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Effects of Soybean Oil or Rumen Protected Conjugated Linoleic Acid Supplementation on Accumulation of Conjugated Linoleic Acid in Dairy Cows' Milk

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ABSTRACT : The effects of feeding soybean oil (SBO) or rumen protected conjugated linoleic acid (RP-CLA) on CLA accumulation in milk, and performance of lactating dairy cows were studied. Twenty four Holstein Friesian crossbred lactating dairy cows, averaging 126±45 days in milk, 15.6±2.43 kg of milk and 452±51 kg body weight were stratified randomly and assigned in a randomized complete block design (RCBD) to three treatments of 8 cows each. The treatments were control, 150 g of SBO and 150 g of RP-CLA supplementation. Performance parameters showed that DM intake, NE_{LP} intake and body weight change were similar across treatments, while CP intake was decreased by SBO and RP-CLA supplementation. Milk yield and milk composition were not significantly different among treatments, except for milk fat percentage and fat yield which were significantly decreased by 27% ($p<0.05$) and by 28% ($p<0.01$), respectively, by RP-CLA supplements compared with control treatment. Feeding RP-CLA reduced 3.5% FCM compared with the other treatments ($p<0.003$). Both SBO and RP-CLA supplementation reduced <C16:0 fatty acids but increased ≥C18:0 and CLA concentration in milk fat. (**Key Words** : Conjugated Linoleic Acid, Rumen-protected Conjugated Linoleic Acid, Soybean Oil, Fatty Acid, Milk Production and Composition)

INTRODUCTION

Conjugated linoleic acids (CLA), a mixture of positional and geometric isomers of octadecadienoic acid with conjugated double bonds, have a range of potent health effects, including suppression of carcinogenesis (Ip et al., 1999; Belury, 2002; Corl et al., 2003), antiobese effect (Park et al., 1997), modulation of the immune system (Cook et al., 1993), reduction in atherosclerosis (Nicolosi et al., 1997), diabetes (Houseknecht et al., 1998) and decreased body fat mass in humans (Blankson et al., 2000; Gaullier et al., 2005). Animal products from ruminants, particularly dairy products are the main dietary source of CLA. It is accepted that CLA are intermediates in the biohydrogenation of linoleic acid, which originate from the incomplete biohydrogenation of unsaturated fat by rumen function (Bauman et al., 1999). However, research work has found that cows can also synthesis CLA from *trans*-11 octadecadienoic acid, another intermediate in the rumen biohydrogenation process, by Δ^9 desaturase in tissue (Griinari et al., 1998; Corl et al., 2001).

Plant oils and oil seeds rich in linoleic acid have been shown to increase CLA in milk fat of cows (Kelly et al., 1998; Leonardi et al., 2005; Loor et al., 2005; Zheng et al., 2005; Shingfield et al., 2006; Bu et al., 2007; Chantaprasarn and Wanapat, 2008) and rumen-protected (RP)-CLA also showed a similar trend (Perfield et al., 2002; Perfield et al., 2004; Piperova et al., 2004; Castaneda-Cutierrez et al., 2005). Comparison between oils and RP-CLA supplementation in dairy cows is very limited. The aim of the present study was to compare soybean oil and RP-CLA supplementation on CLA accumulation in milk fat and performance of lactating dairy cows.

MATERIALS AND METHODS

Animals and treatments

Twenty four Holstein Friesian crossbred (>87.5% Holstein Friesian) lactating dairy cows, averaging 126±45 days in milk, 15.6±2.43 kg of milk and 452±51 kg body weight (BW), were blocked by milking days first and then

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milk yield and body weight into two groups of 12 cows. Cows within each group were further randomly assigned to three treatments of 8 cows each. The first group (control) received approximately 10 kg of concentrate. The second group was fed the same basal diet as the control group and supplemented with 150 g/d of soybean oil (SBO). The third group was fed the same diet as the control group and supplemented with 150 g/d of rumen-protected conjugated linoleic acid (RP-CLA; BASF (Thai) Co., Ltd.). The RP-CLA contained two CLA isomers in equal proportion: *cis*-9, *trans*-11 and *trans*-10, *cis*-12 CLA. The fatty acid profiles of concentrate, SBO and RP-CLA are presented in Table 2. SBO or RP-CLA was thoroughly mixed with concentrate prior to feeding. All cows also received *ad libitum* grass silage, had free access to clean water and were individually housed in a free-stall unit and individually fed according to treatments. The experiment lasted for 10 weeks with the first 2 weeks as the adjustment period, followed by 8 weeks of measurement period.

