Effect Of Linseed Fed As Ground Seed, Oil And Ca-Salt On Milk Yield And Its Fatty Acid Profile In Dairy Cows

Netsanet Beyero, V.Kapoor, B.S.Tewatia

Abstract: Experiment was conducted on twenty-four crossbred dairy cows divided into four groups of six animals in each group. Cows were fed on iso-nitrogenous and iso-caloric ration consisting of concentrate mixture and roughage. The concentrate mixture of four treatment groups differed with regard to the source of fat/oil and contained GNC (T_1), ground linseed (T_2), Ca-salt of linseed oil (T_3) and linseed oil (T_4). Roughages offered were green berseem forage and wheat straw. DMI (kg/d, g/ $W^{0.75}$kg, or % BW) did not vary significantly among treatments. Digestibility (%) of nutrients (DM, OM, CP, CF, EE, NFE, NDF and ADF) did not vary among treatments. Daily average milk yield and 4% FCM yield did not vary significantly among the treatments. Milk yield (kg/d) ranged between 12.40 (T_3) and 13.38 (T_2). Percentage of milk protein, fat, lactose, MUN, Cholesterol, TS and total ash were not affected by different treatments. TLCFA and PUFA were significantly ($P < 0.05$) higher in T_3 than in T_1, T_2 and T_4. Total PUFA yield (g/100g fat) increased by 137.34% in cows fed bypass fat as compared to control group (T_1). Total CLA yield (mg/g fat) improved by 74.97% in cows fed linseed over T_1. The total feeding cost was highest (USD 3.33/d) in T_3, followed by (USD 2.76/d) T_4, (USD 2.56/d) T_2, and (USD 2.49/d) T_1. The feeding cost per g PUFA production reduced by 42% in cows fed bypass fat than control diet. The feeding cost per g CLA production reduced by 44% in cows fed linseed than control diet. In summary, these results suggest that milk fatty acid profile is improved for better human health by feeding linseed products, as shown by higher concentrations of PUFA including CLA recognized as being beneficial to reduce the incidence of cancer, cardiovascular diseases and stimulate the immune system.

Index Terms: Dairy cow, linseed oil, milk fatty acid, conjugated linoleic acid

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1. Introduction

t contains a substantial concentration of saturated fatty acids ($C_{14:0}$ and $C1_{6:0}$) and relatively low concentrations of monounsaturated and polyunsaturated fatty acids (Kennelly and Glimm, 1998). Increasing specific polyunsaturated fatty acids such as linoleic acid ($C_{18:2}$) and linolenic acid ($C_{18:3}$) in milk would increase consumer interest and acceptance of milk due to health benefits associated with these fatty acids [1]. Among the milk fatty acids that has gained attention is conjugated linoleic acid (CLA) ($C_{18:2}$ cis-9 trans-11) because it is unique in its ability to inhibit carcinogenesis in experimental animals. It has been observed to reduce the incidence of tumours in a number of experimental animal models and serve as a cytotoxic agent against existing tumour cells [2]; [3]; [4]. It has also been shown that CLA may have positive effects on cardiovascular risk factors in animal models [5]; [6]. CLA is an intermediary product of ruminal biohydrogenation of dietary lipids.

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The biohydrogenation in the rumen that can alter the CLA production in milk fat is affected by the type and the amount of fatty acid substrate in the diet [7]; [8]. The proportion of milk CLA should increase as the concentration of polyunsaturated fatty acids, especially $C_{18:2}$ and $C_{18:3}$ increase in the lactation diet of dairy cow. Linseed (Linum usitatissimum) is a rich source of α-linolenic acid constituting 53% of the total fatty acids [9]. However, feeding linseed produced relatively small changes in the concentrations of C18:2 and C18:3 in milk fat, likely due to extensive ruminal biohydrogenation of the linseed polyunsaturated fatty acids [6]. We speculated that beneficial effects of feeding linseed to dairy cows may be enhanced by protecting it from extensive rumen biohydrogenation. A variety of fat sources as Ca-salt have been fed to increase the concentration of CLA as a method to protect dietary lipids from ruminal biohydrogenation [10]. This study was designed to assess the effect of different sources of linseed oil (ground linseed, Ca-salt of linseed oil and linseed oil) on milk yield and its fatty acid composition

