

## AN EXERTION OF ENHANCE MILK YIELD OF NATIVE PABNA CATTLE THROUGH USING DIFFERENT TYPES OF CALCIUM SALT OF FATTY ACID

D. Yeasmin<sup>1</sup>, N. Islam<sup>2</sup>, N.R. Sarker<sup>1</sup>, N. Huda<sup>3\*</sup>, A. Habib<sup>1</sup>, F. Tabassum<sup>1</sup>

<sup>1</sup>Fodder Research and Development Project, Bangladesh Livestock Research Institute  
Savar, Dhaka, Bangladesh

<sup>2</sup>Department of Dairy Science, Bangladesh Agricultural University, Mymensingh, Bangladesh

<sup>3</sup>Animal Production Research Division, Bangladesh Livestock Research Institute  
Savar, Dhaka, Bangladesh

### ABSTRACT

Keeping consideration on increasing quality milk production through addition of conventional fat, an experiment accords CRD design was conducted at Bangladesh Livestock Research Institute, Savar, Dhaka for a period of 30 days including 5 days of digestibility trial. Twenty lactating Pabna cattle of average  $250.1 \pm 13.64$  kg (*Bos indicus*) of initial body weight with average milk yield of  $2.88 \pm 0.38$  kg were selected and divided into four groups randomly having five cows in each. The imposed treatments were T<sub>0</sub>, T<sub>1</sub>, T<sub>2</sub>, and T<sub>3</sub>; in where T<sub>0</sub>= German grass + Concentrate mixture without Ca salt of fatty acid + 2.5 % DCP, T<sub>1</sub>= German grass + Concentrate mixture + 2.5 % soybean based Ca salt of fatty acid, T<sub>2</sub>= German grass + Concentrate mixture + 2.5 % mustard based Ca salt of fatty acid and T<sub>3</sub>= German grass + Concentrate mixture + 2.5% palm-oil based Ca salt of fatty acid. The result showed that, calcium salt of fatty acid influenced to increase of milk yield significantly among ( $P < 0.001$ ) the treatments; in where highest yield gained in T<sub>1</sub> ( $3.53 \text{ kg d}^{-1}$ ) followed by T<sub>2</sub>, T<sub>0</sub> and T<sub>3</sub> (3.33, 2.44 and 2.24, respectively). Besides this, all other parameters analytically examined in this experiment i.e. intake, apparent digestibility, gained body weight and chemical composition of milk showed non-significant difference among the treatments. From the analytical value and discussion, it may be suggested that, calcium salt of fatty acid formulated from soybean oil or mustard oil could be used for enhancing better quality milk yield.

**Keywords:** Ca salt of fatty acid, milk yield, chemical composition of milk, nutrient intake, nutrient digestibility, gained weight

### INTRODUCTION

For tactful diet formulation with much more energy than conventional fat consults addition of fat as feed in the diet very commonly. The supplemental fat of diet,

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\* Corresponding author e-mail: [hudanazmul1971@gmail.com](mailto:hudanazmul1971@gmail.com)

