf scours.

Materials And Methods

Animals and Dietary Treatment

Twenty-one crossbred (Local×Friesian, Local× Sahiwal, Sahiwal×Friesian) calves of around one week of age were collected for feeding trial from Central Cattle Breeding Station and Dairy Farm (CCBS & DF), Savar, Dhaka. The calves were distributed into three groups namely farm practices (FP), control and probiotic groups. The selected calves were assigned to these three groups keeping the average body weight and sex ratio equal. Calves of FP group were managed under existing management system practiced in the dairy farm and calves of other two groups were managed under strict hygienic conditions. All the caves under control and probiotic group were fed sterilized milk, whereas the calves of FP group were supplied with non-sterilized milk as per existing feeding system in dairy farm. The calves under probiotic group were fed LAB probiotic mixed with milk. The feeding trial was continued for control and probiotic groups till 70 days. On the other hand, the calves of farm practice group were shifted from individual pen system to group management system after 5 weeks of the experiment as a part of established existing management system in the dairy farm and reared on different feeding system with other calves. Therefore, data of the FP group was available only for 35 days.

Housing of the Animals

The experimental calves were housed in individual calf pen made up of steel and wooden slatted floor and provided with a plastic bucket at the time of feeding milk and calf starter.

Provision of hygienic management

Stalls were washed with clean water and disinfected with phenol before arrival of calves. Rearing stalls were also cleaned and disinfected twice daily throughout the trial period. All equipment and utensils were washed with boiling water and 70% alcohol solution for sterilization every day before and after use. Hands of attendant were washed with disinfectant (70% alcohol solution) before handling of calves and utensils. Potassium per manganate solution was placed as foot bath all the time at the entrance of stalls.

Feeding of milk

All calves irrespective of treatments were supplied with milk at a rate of 12% of their live weight for the first fourteen days and thereafter at the rate of 10% of their live weight throughout the experimental period. The amount of milk to be offered was divided equally into two parts and given at 9.00 am and at 6:30 pm. The calves were not allowed to suck their dams during the trial period. Milk was collected from the bulk collection, filtered to remove extraneous material and boiled at 100^{o}C for 20 minutes in a gas burner. To keep the volume constant certain amount of water was added and samples were collected for laboratory analysis to check the nutrient composition of milk before and after sterilization (Table 1). This was done to avoid any chance of indigestion that may occur due to feeding concentrated milk. After boiling milk was cooled to 37^{o}C and was supplied to the calves of control and probiotic group. On

the other hand, the calves of FP group were supplied with milk just after warming it to 37°C as a daily practice in dairy farm.

Feeding of probiotic

The LAB probiotic mixture (Containing *Lactobacillus spp*.) developed by Bangladesh Livestock Research Institute was administered daily at a dose of 0.5 g/day to each calf belonged to the probiotic group during the evening feeding. The probiotic powder was diluted in warm milk and fed to calves.

Feeding calf starter/ wheat bran

All calves under control and probiotic group were also fed wheat bran as calf starter starting at 50 days of age. The amount of wheat bran offered and duration of feeding were 250g/head/day and 20 days, respectively. All calves irrespective of treatments were provided rock salt for licking mineral. The chemical composition of bran is given in Table 1.

quantity of feed offered was weighed daily and the representative samples of feed were collected and kept for chemical analysis. Faecal samples were also observed daily for its consistency, colour and odour and were collected weekly and stored at -20°C with 10% buffered glycerol solution as done by Agarwal *et al*. (2002).

Chemical analysis of feed

Representative samples of feed were used in duplicate to determine dry matter, organic matter, ash, crude protein and crude fiber following the method of AOAC (1990). The percentage of SNF (solid not fat), fat, protein, and lactose of milk samples were determined by using a Lactostar.