2. Material and Methods

2.1. Experimental animals and management

The study was conducted on the Animal farm of College of Veterinary Sciences, Lala Lajpat Rai University of Veterinary and Animal Sciences (LLRUVAS), Hisar, India. Twenty four Cross bred dairy cattle in second to fourth lactation, with average body weight of 395.4±43.36kg and average daily milk yield 11.65±2.49kg were divided into four dietary groups of six in each group on the basis of milk yield and lactation stage. All the experimental animals were housed in well ventilated shed and maintained under clean and hygienic condition in the barn of having the arrangement of individual feeding. Fresh and clean tap water was provided free choice to all animals. The experimental animals were given twenty-one days adaptation period. Before starting the experiment, the animals were adapted for 21 days to the diets. The duration of experiment was 60 days from February to April 2011.

2.2. Experimental diets and treatments

The experimental animals were kept on a diet containing concentrate mixture and roughage (berseem green forage and wheat straw) (see Table 2.1). Berseem green forage was harvested daily, and chopped manually to approximately 5-7 cm length and mixed before feeding. Cows were fed to meet their requirements [11]. Cows were fed on iso-nitrogenous and iso-caloric ration. The concentrate mixture of four treatment groups differed with regard to the source of fat/oil and contained GNC (T1), ground linseed (T2), Ca-salt of linseed oil (T3) and linseed oil (T4). Calcium salt of linseed oil was prepared using Fusion method.

Table 2.1 Percent composition of ingredients in concentrate mixture

Ingredients	T_1	T_2	T_3	T_4
Groundnut cake	40	–	–	–
Deoiled groundnut cake	–	31.5	35	35
Wheat	30	30	30	30
Barely	27	27	28.5	29
Ground linseed	–	8.5	–	–
Ca-salt of linseed oil	–	–	3.5	–
Linseed oil	–	–	–	3
Mineral mixture	2	2	2	2
Common salt	1	1	1	1

2.3. Data collection

The experimental feedstuffs were weighed before feeding. Feed refusals were collected from individual animals every morning and weighed. Feed offered and refusals were sampled daily, bulked foe each animal and sub-sampled for chemical analysis. Amount of all feedstuffs (Berseem forage, wheat straw and concentrate mix) offered and refused were measured to quantify feed intake. Animals were hand milking twice a day at 4 a.m. and 5 p.m. during the experimental period and milk yield was recorded by using circular dial type spring balance, with the capacity of 20 kg and an accuracy of ± 0.05 kg, at each milking time. Milk samples for analysis were taken from morning and evening milking at weekly interval. Hundred fifty ml of milk sample was taken from eachmilking and each animal in milk sampling bottles. The milk samples from morning and evening samples were pooled and milk was thoroughly mixed before analysis.

2.4. Digestibility trial

A digestibility trial of five days collection period was conducted at the end of experimental period to determine the digestibility of nutrients. The feeding regime during the digestibility trial was the same as in the feeding experiment. During trials, the sample of feed offered, residue left and faeces voided were collected and the representative samples of daily feeds offered, residue left and faeces voided were collected and oven dried. For nitrogen estimation, 1/600th part of the total faeces was weighed and preserved in 40% H_2SO_4, in wide mouthed plastic bottles. At the end of the trial, samples from each animal were pooled, mixed and grounded to 1mm sieve size. The ground samples were stored for further analysis.

2.5. Chemical analysis

Samples of feeds offered, refusals and faeces wereanalyzed for dry matter (DM), total ash, crude protein (CP), ether extract (EE) and crude fibre (CF) as in [12], neutral detergent fiber (NDF), acid detergent fiber (ADF) and acid detergent lignin (ADL) contents [13]. The calcium content of feed samples was determined by the atomic absorption spectrophotometry and Phosphorusconcentration was determined by colorimetric method. Samples of milk were analyzed for total solid, total fat and protein according to [12]. Milk lactose content [14],cholesterol [15], milk urea nitrogen (MUN) [16], Fatty acid composition Sukhjia and Palmquist, 1984 modified by [17], CLA content [18].

2.6. Statistical analysis

The data were subjected to analysis of variance using SPSS statistical software version 16.0 for Windows. The means which showed significant differences at the probability level of $P < 0.05$ were compared with each other by using LSD.