provide calories and as well as impacts on tissue metabolism by altering genetic expression (Sumida et al., 1993) or on supplying precursors (acetyl Co-A) for synthesis of other fatty acids or steroids (Staples et al., 1998), or competing with other cell components (Thatcher et al., 2004). In addition, metabolizable energy in the form of supplemental fat is utilized more efficiently than that contained in cereals and forage (Palmquist and Jenkins, 1980). And, another beautiful mechanism is, rumen inert fat can enhance energy density of lactation diets without adversely affecting fiber digestion; thus, effectively overcome the shortcomings of conventional fat being introduced in the rumen (Jenkins and Palmquist 1984; Grummer, 1988). The development of Ca salts of fatty acids (CaFA), which are considered inert in the rumen, offers a method of increasing production and efficiency without impairing fermentative digestion (Chalupa et al., 1986). In this regard, several experiments had done and they suggested that, inclusion of Ca salts of FA increased overall milk production of dairy cows (Moallem et al., 2000); by means of milk yield or milk fat percentage (Klusmeyer et al., 1991) or both (Rabiee et al., 2012). A major concern is, "Ca salts" are thought to be insoluble in the rumen which resembles an experiment; in where a higher proportion of unsaturated FA in duodenal digesta when a diet containing Ca salts of FA from palm oil was fed (Wu et al., 1991). But, it is more established that, calcium salt of fatty acid have possible protection against bio-hydrogenation depending on the conditions in rumen and if feeding saturated fats or Ca salts of long chain fatty acids are practiced, then it may minimize any detrimental effects on ruminal fermentation and saturated fatty acids are less likely to alter fermentation in the rumen than unsaturated fatty acids, because saturated fatty acids are less soluble and therefore, are less likely to absorb by bacteria (Chalupa et al., 1984). Saturated fatty acids also react more readily with metal ions to form insoluble salts of fatty acids (Palmquist and Jenkins, 1982) and preformed Ca salts of fatty acids (CaS) does not alter fermentation in the rumen because of their insolubility (Chalupa et al., 1984 and Chalupa et. al., 1986). It is exercised and proved that, supplementation of calcium salts of long chain fatty acids (Ca-LCFA) as a rumen inert fat (PF) has no detrimental effect on fermentation and apparent nutrient digestion (Naik et al., 2009). Even ruminal pH, total VFA, molar percentage acetate propionate, and milk yield and fat percentage were not affected by fat supplementation (Grummer, 1988). Moreover, calcium salts of palm oil fatty acids at a 4% level in the concentrate mixture resulted with improved milk production and milk quality in terms of polyunsaturated fatty acids without affecting the digestibility of nutrients (Sajith et al., 2008). For a long time, supplemental fat is increasingly included in the diets of high yielding dairy cows (Kellogg et al., 2001). This allows to modify the fatty acids (FA) pattern of the milk fat (Precht et al., 2001) and to improve the energy supply of the cow. Furthermore, supplemental fat act as nutritional modifier of physiology and metabolism (Voigt et al., 2005). However, unprotected, unsaturated FA can be toxic to the rumen microbes unless saturated by microbial hydrogenation (Harfoot, 1981). There are different commercial sources of

rumen inert fats including hydrogenated fatty acids and calcium salts of fatty acids and these fat sources were originally designed to increase the calorie intake of dairy cows with minimal impact on rumen microbial activity indeed. In aspect of improved milk production and altering composition of milk fat, there has no doubt on influencing supremacy of Calcium salt of fatty acid. But, regarding this context, a few works have done in Bangladesh. So very logically, this experiment was undertaken to find out the effect of feeding different types of calcium salt of fatty acids on feed intake, digestibility, milk yield, milk composition of dairy cow and finally to recommend a perfect one.

### MATERIALS AND METHODS

An experiment accords CRD design was conducted at “Pachutia Cattle Research Farm” of Bangladesh Livestock Research Institute (BLRI), Savar, Dhaka for a period of 30 days including 5 days of digestibility trial from 10 September, 2013 to 10 October, 2013. Twenty lactating Pabna cattle of average  $250.1 \pm 13.64$  kg (*Bos indicus*) of initial body weight with average milk yield of  $2.88 \pm 0.38$  kg were selected and divided into four groups randomly having five cows in each with maintained similarity of body weight and milk yield of each group as much as possible. These groups were then imposed to treatments of  $T_0$ ,  $T_1$ ,  $T_2$ , and  $T_3$ ; in where  $T_0$ = German grass + Concentrate mixture without Ca salt of fatty acid + 2.5 % DCP,  $T_1$ = German grass + Concentrate mixture + 2.5 % soybean based Ca salt of fatty acid,  $T_2$ = German grass + Concentrate mixture + 2.5 % mustard based Ca salt of fatty acid and  $T_3$ = German grass + Concentrate mixture + 2.5% palm oil based Ca salt of fatty acid. Due to the abundance of German grass at research farm it was used as basal diet of cow. Three different types of calcium salt of fatty acid were prepared using sodium hydroxide (NaOH) solution and saturated solution of calcium chloride ( $\text{CaCl}_2$ ). As for soybean oil, at first five parts of soybean oil were added to four parts of aqueous solution of NaOH (6M) and the hydrolysis of oil triacylglycerols was performed at 95 to 100°C with continuous agitation and bubbling Na. When no more soybean oil was visible, the resulting blend was left to stand at 5°C until Na soaps had solidified. The Na soaps then were dissolved in hot water (95 to 100°C) using a 1:5 ratio of soap to water, and a saturated solution of  $\text{CaCl}_2$  at a ratio of 2.5 parts and 4.5 parts of soap to water was added for salting out. A filter cloth was used to filter the Ca salts, and tap water was used to remove residual NaOH and excess  $\text{CaCl}_2$ . The Ca salts were finally dried both in air and sun, and kept at about 20°C until use for feeding. For mustard oil and palm oil same manner was followed. Preparation of Ca salt of fatty acid was done on the basis of availability of ingredient by modifying the method of Chouinard et al. (1998). The concentrate mixture was prepared by weighing and mixing of wheat bran (45%), maize crushed (10%), wheat crushed (10%), soybean meal (5%), til oil cake (10%), kheshari (15%), fish meal (2%), salt (0.5%), DCP or Calcium salt of fatty acids (2.5%) manually. The nutrient composition of concentrate mixture was dry matter (90.21%), organic matter

