

## Effect of feeding ca-salts of fatty acids on fatty acids composition in milk

N.R. Sarker<sup>1\*</sup>, K.S. Huque<sup>1</sup>, H. Islam<sup>2</sup> and N.G. Das<sup>1</sup>

<sup>1</sup>Bangladesh Livestock Research Institute, Bangladesh, <sup>2</sup>M.Sc. Student, Bangladesh Agricultural University (BAU), Mymensingh

\*Corresponding author: sarkernr62@yahoo.com

### Abstract

A study was undertaken to investigate the effects of Calcium Salt of Fatty Acid on the fatty acids composition of milk fed green German grass and concentrate diet. Twenty lactating Pabna cows having similar daily milk yield ( $3.4 \pm 0.842$  Kg) were used and divided into four groups with five animals in each group. The study revealed that saturated fatty acids of milk fat were 61.12, 57.83, 54.93, and 54.56% and unsaturated fatty acids of milk fat were 38.36, 42.22, 45.22 and 45.53% in treatment groups T<sub>0</sub> (without Calcium Salt of Fatty Acid), T<sub>1</sub> (2.0 % Calcium Salt of Fatty Acid), T<sub>2</sub> (2.5 % Calcium Salt of Fatty Acid) and T<sub>3</sub> (3.0 % Calcium Salt of Fatty Acid), respectively. The highest unsaturated fatty acid was found in T<sub>3</sub> treatment and the lowest in T<sub>0</sub> treatment. There is no significant difference between T<sub>2</sub> and T<sub>3</sub> treatments in relation to unsaturated fatty acids. The palmitic acid content in milk fat were 25.47, 27.22, 26.22 and 26.22% in T<sub>0</sub>, T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub> respectively. The highest palmitic acid found in T<sub>2</sub> treatment and the lowest in T<sub>0</sub> treatment. The stearic acid content in milk fat were 22.98, 15.78, 13.74 and 13.68% in T<sub>0</sub>, T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub> respectively. The study showed that there is a significant ( $p < 0.01$ ) difference in the treatments for stearic acid content in milk fat. The linoleic acid content in milk fat were 1.57, 1.64, 1.73 and 1.94 % in treatment groups T<sub>0</sub>, T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub> respectively. It may be concluded that the feeding Ca-Salts of Fatty Acids increased unsaturation of milk fat, oleic acid, linoleic acid and linolenic acid and decrease saturation and stearic acid content in milk fat.

**(Key Words:** Calcium salt of fatty acids, unsaturation, milk fat, oleic, linoleic, and linolenic acids.)

### Introduction

Feeding saturated fats or Ca salts of long chain fatty acids may minimize any detrimental effects on ruminal fermentation of supplementing large amounts of fat to the diet of dairy cows. 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 adsorb onto bacteria (Chalupa et. al. 1984). Saturated fatty acids also react more readily with metal ions to form insoluble salts of fatty acids (Jenkins and Palmquist. 1982). Feeding preformed Ca salts of fatty acids (CaS) does not alter fermentation in the rumen because of their insolubility (Chalupa et. al. 1986), provided the pH of the rumen is maintained above 6.0 (Palmquist et. al. 1986). Liquefying a mixture of fatty acids high in saturated fatty acid content and spraying the mixture of fatty acids under pressure into a cooled atmosphere results in a dried prilled fatty acid supplement that is inert in the rumen and does not alter rumen fermentation (Grummer, 1988). *Trans*-18:1 fatty acids (**tFA**) and conjugated linoleic acids (**CLA**) produced during biohydrogenation of polyunsaturated fatty acids (**PUFA**) in the rumen are mixtures of positional