3. Results and Discussion

3.1 Chemical composition of the experimental diets

The chemical composition of experimental diets (concentrate mixture and roughage as berseem (green) and wheat straw) is given in Table 3.1. Diets were formulated regards to various chemical constituents to be similar among treatments as possible.Protein (12.3-13%) was obtained from T3 and T4 respectively.

Table 3.1 Chemical composition of dietary ingredients (concentrate mixtures, berseem forage and wheat straw).

Attribute	Concentrate mixture				Berseem forage	Wheat straw
	T1	T2	T3	T4		
DM	91.78	91.78	91.80	91.78	35.24	90.35
OM	92.81	93.22	92.4	93.7	89.6	93.27
NFE	54.54	54.77	53.67	53.11	40.51	47.15
CP	23.39	23.61	23.46	23.78	14.58	2.10
EE	7.55	7.46	7.36	7.65	4.32	1.55
CF	7.33	7.48	6.97	7.44	30.19	41.97
Ash	7.19	6.78	7.60	6.30	10.40	6.73
NDF	31.60	31.90	32.30	33.23	56.30	79.00
ADF	11.22	12.26	10.90	11.71	38.40	49.85
ADL	3.90	3.54	2.94	3.21	11.30	12.55
Calcium	1.32	1.36	1.41	1.29	1.44	0.31
Phosphorus	0.74	0.72	0.69	0.69	0.29	0.10

Table 3.2 Dry matter intake (DMI), nutrients intake and digestibility of nutrients in experimental cows

Attribute	T1	T2	T3	T4
Feed Intake (Kg/d)				
Roughage	7.36±0.19	7.48±0.30	6.95±0.32	7.14±0.25
Concentrate mixture	6.88	6.88	6.88	6.88
Total DMI (kg/d)	14.24±0.19	14.36±0.30	13.83±0.32	14.02±0.25
DMI (% Body Wt)	3.40±0.08	3.46±0.11	3.35±0.08	3.42±0.09
DMI (g/ W0.75kg)	153.94±2.77	155.96±3.35	150.75±2.97	153.92±3.02
Nutrient Intake (kg/d)				
DCP	0.71±0.01	0.72±0.03	0.67±0.01	0.69±0.01
TDN	6.70±0.15	6.71±0.11	6.38±0.13	6.49±0.06
Nutrient digestibility (%)				
DM	67.10±0.78	66.37±0.76	67.66±0.91	65.30±0.89
OM	69.92±0.72	69.47±0.64	70.46±0.79	68.60±0.66
NFE	73.06±1.09	71.41±0.68	73.32±1.25	70.69±0.62
CP	70.28±1.04	68.82±1.20	69.67±0.76	67.59±2.07
EE	65.50±0.88	66.30±0.69	66.81±1.21	66.63±1.30
CF	66.22±0.91	65.88±0.82	65.84±0.64	65.00±1.43
NDF	59.95±1.11	58.65±1.09	58.97±0.82	58.36±1.26
ADF	42.12±1.23	41.78±1.31	41.56±1.37	41.06±0.90

The digestibility (%) of DM and OM did not vary significantly among the four treatment groups (Table 3.2). These observations are consistent with the finding of [19] who reported similar ruminal digestibility of DM in the cows fed diets supplemented with 7.5% rolled linseed, 7.5% extruded linseed, or 2.6% linseed oil. However, lower digestibility of DM, OM, NDF, and ADF has been reported in cows fed 12% (DM basis) whole linseed in the diet [20]. In the present experiment, the level of supplementation linseed oil products might not be high enough to depress intake of dry matter and its digestibility as reported by earlier workers. The intake of DCP and TDN did not vary significantly among various treatment groups (Table 3.2). The lack of significant difference in the DCP and TDN intakes in treatment groups may be due to a similar DMI and digestibility of DM.