(91.49%), crude protein (16.07%), ADF (21.29%), NDF (55.69%) and ash (8.51%). All the ingredients were mixed properly and then allowed to animals. The amount of supplied and refused feeds was recorded everyday and from there actual feed intake of each animal was found out periodically after every fortnight and then incremental feed was adjusted to the animal diets. The nutrient requirements of animal (varying amount of milk production) were calculated based on the recommendation of NRC, 1984. Live weight of all animals were also measured fortnightly at morning with empty stomach. Animals were housed in individual well equipped stanchion barn and offered concentrate rations as two equal portions at 6:00 and 13:00 h before milking and roughages was offered thrice a day; as one third at morning after milking and remaining two third at noon after milking again. Abundant clean water was made available all the period of experiment and prior to experiment, all the cows were dewormed for internal parasites using Tetranid Bolus (Techno Drugs, Bangladesh). A conventional digestion trial was performed for 5 days at the end of feeding trial and that time feed intake, refusal of feed and was recorded daily. Composite samples of supplied feed, residue and feces of individual animal were stored at  $-20^{\circ}\text{C}$ . At the time of chemical analysis, 1 mm screen sieve was used for mixed dried feces except DM and CP. Samples of fresh fodder, concentrates, feed refusals and feces were analyzed for dry matter (DM), organic matter (OM), crude protein (CP), neutral detergent fiber (NDF) and acid detergent fiber (ADF). DM contents by oven drying at  $105^{\circ}\text{C}$  for overnight, ash at  $550^{\circ}\text{C}$  for 8 h, total nitrogen (N) by Kjeldahl procedure and CP calculation from N content ( $\text{CP} = \text{N} \times 6.25$ ) according to the official methods of AOAC (2005) were determined. NDF and ADF were determined following procedure proposed by Goering and Van Soest (1970). Apparent digestibility coefficient for DM, OM, CP, NDF and ADF was calculated from dietary intake of constituent and amount recovered in feces. Milk samples were collected from each cow at 15 days interval and were analyzed for fat, protein, lactose, SNF and total ash contents by milk analyzer (Lactostar, Funk Gurbar). The data were analyzed using "MSTAT-C" statistical program to compute analysis of variance (ANOVA) for a Complete Randomized Design (CRD) and the mean values with standard error of difference (SED) were recorded. The difference among the treatment means were determined by Duncan's Multiple Range Test (Steel and Torrie, 1980).

## RESULTS

In this experiment, all the intake parameters of nutrients showed that, they are non-significant among the treatments. In case of total dry matter intake, little bit highest value observed in  $T_1$  (2.5 % soybean oil based calcium salt of fatty acid) treatment with  $7.99 \text{ kg d}^{-1}$  followed by  $T_3$  (2.5% palm oil based calcium salt of fatty acid),  $T_2$  (2.5 % mustard oil based calcium salt of fatty acid) and  $T_0$  (without calcium salt of fatty acid) (7.93, 7.92 and 7.84), respectively. Result of dry matter intake from roughage varied as same before but in case of DMI of concentrate it was different as before (Table1). The metabolizable energy requirement of different treatments were

