



## Effect of feeding live *Lactobacillus acidophilus* NCDC 15 and lactic fermented milk as probiotics on the performance of early weaned crossbred (Landrace × Desi) piglets

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### ABSTRACT

The present study was undertaken to evaluate the effect of feeding live *Lactobacillus acidophilus* NCDC 15 and lactic fermented milk (LFM) as probiotics on the performance of early weaned crossbred (Landrace × Desi) piglets. Crossbred (Landrace × Desi) piglets (36) weaned at 28 days were assigned to treatments T1, T2 and T3 (12) following completely randomized block design. Piglets were fed concentrate mixture as per standards consisting of maize, soyabean, wheat bran and fish meal as a major ingredients. Piglets of T1 were fed basal diet without probiotics, while T2 and T3 were fed basal diet fermented with *L. acidophilus* NCDC 15 (200g/day/piglet,  $5.8 \times 10^7$  cfu/g) and curd (200g/day/piglet,  $6.7 \times 10^7$  cfu/g), respectively, for 120 days. Supplementation of probiotics (*L. acidophilus* NCDC 15 and LFM) to piglet diet improved total feed intake and the net body weight gain as compared to control group. The carcass parameters like carcass weight, dressing percentage, carcass length, back fat thickness and loin eye area was higher in *L. acidophilus* NCDC 15 and LFM supplemented groups as compared to control group where no probiotic was fed. Owing to the comparable affect of both the probiotics tested, it can be concluded that to improve the performance of the piglets, LFM (curd) can be used as probiotic.

**Key words:** Carcass quality, Lactic fermented milk, Piglets, Probiotics

Successful weaning is challenging and very stressful for young piglets. To ameliorate the weaning stress, maintaining gastrointestinal tract (GIT) microflora through probiotics can be one of the strategies. Probiotics are the live microbial cultures that help in improving intestinal microbial balance and reducing the counts of harmful microbes in the piglet's intestine (Giang *et al.* 2012), thereby improving carcass characteristics of pork. Yang *et al.* (2015) reviewed role of probiotics in pig feeding and stated beneficial effects of *Lactobacillus* spp, *Bifidobacterium* spp, *Bacillus* spp feeding on gut health, immunity and growth performance at various physiological stages. Probiotic feeding was also able to ameliorate the weaning stress in piglets. According to Bureau of Indian Standards (1980), LFM (curd) is a product obtained by lactic acid fermentation of cow or buffalo milk or mixed milk through the action of single or mixed strains of lactic acid bacteria. Curd contains mixed culture of bacteria, and these bacteria inhibit the growth of pathogenic organism in the GIT. Therefore, the present study was carried out to evaluate the effects of a pure culture

of *L. acidophilus* NCDC 15 and LFM as probiotics on the performance of early weaned crossbred (Landrace × Desi) piglets.

### MATERIALS AND METHODS

**Ethical approval:** Prior approval for experiments was taken from Institutional Animal Ethics Committee as per CPCSEA (Govt. of India) norms.

**Experimental animals and grouping:** An experiment was conducted on 36 [18 Male (M) and 18 Female (F)] crossbred (Landrace × Desi) piglets weaned at day 28 at Swine production farm, Indian Veterinary Research Institute, Izatnagar by assigning them to 3 different groups (T1, T2 and T3) with 12 piglets in each group. T1 group was fed basal diet without probiotic while T2 and T3 were fed basal diet supplemented with live *L. acidophilus* NCDC 15 and LFM, respectively. All the piglets were fed as per NRC (1998) along with mineral and vitamin supplement (Table 1). Out of total daily feed, 200 g (on fresh basis) per piglet was given after fermenting either with *L. acidophilus* NCDC 15 ( $1.7 \times 10^9$  cfu/g feed) or LFM ( $2.0 \times 10^9$  cfu/g feed) as described below to T2 and T3 groups, respectively. The study was continued for 120 days.

**Preparation of fermented feed:** The lyophilized culture of *L. acidophilus* NCDC 15 was procured from NDRI, Karnal. The culture was inoculated in MRS broth (Rogosa

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*et al.* 1951) and incubated for 24 h at 37°C. Basal feed (2 kg) was mixed with 2,000 ml of tap water, inoculated with 200 ml of 24 h old *Lactobacillus* culture (10% inoculum) and incubated for 24 h at 37°C. The fermented material was used as inoculum (20% of basal feed mixture) for preparation of next day's fermented feed. After 15 days, fresh culture was taken and propagated as described above and used for next 15 days. To ferment the feed, similar procedure was followed as for *L. acidophilus* but here LFM was directly used as inoculum.