Microbiological examination

As a part of microbiological examination, colony forming unit (CFU) determination of *Lactobacillus* in probiotic mixture and *E coli* in faeces were done in the Rumen Microbiology Laboratory of Animal Production Research Division. All works for micro-biological study like media preparation and it's spreading, preparation of faeces sample, inoculation of sample to the media were conducted in the Laminar air flow cabinet to avoid any contamination. The safety cabinet was also sprayed with 70% alcohol solution before starting work. All the equipments

Table 1. Composition of diet offered to the calves of different groups

Feeds	Dry matter (% fresh feed)	Composition of feeds (% DM basis)			
		Protein	Fat	SNF	Lactose
Milk					
Non-sterilized	11.72	3.05	3.67	8.05	4.35
Sterilized	12.21	3.23	3.61	8.6	4.67
Wheat bran	86.0	17.58	-	-	-

Measurement of live weight gain and collection of feed and faecal samples

The calves were weighed at the start of the trial and there after at weekly interval by a weighing balance. Each calf was weighed in the morning before feeding and continued till to the end of 70 days feeding trial. The

were autoclaved at 121°C for 15 minutes followed by drying in an oven at 105°C before being used for the study.

The de Man, Rogosa and Sharpe (MRS) agar culture media (HIMEDIA, India, M 369-500G) was used for the determination of colony forming unit (CFU) of *Lactobacillus* in probiotic mixture. At first, 26.86g of MRS agar powder were taken in a clean 1 litre conical flask and 400ml deionized water was poured into the flask and mixed thoroughly using hot plate and magnetic stirrer. This medium was autoclaved at 121°C for 15 minutes, cooled and spread into petridishes. Probiotic mixture of 0.1 g was diluted with 0.9 ml of distilled water in an Eppendorf tube and mixed well using vortex. Ten sterilized Eppendorf tubes were taken for making ten-fold serial dilutions of probiotic mixture solution. A 0.1 ml of the each diluted solution was taken by micropipette and poured on to agar plate and was spread properly using a ladder. Plates were incubated at 37°C for 48 hours and then the colony forming units of *Lactobacillus* were counted.

One gram of faecal sample was taken into a sterilized test tube and diluted with 9.0 ml of water and mixed well using a vortex. The sample was centrifuged for 15 minutes, and 1.0 ml of supernatant was taken to dilute up to 10 folds gradually for better CFU counting.

Violet Red Bile Agar (VRB) was used for the enumeration of coliforms in faeces. In order to prepare the medium, 39.5g VRB agar was taken in a clean conical flask and deionized water was poured into the flask to make the volume of 1000ml and then mixed properly using a magnetic stirrer. This medium was heated in a hotplate at 100°C for one minute while agitated frequently. After cooling the media at 50°C it was poured into sterilized plates. Then 0.1 ml of prepared faeces sample was taken from different dilution to agar plate and was spread properly using a ladder. These plates were placed in an incubator at 37°C for 48 hours. After 48 hours, colony forming units were counted at different dilutions.

Statistical analysis

The data were tabulated in Excel spread sheet in Microsoft office and were analyzed in an Complete Randomized Design (CRD) by analysis of variance using SPSS 11.70 statistical package program. The treatment means for each parameter were compared for significance of difference using Least Significant Difference (LSD), where necessary.

Results

The data on feed intake and feed conversion ratio, growth performances, incidence of diarrhoea as well as the results of micro-biological study of probiotic mixture and faeces are presented in this section.

Colony Forming Unit (CFU) of *Lactobacillus* bacteria in probiotic mixture

Before starting the feeding trial, micro-biological study was conducted to determine the presence of *Lactobacillus* bacteria and its CFU counts in probiotic mixture. The counts of *Lactobacillus* bacteriain in the mixture were found as 4.8×10^{8} $CFUml^{-1}$.