50.4, 49.4, 47.6 and 50.0 ( $T_0$ ,  $T_1$ ,  $T_2$  and  $T_3$ ), respectively (Table 1). And, after the experiment, increased ME intake was observed with all the treatments. Among the treatments, highest MEI value was observed in  $T_1$  with 55.4 MJ  $d^{-1}$  followed by  $T_0$ ,  $T_3$  and  $T_2$  (54.4, 53.6 and 50.8, respectively), shown in table 1. Same manner was seen in the context of ME intake from roughage i.e. highest in  $T_1$  with 37.0 MJ  $d^{-1}$  followed by  $T_0$ ,  $T_3$  and  $T_2$  (36.2, 34.8 and 32.6, respectively), shown in table 1. But, from concentrate  $T_3$  treatment harvest the highest ME i.e. 18.8 MJ  $kg^{-1}$ . In case of crude protein intake, highest total crude protein intake was observed in  $T_3$  treatment with 995.2 gm  $d^{-1}$  followed by  $T_0$  &  $T_1$  and  $T_2$  (994.4 and 992.4, respectively), shown in Table 1. CP intake from roughage was observed as same manner as intake of total CP. From concentrate, highest crude protein was ingested by  $T_0$  (385.4 gm  $d^{-1}$ ) followed by  $T_1$ ,  $T_3$  and  $T_2$  (384.6, 384.4 and 383.6, respectively), shown in table 1. The effect of calcium salt of fatty acid on apparent digestibility of different treatments for all the parameters were also found non-significant. Highest dry matter and crude protein digestibility was found in  $T_0$  (71.68 and 72.04, respectively) followed by  $T_3$ ,  $T_2$  and  $T_1$  (71.24 & 71.39, 71.05 & 71.29 and 69.31 & 69.23, respectively), shown in table 1. The effect of different types of calcium salt of fatty acid was clearly visible with the milk yield of different treatments. The average daily milk yield of whole experimental period were 2.44, 3.53, 3.33 and 2.24 kg in treatment groups  $T_0$  (without calcium salt of fatty acid),  $T_1$  (2.5 % soybean oil based calcium salt of fatty acid),  $T_2$  (2.5 % mustard oil based calcium salt of fatty acid) and  $T_3$  (2.5% palm oil based calcium salt of fatty acid), respectively; which differed significantly ( $P < 0.001$ ) among treatments with gained highest yield in  $T_1$  (3.53 kg  $d^{-1}$ ) followed by  $T_2$ ,  $T_0$  and  $T_3$  (3.33, 2.44 and 2.24, respectively), shown in table 2. Here,  $T_1$  did not differed significantly with  $T_2$  and  $T_0$  also did not differed significantly with  $T_3$  but  $T_1$  &  $T_2$  together differed significantly ( $P < 0.001$ ) with other two treatment groups i.e.  $T_0$  &  $T_3$ . This result indicates significant improvement in milk yields distinctly due to supplementation of Calcium salt of fatty acid. However, the compositions of milk against fed different diets represented the result differently. The average milk fat content of  $T_0$ ,  $T_1$ ,  $T_2$  and  $T_3$  groups were 4.01, 3.35, 3.55 and 3.66 g  $kg^{-1}$  of milk, respectively. There was no significant difference ( $p > 0.05$ ) in terms of fat, protein, lactose, total minerals (estimated) or solids not fat (g  $kg^{-1}$ ) among treatments (Table 2). Finally, very logically as all animals of each treatment got the same quality feed ingredient except Ca salts of fatty acid then any significant difference of live weight gain among treatments was not occurred (Table 3).

Table 1. Effect of different types of calcium salt of fatty acid on intake and digestibility of different treatment groups of Pabna lactating cow