Measurements, sample collection, and chemical analysis

Feeds offered and residues left after eating of individual cows were weighed for two consecutive days of each period and samples were taken and dried at 60°C for 48 hour. At the end of the experimental period, feed samples were composited and subsamples were taken for further chemical analysis. Samples were ground through a 1 mm screen and subjected to proximate analysis. The crude protein content was determined by Kjeldahl analysis (AOAC, 1998). Ether extract was determined using petroleum ether in a Soxtec System (AOAC, 1998). Neutral detergent fiber and acid detergent fiber were determined using the method described by Van Soest et al. (1991), adapted for Fiber Analyzer. Chemical analysis was expressed on the basis of the final DM.

Cows were milked twice daily at 05.00 and 15.00 h and milk yields were recorded for each cow. Samples of milk (evening+morning) were collected at each milking for two consecutive days weekly and stored at 4°C with a preservative (bronopol tablet; D&F Control System, San Ramon, CA) until analyzed for fat, protein, lactose and solid-not-fat contents using a Milko-Scan S50 analyzer (Tecator, Denmark). All cows were weighed at the start and end of the experiment.

Milk fatty acid analysis

Milk samples were collected from individual cows on day 0, 10, 20 and 30 of the experiment. For fatty acid analysis, an aliquot of milk (~30 ml) was centrifuged at 17,800×3 g for 30 min at 8°C and then 300-350 mg of fat cake was removed. Lipid extraction followed the procedures described by Hara and Radin (1978), using a volume of 18 ml of hexane and isopropanol (3:2, vol/vol)/g of fat cake. After vortexing, a sodium sulfate solution (6.7% $NaSO_4$ in distilled H_2O) was added at 12 ml/g of fat cake. The hexane layer was transferred to a tube containing 1 g of $NaSO_4$ and after 30 min, the hexane layer was removed and stored at -20°C until methylation.

Fatty acid methyl esters (FAME) were prepared by the procedure described by Ostrowska et al. (2000). After methylation was completed, 10 ml of deionized water was added. The solution was transferred to a 40-ml centrifuge tube and 5 ml of hexane was added for FAME extraction. The solution was centrifuged at 2,000 g at 10°C for 20 min, then the hexane layer was dried over sodium sulfate and was placed into a vial for analysis by gas chromatography (GC) (Hewlett Packard GC system HP6890 A; Hewlett Packard, Avondale, PA) using a 100 m×0.25 mm fused silica capillary column (SP2560, Supelco Inc. Bellefonte, PA, USA). Injector and detector temperatures were 240°C. The column temperature was kept at 70°C for 4 min, then increased at 13°C/min to 175°C which was held for 27 min, then increased at 4°C/min to 215°C and held for 31 min.

Statistical analysis

Measurements of intake, milk production and milk fatty acid composition were analyzed by ANOVA for a randomized complete block design using the Statistical Analysis System (SAS, 1996). Differences between treatment means were statistically compared using Duncan's New Multiple Range Test (Steel and Torrie, 1980).

RESULTS AND DISCUSSION

Chemical and fatty acid compositions of feeds used in the experiment are presented in Table 1 and 2, respectively. The ether extract (EE) content and energy values of the control diet were lower than the SBO and RP-CLA supplementation diets. The control diet had higher C12:0 and C14:0 than other diets, while the SBO supplemental diet was rich in C18:2 and C18:3. The RP-CLA diet contained total CLA at 18.5% of total fatty acids. Dry matter (DM) and net energy for lactation (NE_{LP}) intakes of the experimental cows were similar ($p>0.05$), however, crude protein (CP) intake was significantly higher ($p<0.001$) in cows which received the control diet (Table 3). Similar results were previously obtained with diets supplemented with oils (Dhiman et al., 2000; Loor et al., 2005; Zheng et al., 2005) and with RP-CLA (Moore et al., 2004; Piperova et al., 2004; Perfield et al., 2004; Castaneda-Cutierrez et al., 2005; De Veth et al., 2005). The present study showed no significant difference in dry matter intake (DMI) due to SBO and RP-CLA addition. High oil addition in the diet has been showed to limit DMI in previous studies (Gagliostro and Chilliard, 1991; Litherland et al., 2005; Shingfield et al., 2006). There were no significant

Table 1. Chemical composition of the diets

Item	Treatment (% of DM)[1]			Grass silage
	Control	SBO	RP-CLA	
Dry matter	88.03	90.18	89.19	31.11
Ash	7.58	7.56	8.21	8.17
Crude protein	23.57	22.16	22.00	3.95
Ether extract	2.86	4.59	4.04	1.65
Crude fiber	12.53	12.52	12.38	39.88
Neutral detergent fiber	46.98	48.54	42.17	81.41
Acid detergent fiber	22.77	21.42	21.89	52.61
Acid detergent lignin	7.27	5.57	6.56	7.95
Neutral detergent insoluble N	1.16	1.04	0.96	0.40
Acid detergent insoluble N	0.74	0.74	0.77	0.60
TDN_{1X} (%)[2]	59.23	63.48	63.12	45.56
DE_{1X} (Mcal/kg DM)[3]	2.77	2.93	2.92	1.93
DE_P (Mcal/kg DM)[4]	2.74	2.83	2.82	2.11
ME_P (Mcal/kg DM)[5]	2.31	2.41	2.40	1.68
NE_{LP} (Mcal/kg DM)[6]	1.44	1.51	1.50	0.99