Effects of probiotic on feed intake and nutrient intake of calves

The average feed intake of calves are presented in Table 2. Up to 5 weeks of age, the calves under probiotic group consumed 6

and 6.5% higher daily DM (g/d) than that of the farm practice and the control, respectively and these differences were non-significant (P>0.05). After 5 weeks of the experiment, data on feed intake in Farm Practice group was not considered as the calves under this group were started to offer different feeds. On the other hand, calves under probiotic group consumed 12% higher daily DM (g/d) than that of the control up to 10 weeks of age. However, no significant difference (P>0.05) was observed in daily DM intake (g/d) between two groups (Table 2). Similarly, no significant differences in the intake of SNF, fat, protein and lactose among calves belonged to different treatment groups were observed.

Effect of probiotic on live weight gain of calves

The growth performances of calves are presented in Table 3. Initial live weight of calves of all groups were similar (P>0.05). After 35 days of feeding trial, calves under farm practice group (FP) were transferred from individual pen feeding system to group feeding and were reared on a different feeding regime. Therefore, the data of FP

Table 2. Effects of probiotic on intakes of feed and nutrients by calves

Parameter	Mean			SED	Sig. level
	Farm practice	Control	Probiotic		
Up to 5 weeks of age					
Feed intake					
Daily milk intake (kg)	3.18	3.05	3.25	0.261	NS
Total DM intake (g/d)	374.3	372.9	397.1	2.93	NS
Nutrient intake (g/d)					
Protein Intake	97.0	99.0	104.0	0	NS
Fat Intake	117.0	111.0	117.0	0	NS
SNF Intake	257.0	261.0	280.0	2.39	NS
Lactose Intake	132.9	142.9	152.9	1.69	NS
Up to 10 weeks of age					
Feed intake					
Daily milk intake (kg)	-	3.3	3.6	0.371	NS
Total DM intake (g/d	-	427.1	477.1	0.046	NS
Nutrient intake (g/d)					
Protein intake	-	124.3	140.0	0.013	NS
Fat intake	-	118.0	130.0	0.013	NS
SNF intake	-	281.4	311.1	0.031	NS
Lactose intake	-	152.9	169.0	0.017	NS

DM, Dry matter; SNF, Solids not fat; SED, Standard error deviation; NS, Non-significant

group were taken only up to 5 weeks of age. Results shown that live weight gain (Table 3) were reached to a significant level (P <0.05) among the three groups (FP, control and probiotic) at 35 days. On average, the calves of probiotic group gained 82.8% and 74.5% higher live weight gain than that of the farm practice and the control group, respectively. On the contrary, no significant (P>0.05) difference in average live weight gain was observed between probiotic and the control groups at 10 weeks, though numerical difference was observed between two groups. On average, 92 g/d higher live weight gain in calves under probiotic group was observed than that of calves under the control group.

Table 3. Effects of different treatments on live weight change

Parameter	Mean			SED	Sig. level
	Farm practice	Control	Probiotic		
Initial Live wt (kg)	26.79	27.64	27.64	1.53	NS
Live weight (kg) at 5 weeks of age	31.36^{b}	32.43^{b}	36.00^{a}	3.17	*
Live wt (kg) at 10 weeks of age	-	40.6	47.0	6.73	NS
Up to 5 weeks of age					
Average Daily gain (g)	130.61^{b}	136.71^{b}	238.77^{a}	79.14	*
FCR (kg DM /kg gain)	3.41^{b}	3.50^{b}	1.83^{a}	2.34	*
Up to 10 weeks of age					
Average Daily gain (g)	-	184.71	276.57	65.70	NS
FCR(kg DM/ kg gain)	-	2.07	1.99	0.507	NS

FCR, Feed conversion ratio; SED, Standard error of deviation; NS = Non-significant, * P <0.05