isomers (Griinari and Bauman, 1999), which subsequently are incorporated into milk fat of lactating cows (Loor and Herbein, 2001; Piperova et al., 2004; Precht et al., 2002). Studies have demonstrated that levels of tFA (Kalscheur et al., 1997; Griinari et al., 1998) and CLA isomers Peterson et. al., 2003) increase in milk fat of lactating cows fed a variety of diets associated with milk fat depression (MFD). Feeding diets supplemented with Ca salts of fatty acids is a practical way to bypass rumen biohydrogenation. Studies have already shown that diets supplemented with Ca salts of CLA (Ca-CLA) effectively reduce milk fat percentage and yield in lactating cows (Giesy et al., 2002; Perfield II et al., 2002; Bernal-Santos et al., 2003). Dietary Ca salts of tFA (Ca-tFA) supplementation could potentially increase the post ruminal availability of *trans*-18:1 fatty acids and provide an alternative to abomasal infusion to test the potential effects of *trans*-18:1 fatty acids on milk fat synthesis. A current interest in milk fat manipulation is to produce milk with higher proportions of unsaturated fatty acids. Milk with higher proportions of unsaturated fat is likely to be preferred by the consumer because of concerns about too much saturated fat in the diet. An increase in saturated fatty acid intake has been linked to a rise in cardiovascular disease. Dairy feeds that contain lipids with low susceptibility to ruminal biohydrogenation (e.g., oilseeds vs. free oil) can influence milk fatty acid composition. Milk from cows fed high oil corn, an oilseed, has been shown to contain more unsaturated fatty acids than milk from cows fed conventional corn. Therefore, the present was undertaken to study the feeding effect of Ca-salts of fatty acid on fatty acids profile in milk fat.

## **Materials and Methods**

### **Experimental site**

The experiment was conducted at Pachutia Dairy Farm, Animal Production Research Division, Bangladesh Livestock Research Institute (BLRI), Savar, Dhaka for a period of 30 days from 1<sup>st</sup> April to 1<sup>st</sup> May 2010.

### **Dietary treatments**

Twenty cows were divided into five groups considering similar milk yield and parity. The cows in each block were assigned at random to four dietary treatments having five animals in each treatment. The dietary treatments were T<sub>0</sub> = German grass + Concentrate mixture without Ca

salt of fatty acid, T<sub>1</sub>= German grass + Concentrate mixture + 2 % Ca salt of fatty acid, T<sub>2</sub>= German grass + Concentrate mixture + 2.5 % Ca salt of fatty acid, T<sub>3</sub>= German grass + Concentrate mixture + 3 % Ca salt of fatty acid.

### Formulation of diets

Four diets were formulated using green German grass and concentrate mixture with different proportions of Calcium salt of fatty acid. Ingredient and chemical composition of experimental diets are shown in Table 1.

**Table 1. Ingredient and chemical composition of concentrate mixture**

| Items                             | Dietary Treatments <sup>#</sup> |                |                |                |
|-----------------------------------|---------------------------------|----------------|----------------|----------------|
|                                   | T <sub>0</sub>                  | T <sub>1</sub> | T <sub>2</sub> | T <sub>3</sub> |
| <b>Ingredient composition (%)</b> |                                 |                |                |                |
| German grass                      | Ad lib.                         | Ad lib.        | Ad lib.        | Ad lib.        |
| Wheat Bran                        | 46                              | 46.5           | 46             | 45.5           |
| Maize crushed                     | 10                              | 10             | 10             | 10             |
| Wheat broken                      | 20                              | 20             | 20             | 20             |
| Soyabean meal                     | 12                              | 12             | 12             | 12             |
| Til oil cake                      | 8                               | 8              | 8              | 8              |
| Fish meal                         | 1                               | 1              | 1              | 1              |
| DCP                               | 2.5                             | -              | -              | -              |
| Salt                              | 0.5                             | 0.5            | 0.5            | 0.5            |
| Calcium salt of fatty acids       | -                               | 2.0            | 2.5            | 3.0            |
| <b>Chemical composition</b>       |                                 |                |                |                |
| DM (g/100 g sample)               | 89.75                           | 91.32          | 91.69          | 91.06          |
| OM                                | 90.73                           | 89.98          | 88.33          | 89.49          |
| CP                                | 15.53                           | 15.45          | 15.40          | 15.28          |
| ADF                               | 26.05                           | 19.00          | 23.72          | 23.79          |
| NDF                               | 52.04                           | 55.69          | 53.76          | 50.33          |
| Ash                               | 9.27                            | 10.02          | 11.67          | 10.51          |