**Evaluation of probiotic product:** The counting of lactic acid bacteria was done by using pour plate method in MRS agar at pH 6.8. 90 ml normal saline (0.85%) was prepared in 250 ml flasks and distributed into test tubes (9 ml). MRS agar, test tubes having normal saline were autoclaved for 20 min. Fermented feed (1 g) was mixed with 9 ml of sterile normal saline. The content was mixed for 4–5 min in vortex mixture and the supernatant (1 ml) was used for the colony counting. The sample was diluted up to 10 folds with normal saline (0.85%). After serial dilution, 1 ml culture of appropriate dilution was poured in petri plates and 15–20 ml of MRS agar was poured and mixed gently by circular motion. The inoculated plates were incubated at 37°C for 48 h. Individual colonies were counted after 48 h of

incubation and the average of duplicate plates was taken. The average number of colonies was multiplied by the dilution factor to get the number of colony forming units (CFU) per g of sample. The fermented feed was used as probiotic and this fermented feed was evaluated periodically to see the status of the probiotic cells. The number of cells went down gradually (Table 2), hence, after 15 days a fresh culture was used to prepare fermented probiotic products.

**Carcass characteristics:** Three female and 3 male pigs per group (total, 18) were slaughtered at day 120 post-weaning at the institute as per the standard protocol. First live weights of pigs were taken and then, electric stunning (70V, 250mA) was done. The weights of the offals and other components were recorded in the order of their removal during slaughter. The following parameters were recorded:

**Carcass length:** Carcass length was measured from the front of aitchbone to the middle of the front of first rib using a metal scale.

**Back fat thickness:** Back fat thickness was measured at first rib, last rib and last lumbar using plastic measuring scale.

**Loin eye area:** The depth of the longissimus dorsi (loin eye) muscle was measured immediately posterior to the last rib. The width of loin eye was measured at a right angle to the depth measurement. The product of these 2 measurements was referred as the loin eye cross section area (mm<sup>2</sup>).

**Dressing percentage:** Dressing percent was calculated by dividing the chilled carcass weight by the live weight and multiplied by 100.

**Primal cuts:** Important primal cuts of swine, viz. jowl, picnic shoulder, loin, belly, boston butt, ham were weighed.

## RESULTS AND DISCUSSION

**Performance:** The total feed intake and net BW gain was significantly higher in the T2 and T3 groups as compared to T1 wherein no probiotic was given (Table 3). The performance of the LFM fed piglets was tended to be better than those receiving the *L. acidophilus*, hence we can say that LFM can also be used as probiotic for pigs. Giang *et al.* (2012) demonstrated improved average daily body weight gain and FCR in piglets during growing phase

Table 1. Physical composition of diets for piglets

Ingredients (%)	Body weights (kg)			
	5–10	10–20	20–50	50–80
Crushed maize	46	54	62	71
Deoiled soybean meal	30	22	15	10
Wheat bran	16	16	15	13
Fish meal	06	06	06	04
*Mineral mixture	1.5	1.5	1.5	1.5
Common salt	0.5	0.5	0.5	0.5
CP (%)	23.7	20.8	18.4	15.5
DE (kcal/kg)	3400	3399	3399	3390

\*Mineral mixture contained Ca, 25.50, P, 12.75 and S, 0.72; %; vitamin A, 700,000 and vitamin D<sub>3</sub>, 70,000 IU/kg; vitamin E, 250; nicotinamide, 1,000; Cobalt, 150; iodine, 325; iron, 1,500; magnesium, 6,000; zinc, 9,600; potassium, 100 and sodium, 5.9 mg/kg.

Table 2. Microbial profile of probiotics products at different periods

Mean of each 15 days batch	<i>Lactobacillus acidophilus</i>	<i>Lactobacillus</i> spp.
	NCDC 15 (cfu/g)	(cfu/g) (lactic fermented milk)
1 <sup>st</sup> d	3.1 × 10 <sup>6</sup>	3.3 × 10 <sup>6</sup>
4 <sup>th</sup> d	2.8 × 10 <sup>6</sup>	3.2 × 10 <sup>6</sup>
7 <sup>th</sup> d	2.2 × 10 <sup>6</sup>	2.9 × 10 <sup>6</sup>
9 <sup>th</sup> d	2.0 × 10 <sup>6</sup>	2.3 × 10 <sup>6</sup>
12 <sup>th</sup> d	1.8 × 10 <sup>8</sup>	2.2 × 10 <sup>8</sup>
15 <sup>th</sup> d	1.6 × 10 <sup>8</sup>	1.7 × 10 <sup>8</sup>
Average	1.7 × 10 <sup>6</sup>	2.0 × 10 <sup>6</sup>
pH	3.9–4.6	4–4.8

Table 3. Performance of early weaned piglets in different groups

Attributes	T1	T2	T3	SEM	P value
Total feed intake (kg)	406.3 <sup>a</sup>	440.7 <sup>b</sup>	453.0 <sup>b</sup>	7.49	0.011
Initial weight (kg)	6.40	6.45	6.74	0.20	0.756
Final weight (kg)	50.22 <sup>a</sup>	60.87 <sup>b</sup>	66.19 <sup>b</sup>	1.57	0.013
Total gain (kg)	43.82 <sup>a</sup>	54.41 <sup>b</sup>	59.43 <sup>b</sup>	1.50	<0.001

<sup>abc</sup>Means bearing different superscripts in a row differ significantly.

by feeding a LAB complex consisting of *Enterococcus faecium* 6H2, *L. acidophilus* C3, *Pediococcus pentosaceus* D7, and *L. fermentum* NC1. Yang *et al.* (2015) also documented beneficial effects of lactic acid bacteria on growth performance of piglets.