Parameter	Mean $\pm$ SE of different treatment group				Level of significance
	T <sub>0</sub>	T <sub>1</sub>	T <sub>2</sub>	T <sub>3</sub>	
ME (MJ) requirement	50.40 $\pm$ 2.03	49.40 $\pm$ 3.05	47.60 $\pm$ 3.74	50.00 $\pm$ 3.78	NS
DMI from roughage (kg)	5.45 $\pm$ .068	5.62 $\pm$ .094	5.53 $\pm$ .14	5.58 $\pm$ .079	NS
DMI from concentrate (kg)	2.37 $\pm$ .046	2.37 $\pm$ .085	2.38 $\pm$ .076	2.35 $\pm$ .022	NS
Total DMI (kg)	7.84 $\pm$ .06	7.99 $\pm$ .12	7.92 $\pm$ .21	7.93 $\pm$ .09	NS
ME from roughage (MJ)	36.20 $\pm$ 1.83	37.00 $\pm$ 2.47	32.60 $\pm$ 2.06	34.80 $\pm$ 2.48	NS
ME from conc. (MJ)	18.20 $\pm$ .58	18.40 $\pm$ .51	18.20 $\pm$ .80	18.80 $\pm$ .66	NS
Total ME intake (MJ)	54.40 $\pm$ 1.75	55.40 $\pm$ 2.25	50.80 $\pm$ 2.39	53.60 $\pm$ 3.08	NS
CP from roughage (g)	609.00 $\pm$ 2.19	609.80 $\pm$ 3.01	608.80 $\pm$ 2.73	610.80 $\pm$ 1.36	NS
CP from conc. (g)	385.40 $\pm$ 1.21	384.60 $\pm$ 3.19	383.60 $\pm$ .93	384.40 $\pm$ .68	NS
Total CP intake (g)	994.40 $\pm$ 1.50	994.40 $\pm$ 1.78	992.40 $\pm$ 2.23	995.20 $\pm$ 1.32	NS
Digestibility of DM	71.68 $\pm$ 2.08	69.31 $\pm$ 1.65	71.05 $\pm$ 1.81	71.24 $\pm$ 1.17	NS
Digestibility of OM	69.05 $\pm$ 1.18	66.74 $\pm$ 1.57	69.05 $\pm$ 1.18	69.05 $\pm$ 1.18	NS
Digestibility of CP	72.04 $\pm$ 2.21	69.23 $\pm$ 1.44	71.29 $\pm$ 1.14	71.39 $\pm$ 1.07	NS
Digestibility of ADF	68.26 $\pm$ 1.21	65.89 $\pm$ 1.61	68.26 $\pm$ 1.21	68.26 $\pm$ 1.21	NS
Digestibility of NDF	71.60 $\pm$ .60	72.70 $\pm$ .51	71.40 $\pm$ .76	71.90 $\pm$ .58	NS

Significant level= NS: Non Significant= $P>0.05$ ; SE: Standard error; T<sub>0</sub>= without calcium salt of fatty acid; T<sub>1</sub>= 2.5 % soybean oil based calcium salt of fatty acid; T<sub>2</sub>= 2.5 % mustard oil based calcium salt of fatty acid; T<sub>3</sub>= 2.5% palm oil based calcium salt of fatty acid

Table 2. Effect of different types of calcium salt of fatty acid on milk yield and composition of different treatment group of Pabna lactating cows

Parameter	Mean $\pm$ SE of different treatment group				Level of significance
	T <sub>0</sub>	T <sub>1</sub>	T <sub>2</sub>	T <sub>3</sub>	
Initial milk yield	2.86 $\pm$ .21	2.9 $\pm$ .22	2.84 $\pm$ .48	2.94 $\pm$ .29	NS
Final milk yield	2.44 <sup>b</sup> $\pm$ .15	3.53 <sup>a</sup> $\pm$ .13	3.33 <sup>a</sup> $\pm$ .21	2.24 <sup>b</sup> $\pm$ .28	***
Fat (g kg <sup>-1</sup> )	4.01 $\pm$ .21	3.35 $\pm$ .19	3.55 $\pm$ .19	3.73 $\pm$ .26	NS
Protein (g kg <sup>-1</sup> )	3.65 $\pm$ .02	3.61 $\pm$ .05	3.70 $\pm$ .05	3.71 $\pm$ .04	NS
Lactose (g kg <sup>-1</sup> )	4.95 $\pm$ .30	5.21 $\pm$ .07	5.33 $\pm$ .07	5.34 $\pm$ .06	NS
Solids not fat (g kg <sup>-1</sup> )	9.67 $\pm$ .06	9.60 $\pm$ .14	9.83 $\pm$ .13	9.84 $\pm$ .11	NS
Estimated mineral (g kg <sup>-1</sup> )	0.65 $\pm$ .02	0.62 $\pm$ .06	0.67 $\pm$ .02	0.72 $\pm$ .03	NS