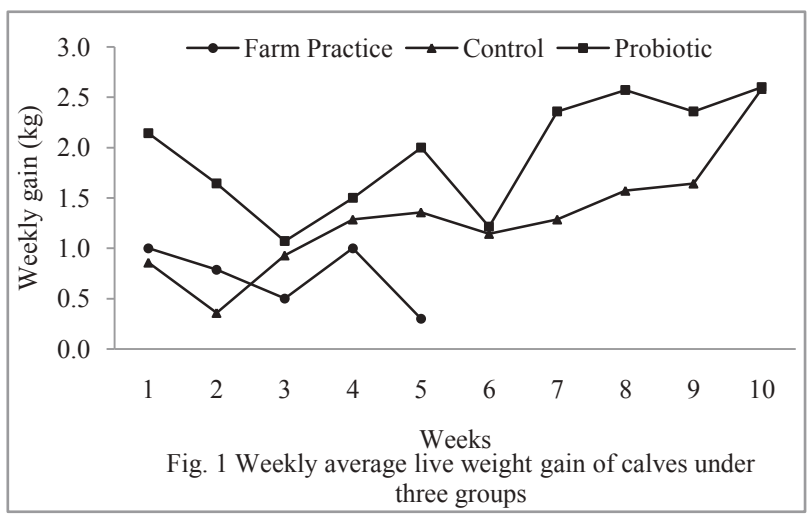


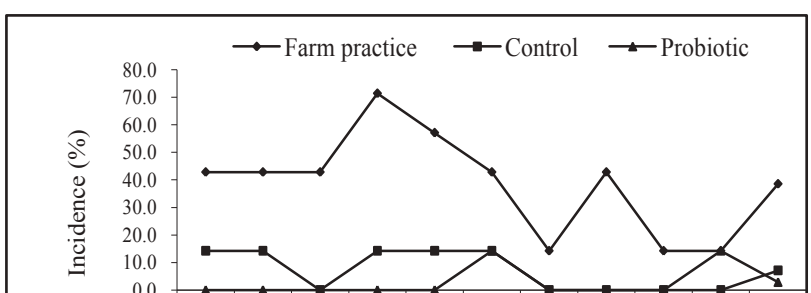
Fig. 1 Weekly average live weight gain of calves under three groups

Figure 1 shows the average weekly gain of calves under three groups. Calves under probiotic group showed higher gain throughout the whole experimental period than that of the control and FP groups. But, the calves under control group achieved almost similar live weight gain at 70 days of the experimental period.

Table 3 indicated a significantly improved FCR ($P < 0.05$) in the probiotic group than that of the other two groups (Farm practice and Control) at 5th week of the experiment. On an average, the calves under the probiotic group consumed 1.58 kg and 1.67 kg less feed than that of the farm practice and the control group, respectively to yield 1.0 kg gain up to 5 weeks of age. This has resulted a superior feed conversion ratio of the calves of probiotic group than that of the control group. However, this significant difference was level off (only 3.8%) between the control and probiotic groups at 10th week of the feeding experiment.

Effects of probiotic on incidence of diarrhoea

The consistency of calf faeces was checked daily in the morning and was graded from solid (+) to watery (++++). The incidence of diarrhoea depicted was based on the release of loose faeces as well as the load of coliform bacteria in faeces. The overall incidence (%) of diarrhoea was calculated considering both the number of animals affected with diarrhoea and frequency of diarrhoeal attack throughout the experimental period. Figure 2 illustrated the weekly and average incidences (%) of diarrhoea in calves under different groups. At 1st week of experiment no diarrhoea was reported in probiotic group and this condition was continued almost throughout the experimental period except 6th and 10th week, when the incidence (14.3%) of diarrhoea in this group was reported. Apart from this overall incidence of diarrhoea in probiotic group was reported as 2.9%. In the control group, incidence of diarrhoea was reported 14.3% from the first week to 6th week of experiment, with the exception in 3rd week, when there was no diarrhoea case reported in this group. After 6 weeks of age calves under the control group remained unaffected till the end of the study. The overall diarrhoeal incidence in this group



	1	2	3	4	5	6	7	8	9	10	Overall
Farm practice	42.9	42.9	42.9	71.4	57.1	42.9	14.3	42.9	14.3	14.3	38.6
Control	14.3	14.3	0	14.3	14.3	14.3	0	0	0	0	7.1
Probiotic	0	0	0	0	0	14.3	0	0	0	14.3	2.9

Fig. 2 Incidence (%) of diarrhoea in different groups

was 59.2% higher than that of the probiotic fed group.