<sup>#</sup>T<sub>0</sub> = German grass + Concentrate mixture without Ca salt of fatty acid, T<sub>1</sub>= German grass + Concentrate mixture + 2 % Ca salt of fatty acid, T<sub>2</sub>= German grass + Concentrate mixture + 2.5 % Ca salt of fatty acid, T<sub>3</sub>= German grass + Concentrate mixture + 3% Ca salt of fatty acid \* Fresh basis

### Nutritional analysis

Dry matter, OM, CP and total ash of feed and fecal materials were determined by following the methods described by AOAC (1990). Neutral detergent fiber (NDF) and acid detergent fiber (ADF) were estimated by the methods of Van Soest et al., (1991). The fatty acid profile of milk fat were determined according to the procedure developed by Ruiz et al., (2004) through using Gas Chromatography in the Institute of Food Science and Technology (IFST), BCSIR, Dhaka.

### Milk analysis

Milk samples were collected from each cow at 7 days interval and were analyzed for fat, protein, lactose, SNF and minerals contents by milk analyzer (Lactostar, Funke Gerber).

#### **Analysis of fatty acids profile**

The fatty acid profile of milk fat was analyzed at the Institute of Food Science and Technology (IFST), BCSIR, Dhaka following the method of Delve et al., (2004) by using GC.

#### **Statistical analysis**

The data were analyzed using “MSTAT” statistical program to compute analysis of variance (ANOVA) for a Randomized Complete Block Design (RCBD). Least significant difference (LSD) test was also done to compare the treatment means for different parameters. In the analysis of milk parameters, initial milk yield was taken as covariate and then analyzed accordingly.

### **Results and Discussion**

The results of the present study showed that the saturated fatty acids in milk fat were 61.12, 57.83, 54.93 and 54.56 % and unsaturated fatty acids of milk fat were 38.36, 42.22, 45.22 and 45.5 % in treatment groups  $T_0$  ,  $T_1$  ,  $T_2$  and  $T_3$ , respectively (Table 2). Significantly higher ( $p<0.01$ ) unsaturated fatty acid was found in milk under  $T_3$  treatment group and the lowest in  $T_0$  treatment group. There is no significant difference ( $p>0.05$ ) between  $T_2$  and  $T_3$  treatments (Table 3). The palmetic acid content in milk fat were 25.47, 27.22, 26.22 and 26.22% in treatment  $T_0$  ,  $T_1$  ,  $T_2$  and  $T_3$  respectively. The highest ( $p<0.05$ ) palmetic acid was found in  $T_2$  treatment and the lowest in  $T_0$  treatment. The stearic acid content in milk fat were 22.98, 15.78, 13.74 and 13.68% in treatment groups  $T_0$  ,  $T_1$  ,  $T_2$  and  $T_3$  groups, respectively. The ( $p<0.01$ ) difference in the treatment groups for stearic acid in milk fat was significant. However, LSD value indicates that the difference between  $T_2$  and  $T_3$ . The highest stearic acid content was found in  $T_0$  treatment and lowest in  $T_3$  treatment groups (Table 2). The oleic acid content in milk fat were 38.70, 37.27, 40.38 and 40.29% in treatment groups  $T_0$  ,  $T_1$  ,  $T_2$  and  $T_3$  groups, respectively. There was a significant ( $p<0.01$ ) difference between treatment groups  $T_0$  and  $T_1$  for oleic acid content of milk fat, but no significant difference was found between  $T_2$  and  $T_3$ . The highest oleic acid was found in  $T_3$  treatment and lowest in  $T_1$  treatment. This finding corroborate with the findings of Sultana et al., (2008), who reported that feeding of Ca-Salt of Linseed oil significant decreased the proportion of saturated FA C16:0. They also observed that the proportions of long chain fatty acids, namely, C18:0, C18:1, C18:2, VA and CLA increased significantly ( $p<0.01$  and  $p<0.05$ )