Significant level= (Non Significant= $P>0.05$ ; \*\*\*= $P<0.001$ , highly significant), <sup>abcd</sup> values with different

superscripts in the same row differ significantly; SE: Standard error, T<sub>0</sub>= without calcium salt of fatty acid; T<sub>1</sub>=2.5 % soybean oil based calcium salt of fatty acid; T<sub>2</sub>= 2.5 % mustard oil based calcium salt of fatty acid; T<sub>3</sub>= 2.5% palm oil based calcium salt of fatty acid

Table 3. Effect of different types of calcium salt of fatty acid on live weight changes of different treatment groups of Pabna lactating cow

Parameter	Mean ( $\pm$ SE) of different treatment group				Level of significance
	T <sub>0</sub>	T <sub>1</sub>	T <sub>2</sub>	T <sub>3</sub>	
Initial Live weight (kg)	258.2 $\pm$ 9.26	265 $\pm$ 13	224 $\pm$ 19.21	253.2 $\pm$ 13.09	NS
Final live weight (kg)	279.2 $\pm$ 3.48	285 $\pm$ 7.73	244.2 $\pm$ 17.16	269.8 $\pm$ 10.01	NS
Live weight gain (kg)	21 $\pm$ 10.47	20 $\pm$ 8.05	20.2 $\pm$ 5.77	16.6 $\pm$ 5.90	NS
Daily live weight gain (kg d <sup>-1</sup> )	0.70 $\pm$ 0.34	0.66 $\pm$ 0.26	0.67 $\pm$ 0.19	0.55 $\pm$ 0.19	NS

Significant level= NS: Non Significant= $P > 0.05$ ; SE: Standard error, T<sub>0</sub>= without calcium salt of fatty acid; T<sub>1</sub>=2.5 % soybean oil based calcium salt of fatty acid; T<sub>2</sub>= 2.5 % mustard oil based calcium salt of fatty acid; T<sub>3</sub>= 2.5% palm oil based calcium salt of fatty acid

## DISCUSSION

The intake of total DM was higher in T<sub>2</sub> group (2.5 % calcium salt of fatty acid) may be due to efficient microbial growth which may not be the exact cause. Wanapat and Khampa (2006) described that, the combined use of feeding with roughage and concentrate after 4h with MSPF could improve rumen P<sup>H</sup>, microbial protein synthesis but reduced protozoal population in dairy steers. Hristov et al. (2004) viewed long chain unsaturated fatty acid (C<sub>18:3</sub>, C<sub>18:2</sub>, C<sub>18:1</sub>) decreased protozoal numbers. But discrete or overall performances of roughage and concentrate did not differ significantly which resembles to the findings of Klusmeyer et al., 1991. Feeding vegetable oils rich in linoleic acid (Go mez-Corte et al., 2008) also respond similarly was found. In case of apparent digestibility, Naik et al. (2009) evaluated that, there had no effect on apparent digestibility of DM and organic matter (OM) when calcium salts of long chain fatty acids (Ca-LCFA) as a rumen inert fat (PF) was used as adult buffalo feed. CP digestibility was not significantly differed among treatments as like as the findings of Voigt et al. (2006), in where carbohydrates and crude protein did not differ significantly. And, Schauff and Clark (1989) suggest that Ca salts of fatty acids were inert in the rumen and did not greatly alter fermentation in the rumen, apparent total tract digestibility of DM, organic matter, ADF, NDF, and CP. Significant improvement of milk yield was observed among treatments through incorporation of different types of Ca salts of fatty acid at 2.5% level in the concentrate mixture in this experiment. In the study of Sajith et al. (2008) got improved quality milk production without affecting the digestibility of nutrients through incorporation of polyunsaturated calcium salts of fatty acids at a 4% level in the concentrate mixture. By using calcium salts of fatty acids Rabiee et al. (2012) and Fahey et al. (2002) also obtained the similar result. There was no significant ( $p > 0.05$ )

difference for the parameters of milk protein, lactose, total minerals and SNF. Similar result was also reported by Perfield et al. (2002) except milk fat who conducted an experiment by feeding rumen protected CLA to pregnant cows.

### CONCLUSION

Based on the above finding it may be concluded that, calcium salt of fatty acid formulated from soybean oil or mustard oil could be used for enhancing better quality milk yield and it may recognized as an effective technique.

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