In contrast, higher incidence of diarrhoea in calves of farm practice was observed and highest incidence was observed at 4th week (71.4%) and the lowest (14.3%) at 7th, 9th and 10th week. The overall incidence of diarrhoea in this group was reported 38.6% as shown in figure 2.

Effects of probiotic on fecal coliform shedding

The presence of *E. coli* in the faeces of experimental calves was confirmed from the metallic sheen growth in EMB Agar plate. Table 4. shows the percentage of *E. coli* positive samples in the faeces of calves under different groups. The presence of *E. coli* in faeces of calves as percentage of sample collected were varied at different week ranging from 71.4 to 0.0%, 57.1 to 0.0% and 33.3 to 0.0% in farm practice, control and the probiotic group, respectively (Table 4).

Table 4. Faeces samples (%) positive for *E.coli* presence

Treatment	Weekly infestation with *E. coli* (%)										Overall
	1	2	3	4	5	6	7	8	9	10	
Farm practice	0	0	57.1	28.6	28.6	28.6		-	-	-	25.0
Control	0	28.6	14.3	14.3	57.1	71.4	50	57.1	71.4	66.7	39.7
Probiotic	0	14.3	14.3	28.6	0	33.3	28.6	28.6	28.6	0	17.9

The respective faeces samples found *E. coli* positive were further cultured in VRB agar plate to determine the CFU of *E. coli* (log no. of cells ml^{-1}) and the results were illustrated in the Figures 3(a), (b) and (c). Irrespective of treatments all these figures showed an irregular and unusual pattern of coliform count, varied from 11.6 to 0.0 CFU ml^{-1} (log no. of cells) at different weeks irrespective of treatments. However, Figures showed that there was a trend for higher coliform count in faeces of calves in control group followed by farm practices and the probiotic group.

Discussion

Non-significant effect (P>0.05) of probiotic on feed intake found in this study is in agreement with the results found earlier by many researcher (Cruywagen *et al*.,1996 and Quigley *et al*., 1992). This result is in contrast with those reported by Ruppert *et al*. (1994) and by Higginbotham and Bath (1993). Probiotic supplementation in feed may affect intake of calves only when calves were kept under stressful conditions (Ruppert *et al*., 1994). Better management and feeding systems in this study might be the underlying reason for this non-significant effect (P>0.05) of probiotic on feed intake of calves.

In this study, significant (P<0.05) effect of feeding probiotic on growth performances at an early stage of calves is in agreement with results demonstrated earlier by many researchers (Sarker *et al*., 2010; Lesmeister *et al*., 2004). This significant effect (P<0.05) may be resulted from reduction of *E. coli* in intestine as evident from the lower fecal count (Figure 3 a, b, c) in probiotic group

than for the control and FP group and supports the previous report (Abe *et al.,* 1995). Their results indicated that effect of probiotic in early stages of life was greater which is similar to findings of Cruywagen *et al*. (1996) and Abe *et al*. (1995) and Quintero- Gonzalez *et al.* (1994).

The FCR in this study also confirm the earlier reports (Poonam-Pandey *et al.*, 2001, Strzetelski *et al.* 1998; Abou-Tarboush *et al*., 1996; Abe *et al*., 1995; Jenny *et al*., 1991). Similarly, Gill *et al*. (1987) reported a 9.5% improved feed conversion efficiency in probiotic fed group. Ramaswami *et al*. (2005) observed 5% reduction in feed: gain ratio in his experiment. In this study, calves in probiotic group attained 75-83% extra gain by consuming a similar amount of feed DM which appeared to promote better FCR value in the probiotic group up to 35 days (Table 3). No significant differences were observed in FCR between the control and probiotic groups at 70 days of experimental period. The reason for higher FCR during the early life of calves (5 week) may be explained by permeability of the gut and uptake of nutrients might have increased by the colonization of *lactobacillus* bacteria at an early stage of life (Roth, 2000) and resulted better utilization of feed nutrients by calves.