for CaSO or CaLO treatments compared with controls. Those findings also further supported by the previous study of Chouinard et al. (1998).

**Table 2. Effect of Ca salt of fatty acid on Fatty acid profile (%) of milk fat**

| Parameters                     | Dietary Treatments <sup>#</sup> |                    |                    |                    | SED   | Level of Sig. |
|--------------------------------|---------------------------------|--------------------|--------------------|--------------------|-------|---------------|
|                                | T <sub>0</sub>                  | T <sub>1</sub>     | T <sub>2</sub>     | T <sub>3</sub>     |       |               |
| <b>Saturated fatty acids</b>   | 61.12 <sup>c</sup>              | 57.83 <sup>b</sup> | 54.93 <sup>a</sup> | 54.56 <sup>a</sup> | 0.798 | **            |
| Palmetic acid                  | 25.47 <sup>a</sup>              | 27.22 <sup>d</sup> | 26.22 <sup>c</sup> | 26.22 <sup>b</sup> | 0.195 | **            |
| Stearic acid                   | 22.98 <sup>c</sup>              | 15.78 <sup>b</sup> | 13.74 <sup>a</sup> | 13.68 <sup>a</sup> | 1.150 | **            |
| <b>Unsaturated fatty acids</b> | 38.36 <sup>a</sup>              | 42.22 <sup>b</sup> | 45.22 <sup>c</sup> | 45.5 <sup>c</sup>  | 0.875 | **            |
| Oleic acid                     | 38.70 <sup>a</sup>              | 37.27 <sup>b</sup> | 40.38 <sup>c</sup> | 40.29 <sup>c</sup> | 0.943 | **            |
| Linoleic acid                  | 1.57 <sup>a</sup>               | 1.64 <sup>a</sup>  | 1.73 <sup>a</sup>  | 1.94 <sup>b</sup>  | 0.482 | **            |
| Linolenic acid                 | 0.38 <sup>a</sup>               | 0.64 <sup>b</sup>  | 0.68 <sup>b</sup>  | 1.25 <sup>c</sup>  | 0.346 | **            |

\*\*Significant at (p<0.01) level of significant, <sup>a,b,c,d</sup> mean values having different superscripts in a row different significantly (P<0.01)