**Carcass characteristics:** Slaughter weights of pigs were comparable in different treatment groups (Table 4). Carcass weight (kg) was significantly higher ( $P<0.05$ ) in T3 as compared to T1, whereas T2 was comparable to both the groups. Higher carcass weight of the pigs was achieved by feeding a commercial probiotic (Ganeshkumar *et al.* 2005) and also by feeding *Bacillus* sp. inoculated feather meal to the pigs (Kim 2005). Increased carcass weight by probiotic feeding was also demonstrated by Ceslovas *et al.* (2005). The carcass length and dressing % were significantly ( $P<0.01$ ) higher in *L. acidophilus* NCDC 15 (T2) and LFM (T3) supplemented groups as compared to the piglets without any probiotics (T1). In the study, the higher dressing percentage observed in piglets of probiotic supplemented groups might be attributed to their increased body weight since most of the carcass characteristics are directly proportional to the body weight at the time of slaughter. Higher dressing per cent was observed in piglets fed waste food fermented by probiotic culture (Moon *et al.* 2004), whereas Anna *et al.* (2005a) did not observe any difference in dressing per cent in probiotic supplemented group and non-supplemented group. Ganeshkumar *et al.* (2005) did not find any effect of probiotics feeding on efficiency of dressed carcass and carcass length in the piglets, whereas, higher dressing percentage in probiotic supplemented piglets was observed by Ceslovas *et al.* (2005). In our study, back fat thickness was significantly ( $P<0.01$ ) higher in pigs

of probiotic supplemented groups than the control, whereas, Anna *et al.* (2005b) reported that no significant difference in back fat thickness was found between the 2 treatment groups. Loin eye area was significantly ( $P<0.01$ ) higher in LFM supplemented group (T3) followed by *L. acidophilus* supplemented group (T2) and without any probiotic group (T1). The primal cuts (kg, %) were comparable among all the treatment groups. It may be due to higher muscle and tissue growth in probiotics supplemented groups. No effect of probiotics feeding on loin eye area was reported by Ganeshkumar *et al.* (2005) and Anna *et al.* (2005a). Though there was variability in the results of different experiments conducted on probiotic feeding but a positive response in terms of carcass characteristics was observed. The variability in results might be due to difference in microbial cultures and also the mode of feeding.

In conclusion, supplementation of *L. acidophilus* NCDC 15 and LFM had improved total feed intake, body weight gain and carcass characteristics. Both the tested probiotic products were equally effective. Based on the results of our study, LFM can be propagated as an effective probiotic in swine nutrition to improve the performance of the growing piglets.

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Table 4. Effect of probiotics on carcass characteristics of pigs in different groups

Attributes	T1	T2	T3	SEM	P value
Slaughter weight (kg)	62.28	69.08	70.45	1.80	0.140
Carcass weight (kg)	44.85 <sup>a</sup>	52.21 <sup>ab</sup>	54.02 <sup>b</sup>	1.64	0.042
Dressing %	72.02 <sup>a</sup>	75.43 <sup>b</sup>	76.60 <sup>b</sup>	0.56	<0.001
Carcass length (inch)	66.20 <sup>a</sup>	77.43 <sup>b</sup>	79.37 <sup>b</sup>	1.55	<0.001
Back fat thickness (inch)	1.93 <sup>a</sup>	2.18 <sup>b</sup>	2.20 <sup>b</sup>	0.04	0.011
Loin eye area (cm <sup>2</sup> )	4.12 <sup>a</sup>	4.58 <sup>b</sup>	4.78 <sup>c</sup>	0.07	<0.001
<b>Primal cuts</b>					
Jawl (kg)	1.12	1.25	1.27	0.03	0.139
Butt (kg)	4.30	4.77	4.86	0.12	0.140
Picnic (kg)	6.86	7.61	7.76	0.20	0.140
Belly (kg)	6.72	7.46	7.61	0.19	0.140
Ham (kg)	10.74	11.91	12.15	0.31	0.140
Loin (kg)	15.41	17.09	17.43	0.45	0.140

<sup>abc</sup>Means bearing different superscripts in a row differ significantly.

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