Feeding probiotic has also decreased incidence rate of diarrhea in this study. On average, the incidence of diarrhoea in the probiotic group was 3.4 and 13.0 times lower than that in the control and in FP group, respectively. This result is in agreement with findings of earlier researchers (Isyk *et al*., 2004 and Abe *et al*., 1995). A trend for reduced diarrhoea in this study may be explained by an antagonistic action of probiotic *Lactobacilli* towards diarrhoeagenic

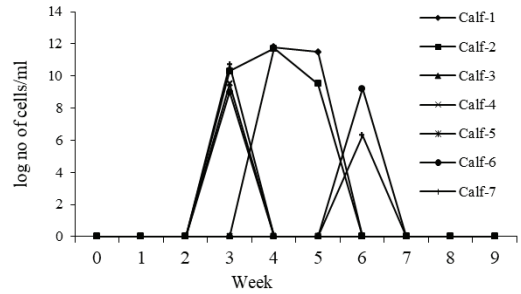


Fig. 3(a) Coliform counts in faeces of calves under farm practices

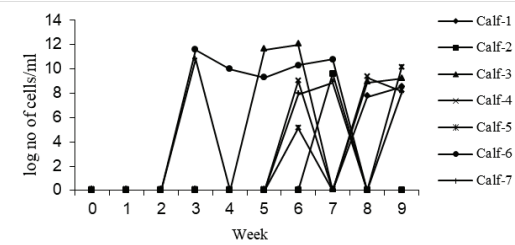


Fig. 3(b) Coliform counts in faeces of calves under control group

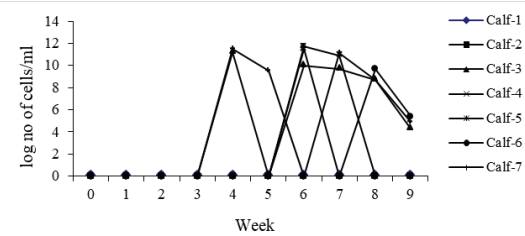


Fig. 3(c) Coliform counts in faeces of calves under probiotic group

E. coli and implantation of probiotic micro-organisms in the intestinal tract (Namioka *et al*. 1991; Yamazaki *et al*.,1991). The calves of FP group put to group management system after 5 weeks may be the underlying reason for the higher incidence rate of diarrhea in calves of this group.

Since, it was difficult to obtain intestinal samples for microbial analyses, enumeration of faecal microbial flora was used as an indirect method of determining bacterial inhabition in the intestinal tract in this study.

This method was also followed by many authors (Bruce *et al*., 1979; Gilliland *et al*.1978). It is assumed that *E. coli* represent only the luminal *E. coli* and not is associated with mucosal epithelial surfaces. The overall results of coliform count in the faeces showed no significant effects of probiotic feeding on fecal coliform shedding (Figure 3 a, b, c). Moreover, the figures portrait an unusual pattern of coliform count and, in some cases zero coliform count was observed, which is unusual because faeces is the normal habitat for *E. coli* at an optimum load. Calves were subjected to treatment with antibiotics and sulfa drugs for diarrhoea just before they were included in this experiment as well as in some cases for swelling of joint during the experimental period might have caused for producing such unusual results of coliform count. In addition, sampling procedure, transportation of sample from farm to laboratory and sample preservation technique could have contributed to this finding. For example, sample collection directly from the rectum may be the suitable procedure for such microbiological studies, whereas in this study faeces were collected from the floor after voiding.

Conclusion

It may be concluded that feeding probiotic to calves was shown to have increased growth performances up to 35 days of calves by reducing incidence of diarrhoea, which, however, not persisted up to 70 days. Feeding probiotics also improved feed conversion ratio in calves and reduce faecal shedding of *E. coli*. Further study may be done to observe the effect of probiotic feeding to the calves at different doses.

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