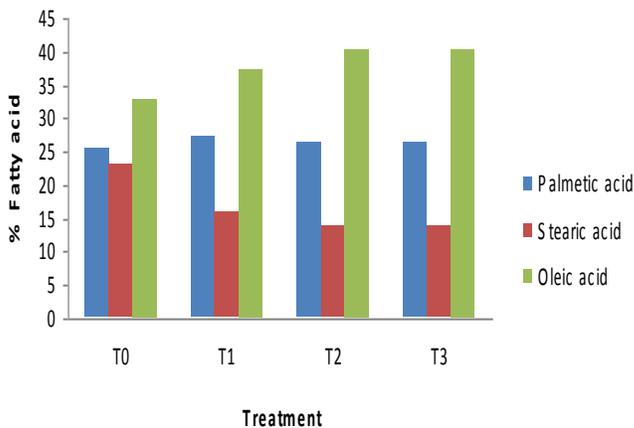
The linoleic acid content in milk fat is shown in Table 2. It can be seen that the difference among the treatment groups of T<sub>0</sub>, T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub> for linoleic acid content of milk fat, but no significant differences were found among the T<sub>0</sub>, T<sub>1</sub> and T<sub>2</sub> treatment groups. The linolenic acid content in milk fat were 0.38, 0.64, 0.68 and 1.25% in treatment groups T<sub>0</sub>, T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub> respectively. There is a significant (p<0.01) difference in the treatment groups for linolenic acid of milk fat, except between T<sub>1</sub> and T<sub>2</sub>. The percent increase and/or decrease of different saturated and unsaturated fatty acids due to feeding of Ca-salts of fatty acid is shown in Table 3. The results indicated that the percent decreased of total saturated fatty acids were higher in T<sub>3</sub> (-10.73%) followed by T<sub>2</sub> (-10.12 %) and T<sub>1</sub> (-5.38%). Meanwhile, the percent increased of total unsaturated fatty acids were higher in T<sub>3</sub> (+18.61%) followed by T<sub>2</sub> (+17.88%) and T<sub>1</sub> (+10.06%). Considering the individual saturated fatty acid, the percent increased of palmetic acid (6.87%) was highest in T<sub>1</sub> treatment compared to that of T<sub>2</sub> and T<sub>3</sub>. On the contrary, the percent decreased of stearic acid was the highest (-40.46%) in T<sub>3</sub> group followed by T<sub>2</sub> (-40.20%) and T<sub>1</sub> (31.33%), treatments, respectively. In the case of unsaturated fatty acids, the percent increased of linolenic acid was higher compared to that of oleic and linoleic acids. The values of percent increased of linolenic acid were +68.42, +78.94 and +228.94 %, respectively for T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub> treatment groups. While in the case of linoleic acid, the percent increased was higher in T<sub>3</sub>

followed by T<sub>2</sub> and T<sub>1</sub>. On the other hand, the percent increased of oleic acid was higher in T<sub>2</sub> compared to that of T<sub>3</sub> and T<sub>1</sub>. The effect of feeding Ca salt of fatty acid on palmitic, stearic and oleic acids are also shown in Figure 1. It can be seen from the Figure 1 that with the increase of Ca-salt of fatty acid in the diet the oleic acid % was increased also except T<sub>3</sub> treatment. The coefficients of regression between feeding Ca-salts of fatty acids with milk fatty acids composition are shown in Figure 2, and 3. Figure 2 shows the regression coefficient between levels of feeding of Ca-salt of fatty acid with saturated and unsaturated fatty acids.

**Table 3 . Effect of feeding Ca salt of fatty acid on Fatty acid profile (%) of milk fat**

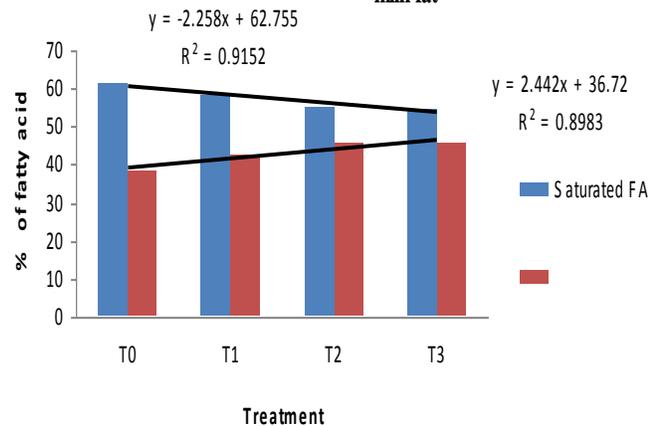
| Parameters                | Dietary Treatments <sup>#</sup> |                |                |                | % increase (+) or decrease (-) compare to control (T <sub>0</sub> ) |                |                |
|---------------------------|---------------------------------|----------------|----------------|----------------|---------------------------------------------------------------------|----------------|----------------|
|                           | T <sub>0</sub>                  | T <sub>1</sub> | T <sub>2</sub> | T <sub>3</sub> | T <sub>1</sub>                                                      | T <sub>2</sub> | T <sub>3</sub> |
| Saturated Fatty acid      | 61.12                           | 57.83          | 54.93          | 54.56          | -5.38                                                               | -10.12         | -10.73         |
| Unsaturated Fatty acid    | 38.36                           | 42.22          | 45.22          | 45.5           | +10.06                                                              | +17.88         | +18.61         |
| <b>Fatty acid profile</b> |                                 |                |                |                |                                                                     |                |                |
| Palmitic Acid             | 25.47                           | 27.22          | 26.22          | 26.22          | +6.87                                                               | +2.94          | +2.94          |
| Stearic Acid              | 22.98                           | 15.78          | 13.74          | 13.68          | -31.33                                                              | -40.20         | -40.46         |
| Oleic Acid                | 38.70                           | 37.27          | 40.38          | 40.29          | -3.69                                                               | +4.34          | +4.10          |
| Linoleic acid             | 1.57                            | 1.64           | 1.73           | 1.94           | +4.45                                                               | +10.19         | +23.56         |
| Linolenic acid            | 0.38                            | 0.64           | 0.68           | 1.25           | +68.42                                                              | +78.94         | +228.94        |