Table 3.3 Milk yield and milk composition of cows fed experimental diets

Attribute	**T_1**	**T_2**	**T_3**	**T_4**
Milk yield (kg/d)	12.62±0.77	13.38±0.76	12.40±0.76	12.91±0.89
FCM (kg/d)	12.18±0.78	12.78±0.75	11.94±0.77	12.41±0.89
Fat yield (kg/d)	0.48±0.03	0.50±0.03	0.47±0.03	0.48±0.04
Milk composition (%)				
Fat	3.77±0.03	3.74±0.03	3.75±0.04	3.70±0.02
Crude protein	3.27±0.05	3.25±0.05	3.23±0.01	3.24±0.02
Lactose	4.47±0.02	4.45±0.01	4.44±0.01	4.46±0.02
Total solids	12.01±0.12	12.06±0.05	12.22±0.08	12.07±0.10
SNF	8.25±0.13	8.32±0.04	8.48±0.10	8.34±0.09
Total ash	0.71±0.01	0.72±0.01	0.74±0.01	0.71±0.01
Calcium	0.104±0.005	0.106±0.004	0.110±0.002	0.105±0.002
Phosphorus	0.062±0.001	0.064±0.002	0.065±0.001	0.062±0.001
MUN (mg/100 ml)	13.64±0.08	14.07±0.11	13.04±0.10	13.21±0.07
Cholesterol (mg/100 ml)	6.65±0.20	6.73±0.20	6.72±0.28	6.15±0.16

The milk yield was numerically higher in cows T2 group and lower in T3 (Table 3.3) yet non-significant differences were observed among different treatment groups. The results of the present study are in agreement with the results obtained by [21]; [6] who found that non-significant different in milk yield by adding rolled flaxseed to dairy cow diets up to 10% of DM or as in [22] ground linseed up to 7.5% of the DM and [23] cows fed with 4% Ca-salts of canola oil, soybean oil, or linseed oil. The data obtained in the present study revealed that milk fat percentage was not affected by dietary treatments; however, cows fed GNC based concentrate (T1) tended to have higher milk fat as compared to other groups (Table 3.3). On the contrary, a reduction of milk fat due to inclusion of linseed in the diets at the rate of 4-5% [24] or 5.1-7.5% as ground linseed [22]; [25] in dairy cows has been reported. However, when the effect of supplementary oilseeds and Ca-salt of fatty acids on milk fat is compared, milk fat concentration was found to be higher or similar [9] in cows fed Ca-salt of fatty acids of palm oil than those fed formaldehyde-treated linseed, while in other studies feeding of Ca-salt of linseed oil (4% of DM) to dairy cows resulted in decreased milk fat percentage compared with a control diet [23]. Milk fat depression can occur if fat supplementation increases the synthesis of certain trans FA such as trans-10 C18:1 or trans-10, cis-12 C18:2 in ruminal fluid [26]; [27]. However, in the present study lower doses of linseed products in the diet might be a reason for the lack of significant differences between treatments. Milk protein, lactose and MUN concentrations were did not show significant difference among the treatment groups (Table 3.3).

3.2. Fatty acid intake and milk fatty acid profile

Table 3.4 Fatty acid composition (mg/g DM) of dietary ingredients (concentrate mixtures, berseem forage and wheat straw)

Fatty acid	Concentrate mixture				Berseem forage	Wheat straw
	T1	T2	T3	T4		
C8:0	0.046	0.056	0.061	0.056	0.044	0.031
C10:0	0.068	0.072	0.069	0.068	0.076	0.013
C12:0	0.040	0.036	0.042	0.04	0.076	0.036
C14:0	0.088	0.112	0.096	0.105	0.092	0.058
C14:1	0.233	0.235	0.252	0.248	0.344	ND
C16:0	12.078	5.244	5.682	5.477	4.233	0.506
C16:1	0.216	0.866	0.901	0.804	0.832	0.039
C18:0	16.344	8.256	8.235	8.342	1.668	0.254
C18:1	18.476	22.455	21.986	22.094	3.135	0.404
C18:2	14.135	18.608	18.772	18.44	7.234	0.388
C18:3	2.446	18.232	18.332	18.078	19.12	0.078
C20:0	0.566	0.403	0.506	0.472	0.734	0.022
Total fatty acids	64.736	74.575	74.934	74.224	37.588	1.829
Total saturated fatty acids	29.23	14.179	14.691	14.56	6.923	0.92
Total unsaturated fatty acids	35.506	60.396	60.243	59.664	30.665	0.909
Total long chain fatty acids	64.261	74.064	74.414	73.707	36.956	1.691
Monounsaturated fatty acids	18.925	23.556	23.139	23.146	4.311	0.443
Polyunsaturated fatty acids	16.581	36.84	37.104	36.518	26.354	0.466