[1] SBO = 150 g of soybean oil/cow/d; RP-CLA = 150 g of rumen protected conjugated linoleic acid/cow/d.

[2] TDN_{1X} (%) = tdNFC+tdCP+(tdFA×2.25)+tdNDF-7 (NRC, 2001).

[3] DE_{1X} (Mcal/kg) = ((tdNFC/100)×4.2)+((tdNDF/100)×4.2)+((tdCP/100)×5.6)+((FA/100)×9.4)-0.3

[4] DE_P (Mcal/kg DM) = DE_{1X}×Discount (NRC, 2001).

[5] ME_p = (1.01×(DE_p)-0.45)+(0.0046×(EE-3)) (NRC, 2001).

[6] NE_{LP} = ((0.703×ME_p (Mcal/kg)]-0.19)+(((0.097×ME_p+0.19)/97)×(EE-3)) (NRC, 2001).

Ingredient composition of concentrate (control diet): 26% cassava chip, 10% rice bran, 16% oil palm meal, 13% coconut meal, 12% soybean meal, 10% sunflower meal, 8% molasses, 2% urea, 2.5% mineral mix and 0.5% premix.

differences in milk, protein, lactose and solid-not-fat (SNF) yields, however, 3.5% fat corrected milk (FCM), fat yield and total solid yield were significantly reduced when RP-CLA was supplemented (Table 4). Milk compositions were unaffected by SBO and RP-CLA addition except for fat percentage, which was significantly ($p<0.05$) decreased when RP-CLA was added. The reduction in 3.5% FCM yield and total solid yield reflected the depression in milk fat percentage and yield. Many previous studies showed no difference in milk yield (Moore et al., 2004; Piperova et al., 2004; Perfield et al., 2004; Castaneda-Cutierrez et al., 2005; De Veth et al., 2005). Zheng et al. (2005) also reported that milk yield was unaffected when cows received oils from cottonseed, soybean and corn. However, Leonardi et al. (2005) found that milk yield was significantly ($p<0.005$) increased by 5.8% with 1.5% corn oil supplementation.

Table 2. Fatty acid compositions of concentrate, SBO, RP-CLA and grass silage

Item	Fatty acid (% of total fatty acid)[1]			Grass silage
	Control	SBO	RP-CLA	
C 12:0	25.9	ND	ND	5.3
C 14:0	9.4	ND	ND	3.5
C 16:0	13.0	0.5	10.6	32.3
C 18:0	2.9	5.0	55.4	10.7
C 18:1	21.5	ND	8.2	6.4
C 18:2	22.8	54.4	1.7	23.2
C 18:3	ND	34.5	ND	ND
C 20:1	ND	ND	ND	18.6
Other[2]	4.5	5.7	5.6	ND
c-9, t-11CLA[3]	ND	ND	9.3	ND
t-10, c-12 CLA[3]	ND	ND	9.2	ND
Total CLA[4]	ND	ND	18.5	ND

[1] SBO = 150 g of soybean oil/cow/d; RP-CLA = 150 g of rumen protected conjugated linoleic acid/cow/d.

[2] Other = Sum of C6:0, C8:0, C10:0, C16:1, C17:1, C20:1, C20:2, C22:0, C20:3n6, C22:1n9+C20:3n3, C23:0, C20:5n3, C24:1.

[3] CLA = Conjugated linoleic acid.

[4] Total CLA = Sum of *cis*-9,*trans*-11 CLA; *trans* -10, *cis* -12 CLA.