**Effect of Ca salt of fatty acid on fatty acid profile of milk fat**



**Figure 1.** Feeding effect of Ca-salt on milk fatty acid

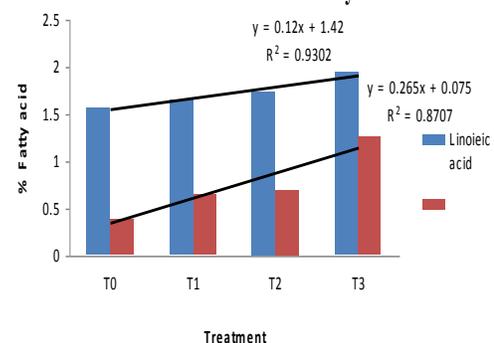
**Effect of Ca salt of fatty acid on saturation and unsaturation of milk fat**



**Figure 2.** Regression co-efficients between levels of Ca-salt of FA and saturated FA

It can be seen that the regression coefficient between levels of Ca-salt of fatty acid and saturated fatty acid

**% of linoleic and linolenic fatty acid of milk fat**



was -2.258 (slope of the curve), while the regression value was 0.91( $R^2$ ). It should be noted that one unit increase of Ca-salt of fatty acid in the diet, corresponds the respective reduction of saturated fatty acid was -2.258. Similarly, the regression coefficient between levels of Ca-salt of fatty acid and unsaturated acid was 2.44, while the regression value was 0.89. The relation indicated that a unit increase Ca-salt in the diet was associated with an increase of 2.44 kg unsaturated fatty acid in milk fat. Hence, feeding Ca-salt of fatty acid was better correlated with both saturated and unsaturated fatty acids. On the other hand, Figure 3 shows the regression coefficient between Ca- salt of fatty and unsaturated fatty acids. The results revealed that the regression coefficient between levels of Ca-salt of fatty acid and linoleic acid was 0.12, while the regression ( $R^2$ ) correlation was 0.9312. In the case of linolenic acid, it was observed that the regression coefficient between levels of Ca-salt of fatty acid and linolenic acid was 0.26, while the correlation was 0.87 (Figure 3). It can be explained from the results that a unit increase of Ca-salt of fatty acid in the diet was associated with an increase of 0.26 kg linolenic acid in milk fat.

### **Conclusions**

It may be concluded that the feeding Ca-salts of fatty acids increased unsaturated fatty acids content e.g oleic acid, linoleic and linolenic acid of milk fat, and decreased saturated fatty acid (stearic acid) content in milk fat. Based on the above findings it may be concluded that 2.5 % Calcium salt of fatty acid may be recommended for increasing the unsaturated fatty acids content in milk of dairy cattle.

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