ND- Not detectable

i. Fatty acids intake

The total saturated fatty acids (TSFA) intake was significantly (P <0.05) higher in T_1 than T_2, T_3 and T_4 while TLCFA, TUFA, MUFA and PUFA intake significantly higher in T_2, T_3 and T_4 (Table 3.5). The stearic acid intake was significantly higher in T_1 (GNC based concentrate), and linoleic and linolenic acid intakes were significantly (P <0.05) lower as compared to diets supplemented with linseed products (T_2, T_3 and T_4) Table 3.5. Perusal of Table 3.4 (fatty acid composition of dietary ingredients) revealed that T_1 group had higher stearic acid concentration while lower linoleic and linolenic acids as compared to T_2, T_3 and T_4 diets. Similarly, total saturated fatty acids (TSFA) intake was significantly (P <0.05) higher in T_1 than T_2, T_3 and T_4 while TLCFA, TUFA, MUFA and PUFA intake was significantly higher in T_2, T_3 and T_4 (Table 4.5). TSFA intake ranged between 141.67 (T_2) and 244.60g/d (T_1) while TUFA intake varied from 433.01 in T_1 to 670.30g/d in T_2. The intake of PUFA (g/d) was in the range of 275.88 and 417.55 in T_1 and T_2, respectively. Generally, the higher intake of saturated fatty acids and lower intake of unsaturated fatty acids in T_1 over T_2, T_3 and T_4 was the reflection of fatty acid composition of the diets.

Table 3.5 Fatty acids intake of cows fed diets containing linseed, Ca-salts of linseed oil and linseed oil

Fatty acid Intake (g/d)	T_1	T_2	T_3	T_4
$C_{8:0}$	0.63±0.01^{d}	0.70±0.01^{b}	0.71±0.01^{a}	0.68±0.01^{c}
$C_{10:0}$	0.95±0.01^{ab}	0.98±0.02^{a}	0.93±0.02^{b}	0.94±0.02^{ab}
$C_{12:0}$	0.79±0.01	0.77±0.02	0.77±0.02	0.77±0.02
$C_{14:0}$	1.24±0.02^{c}	1.42±0.03^{a}	1.26±0.03^{c}	1.34±0.02^{b}
$C_{14:1}$	3.71±0.05	3.75±0.08	3.73±0.08	3.75±0.07
$C_{16:0}$	109.62±0.62^{a}	62.99±0.98^{b}	64.24±1.05^{b}	63.46±0.82^{b}
$C_{16:1}$	6.62±0.1	11.17±0.	11.07±0.	10.53±0.

	2^{c}	19^{a}	20^{a}	16^{b}
$C_{18:0}$	122.97 ± 0.25^{a}	67.47 ± 0.39^{b}	66.63 ± 0.42^{b}	67.62 ± 0.33^{b}
$C_{18:1}$	146.80 ± 0.46^{c}	174.45 ± 0.73^{a}	169.92 ± 0.78^{b}	171.13 ± 0.61^{ab}
$C_{18:2}$	141.99 ± 1.04^{b}	173.39 ± 1.64^{a}	171.59 ± 1.76^{a}	170.35 ± 1.38^{a}
$C_{18:3}$	133.89 ± 2.69^{b}	244.15 ± 4.26^{a}	237.20 ± 4.58^{a}	238.19 ± 3.59^{a}
$C_{20:0}$	8.41 ± 0.10^{a}	7.36 ± 0.17^{b}	7.77 ± 0.18^{b}	7.64 ± 0.14^{b}
Total fatty acids	677.61 ± 5.37^{b}	748.59 ± 8.50^{a}	735.83 ± 9.14^{a}	736.39 ± 7.15^{a}
Total saturated fatty acids	244.60 ± 1.02^{a}	141.67 ± 1.61^{b}	142.31 ± 1.73^{b}	142.44 ± 1.35^{b}
Total unsaturated fatty acids	433.01 ± 4.36^{b}	606.92 ± 6.89^{a}	593.52 ± 7.41^{a}	593.95 ± 5.80^{a}
Total long chain fatty acids	670.30 ± 5.28^{b}	740.98 ± 8.35^{a}	728.43 ± 8.98^{a}	728.91 ± 7.03^{a}
Monounsaturated fatty acids	157.13 ± 0.63^{c}	189.38 ± 0.99^{a}	184.73 ± 1.07^{b}	185.41 ± 0.83^{b}
Polyunsaturated fatty acids	275.88 ± 3.73^{b}	417.55 ± 5.90^{a}	408.79 ± 6.34^{a}	408.54 ± 4.96^{a}

Values bearing different superscripts in a raw differ significantly (p< 0.05).

ii. Fatty acid profile of milk fat

Feeding linseed oil (in the form of linseed, Ca-salt of linseed oil and linseed oil) to cows of groups T_2, T_3 and T_4 led to the lower concentration of milk fatty acids $C_{14:0}$ and $C_{16:0}$ as compared to T_1. The results are in agreement with those of [28]; [6];[27]; [29]who reported a decrease in $C_{14:0}$ and $C_{16:0}$ FA in milk from cows fed either whole or rolled linseed. A lower proportion of short – and medium – chain fatty acids indicated a reduction in de novo synthesis of fatty acids in the mammary gland [30]. As in [31] who cited two possible reasons for reduction of the short chain fatty acids in milk; an adverse effect of polyunsaturated fatty acids which decrease the supply of acetate and β-hydroxybutrate for de novo synthesis of these fatty acids and direct inhibitory effect of dietary long chain fatty acids on acetyl-coenenzyme-A carboxylase activity (rate-limiting enzyme for de novo fatty acid synthesis). Cows in T_4 group had significantly (P <0.05) higher concentration of $C_{18:0}$ (21.67±0.21) than those in T_1 (19.62±0.30), T_2 (18.95±0.24) and T_3 (19.03±0.14g/100g of milk fat) groups while the concentration of C18:1 was highest in cows fed diet T_1. The concentration of total MUFA followed the trend of $C_{18:1}$, which comprised 87.45% of total MUFA, and was highest in cows fed GNC based concentrate mixture. The concentration of PUFAs, namely $C_{18:2}$ and $C_{18:3}$ were significantly (P <0.05) higher in cows fed diets having Ca-salt of linseed oil as compared to cows fed GNC, ground linseed and linseed oil.

Table 3.6 Fatty acid composition (g/100g) of milk fat in cows fed linseed, Ca-salts of linseed oil, and linseed oil

Fatty acids	T_1	T_2	T_3	T_4
$C_{8:0}$	1.14±0.09	1.12±0.07	1.38±0.15	1.30±0.08
$C_{10:0}$	2.03 ± 0.13^{c}	2.13 ± 0.09^{bc}	2.50 ± 0.16^{ab}	2.65 ± 0.14^{a}
$C_{12:0}$	2.57 ± 0.14^{b}	2.56 ± 0.13^{b}	2.97 ± 0.08^{a}	3.01 ± 0.10^{a}
$C_{14:0}$	14.57 ± 0.24^{a}	14.00 ± 0.18^{b}	12.02 ± 0.13^{c}	14.07 ± 0.19^{b}
$C_{14:1}$	1.44 ± 0.11^{ab}	1.25 ± 0.07^{b}	1.58 ± 0.10^{a}	1.52 ± 0.04^{a}
$C_{16:0}$	29.66 ± 0.26^{a}	27.32 ± 0.23^{b}	27.26 ± 0.15^{b}	27.78 ± 1.79^{b}
$C_{16:1}$	1.64 ± 0.08^{b}	1.63 ± 0.10^{b}	2.06 ± 0.08^{a}	1.65 ± 0.05^{b}
$C_{18:0}$	19.62 ± 0.30^{b}	18.95 ± 0.24^{b}	19.03 ± 0.14^{b}	21.67 ± 0.21^{a}
$C_{18:1}$	21.39 ± 0.17^{a}	20.50 ± 0.24^{b}	20.05 ± 0.13^{b}	17.98 ± 0.23^{c}
$C_{18:2}$	2.48 ± 0.07^{c}	3.33 ± 0.10^{b}	5.05 ± 0.06^{a}	2.59 ± 0.13^{c}
$C_{18:3}$	0.86 ± 0.05^{c}	1.64 ± 0.09^{b}	2.86 ± 0.06^{a}	1.30 ± 0.06^{b}
$C_{20:0}$	0.71 ± 0.05^{c}	0.80 ± 0.05^{bc}	0.95 ± 0.05^{ab}	0.99 ± 0.06^{a}
Total saturated fatty acids	70.29 ± 0.49^{a}	66.76 ± 0.41^{b}	66.10 ± 0.30^{b}	70.17 ± 1.94^{a}
Total unsaturated fatty acids	27.79 ± 0.16^{b}	28.35 ± 0.33^{b}	31.60 ± 0.25^{a}	25.03 ± 0.32^{c}
Total long chain fatty acids	76.35 ± 0.48^{ab}	75.87 ± 0.42^{ab}	77.25 ± 0.27^{a}	73.95 ± 1.87^{b}
Monounsaturated fatty acids	24.46 ± 0.17^{a}	23.39 ± 0.29^{b}	23.69 ± 0.20^{b}	21.14 ± 0.25^{c}
Polyunsaturated fatty acids	3.33 ± 0.06^{d}	4.96 ± 0.13^{b}	7.90 ± 0.08^{a}	3.88 ± 0.16^{c}
Total CLA (mg/g fat)	8.47 ± 0.19^{d}	14.82 ± 0.28^{a}	11.19 ± 0.35^{b}	9.46 ± 0.23^{c}

Values bearing different superscripts in a raw differ significantly (p< 0.05).

According to [32]the concentrations of these fatty acids can be increased significantly by feeding linseed that is well protected from ruminal biohydrogenation. Similarly, an increase in the level of α-linolenic acid from 0.49-1.95% was observed by Aii et al. (1991) in the lactating dairy cows upon feeding 0-500 g/d of Ca-salt of linseed oil. In another study [33] observed that the increase was from 0.84 to 2.19% for the 0-5.40% range of Ca-salts of linseed oil in the diets of dairy cows diet. In the present study, the concentration of $C_{18:3}$ increased from 0.86g/100g fat (T_1) to 2.86 (T_3),233% increase in T_3 over control (T_1) group. In spite of the fact that the intake of TUFA and PUFA were statistically similar in the treatment groups T_2, T_3 and T_4, the concentrations of TUFA and PUFA was higher (P <0.05) in the milk of cows in treatment group T_3 fed Ca-salt of linseed oil (Table 3.6). PUFA concentration of milk varied from

3.33±0.06 in T_1 to 7.90±0.08g/100g fat in T_3. The mean values were highest in T_3 and lowest in T_1, which accounted a 137.34% increase in PUFA yield (g/100g fat) of cows fed bypass fat as compared to control group. The lower concentration of TUFA and PUFA in T_2 and T_4 than T_3 might probably be due extensive biohydrogenation of fatty acids in linseed and linseed oil in the rumen, whereas PUFA from bypass fat (Ca-salts of linseed oil) could escape from rumen biohydrogenation. The greater production of long-chain fatty acids in the milk is associated with the increase of mammary uptake of long chain FA from plasma triacylglycerols. Total CLA yield (mg/g fat) increased significantly ($P < 0.05$) by linseed product supplementation. The mean values of CLA were in the order of 14.82±0.28, 11.19±0.35, 9.46±0.23, and 8.47±0.19(mg/g fat)in T_2, T_3, T_4, and T_1, respectively (Table 4.6). Total CLA yield (mg/g fat) improved by 74.97, 32.11, and 11.69% in T_2, T_3, and T_4, respectively over T_1. Results obtained in the present study are in agreement with the work of [8], who supplemented the diet of dairy cows with Ca-salts of linseed oil and observed that cis-9, -trans11-CLA production was increased by 125% compared to control. A similar increase (150%) in cis-9, trans-11 CLA production was observed by [34] upon supplementation of 4% linseed oil in the diets of dairy cows. Milk CLA is derived from rumen CLA as a result of incomplete biohydrogenation of $C_{18:2}$ and $C_{18:3}$ [35] and from endogenous synthesis of CLA in the mammary gland from C18:1 trans-11 [36], which is an intermediate in ruminal biohydrogenation of polyunsaturated fatty acids [37]. It is probable that concentration of milk CLA increases as the concentration of polyunsaturated fatty acids ($C_{18:2}$ and $C_{18:3}$) increases in the diet of the lactating dairy cow (Table 3.4). The value obtained in the present study was substantially higher than the average values of CLA (0.3 to 0.6% milk fat) reported in the literature [17]. In the present investigation, milk fatty acid profile was improved for better human health by feeding linseed products (linseed and Ca-salt of linseed oil), as shown by higher concentrations of PUFA including CLA which is recognized as being beneficial to reduce the incidence of cancer, cardiovascular diseases, hypertension, and arthritis and an improvement of visual acuity.

3.3. Economics of feeding

Table 3.7 Economics of feeding for milk production and milk quality parameters in cows fed different sources of fat

Attribute	T_1	T_2	T_3	T_4
Cost of feeding concentrate mixture (USD/d)	1.98±0.013^{d}	2.05±0.020^{c}	2.85±0.022^{a}	2.267±0.017^{b}
Cost of feeding roughage (Rs/d)	0.51±0.00	0.51±0.00	0.48±0.00	0.491±0.00
Total feeding cost (USD /d)	2.49±0.013^{d}	2.56±0.020^{c}	3.33±0.022^{a}	2.758±0.017^{b}
Total milk production (kg/d)	12.62	13.38	12.40	12.91
Feeding cost/ kg milk production (USD)	0.20±0.012^{b}	0.19±0.012^{b}	0.27±0.016^{a}	0.214±0.015^{b}
Total fat yield (g/d)	0.48	0.50	0.47	0.48
Feeding cost/ kg fat yield (USD)	5.18±0.324^{b}	5.12±0.339^{b}	7.08±0.454^{a}	5.747±0.434^{b}
PUFA yield (g/100g fat)	3.33^{d}	4.96^{b}	7.90^{a}	3.88^{c}
PUFA yield (g/d)	15.98^{d}	24.80^{b}	37.13^{a}	18.62^{c}
Feeding cost/g PUFA (USD)	0.16±0.011^{a}	0.10±0.008^{b}	0.09±0.006^{b}	0.148±0.013^{a}
Total CLA yield (mg/g fat)	8.47^{d}	14.82^{a}	11.19^{b}	9.46^{c}
Total CLA yield (g/d)	4.07^{d}	7.41^{a}	5.26^{b}	4.54^{c}
Feeding cost/ g CLA (USD)	0.61±0.046^{a}	0.35±0.027^{b}	0.63±0.043^{a}	0.608±0.048^{a}

Values bearing different superscripts in a raw differ significantly ($P < 0.05$).

The feeding cost was calculated by taking into account the cost of feed ingredients i.e. roughage and concentrate including linseed oil and its processing costs. Total feeding cost differed significantly ($P < 0.05$) among the four treatments. The total feeding cost was highest (USD 3.33/d) in T_3, followed by (USD 2.76/d) T_4, (USD 2.56/d) T_2, and (USD 2.49/d) T_1. Because of the higher cost of linseed oil total feeding cost was higher when the diets were supplemented with linseed products and the processing of linseed oil for preparing Ca-salt increased the costs further in T_3. The feeding cost per g PUFA production was significantly higher in T_1 and T_4 than T_2 and T_3. Feeding cost per g PUFA production reduced by 42% in cows fed bypass fat (Ca-salt of linseed oil) than the control group (T_1).The feeding cost per g CLA was significantly ($P < 0.05$) lower in T_2 than T_1, T_3 and T_4. The feeding cost per g CLA production was reduced by 44% in cows fed linseed than the control group. These finding, suggested that though the linseed supplementation increased the total feeding cost for milk production, but in terms of the quality of milk with respect to the CLA content, ground linseed proved to be the best while the cost of PUFA production was also reduced upon linseed supplementation and Ca-salt of linseed supplementation as well.

4. CONCLUSION

Feeding linseed products to dairy cows reduced medium chain saturated fatty acids and increased the level of long chain fatty acids in milk fat while maintaining milk yield and milk fat content. In general, milk fatty acid profile is improved for better human health by feeding linseed products (ground linseed and Ca-salt of linseed oil), as shown by higher concentrations of PUFA including CLA recognized as being beneficial to reduce the incidence of cancer, cardiovascular diseases and stimulate the immune system.

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