In the present study, milk fat percentage and yield were significantly decreased by 27% ($p<0.05$) and 28% ($p<0.01$), respectively, due to RP-CLA supplementation. Similar results were previously reported (Moore et al., 2004; Piperova et al., 2004; Perfield et al., 2004; Castaneda-Cutierrez et al., 2005; De Veth et al., 2005).The *trans*-10, *cis*-12 CLA isomer inhibited milk fat synthesis in dairy cows (Baumgard et al., 2000; 2001), whereas the *cis*-9, *trans*-11 CLA had no effect. The mechanism by which *trans*-10, *cis*-12 CLA alters lipid mechanism involves many aspects of milk fat synthesis. Specifically, this CLA isomer dramatically reduced lipogenesis by the mammary gland (rates of acetate incorporation into fatty acids) and decreased the expression of genes encoding enzyme (mRNA abundance of acetyl CoA carboxylase) involved in the uptake and transport of circulating fatty acids, *de novo* fatty acid synthesis, desaturation of fatty acids and formation of triglycerides, as found in mice (Baumgard et al., 2000; 2001; Piperova et al., 2000). Lin et al. (2004) also indicated that reduced lipogenesis in the mammary gland of lactating mice was caused by reducing acetyl CoA carboxylase activity and mRNA abundance of acetyl CoA

Table 3. Effect of soybean oil and rumen protected conjugated linoleic acid on nutrient intake of Crossbred Holstein Friesian dairy cows

Item	Treatment[1]			SEM	p-value
	Control	SBO	RP-CLA		
DM intake (kg/d)					
Concentrate	8.91	8.91	8.91	-	-
Grass silage	5.07	4.49	4.52	0.39	0.4143
Total	13.98	13.40	13.44	0.39	0.4153
CP intake (g/d)					
Concentrate	2,100	1,975	1,961	-	-
Grass silage	211	195	196	13.13	0.4194
Total	2,311[a]	2,170[b]	2,157[b]	13.12	0.0001
Fat intake (g/d)					
Concentrate	255[c]	409[a]	360[b]	4.52	0.0001
Grass silage	84	74	75	4.51	0.4368
Total	339[c]	483[a]	435[b]	4.52	0.0001
NE_{LP} intake (Mcal/d)					
Concentrate	12.83	13.45	13.37	-	-
Grass silage	5.02	4.44	4.48	0.38	0.2715
Total	17.85	17.89	17.84	0.38	0.7227

[a, b] Means within row with different superscripts differ.
[1] SBO = 150 g of soybean oil/cow/d; RP-CLA = 150 g of rumen protected conjugated linoleic acid/cow/d.

carboxylase, the critical enzyme in *de novo* fatty acid synthesis, and also inhibited mammary desaturation by reducing mammary stearoyl-CoA desaturase activity and mRNA abundance.

Fatty acids ≤C14:0 (C4:0, C6:0, C8:0, C10:0, C12:0 and C14:0) in milk fat were significantly decreased ($p<0.01$) by supplementation with SBO and RP-CLA (Table 5). However, C16:0 was not altered across treatments. Fatty acids, C18:0, C18:1 and C18:2 in milk fat were significantly increased ($p<0.001$) by SBO supplementation in the diet compared with respective values for the control treatment. Milk fat content of short- and medium-chain fatty acids was reduced ($p<0.001$) when cows received both SBO and RP-CLA supplementation compared with the control treatment. This resulted in decreasing ($p<0.001$) *de novo* (≤C16:0) fatty acids and an increase ($p<0.001$) in preformed (>C16:1) fatty acids in milk fat. Similar patterns of shift in these fatty acids were also observed with oils (Dhiman et al., 2000; Kay et al., 2004; Zheng et al., 2005; Shingfield et al., 2006; Bu et al., 2007) and with RP-CLA (Chouinard et al., 1999; Perfield et al., 2002; Viswanadha et al., 2003; Moore et al., 2004; Perfield et al., 2004).

In the present study, addition of dietary SBO significantly increased ($p<0.001$) *cis*-9, *trans*-11 CLA

Table 4. Milk yield and milk composition of crossbred Holstein Friesian dairy cows supplemented with soybean oil and rumen-protected conjugated linoleic acid

Item	Treatment[1]			SEM	p-value
	Control	SBO	RP-CLA		
Milk yield (kg/d)	15.16	16.05	14.46	1.29	0.2478
3.5% FCM yield (kg/d)	14.87[a]	16.07[a]	12.02[b]	0.72	0.0003
Milk composition yield (g/d)					
Fat yield	529[a]	581[a]	369[b]	26	0.0001
Protein yield	399	441	418	23	0.1570
Lactose yield	628	674	630	47	0.2908
SNF	1,204	1,306	1,180	87	0.1430
Total solid	1,733[ab]	1,884[a]	1,562[b]	91	0.0084
Milk composition (%)					
Fat	3.49[a]	3.62[a]	2.55[b]	0.34	0.0481
Protein	2.63	2.75	2.89	0.15	0.3677
Lactose	4.14	4.2	4.36	0.13	0.4554
SNF	7.94	8.14	8.16	0.21	0.7573
Total solid	11.43	11.74	10.8	0.54	0.4288
Body weight (kg)					
Pre-experiment	452	454	450	25	0.3265
Post-experiment	448	428	449	21	0.1903
Body weight change (g/d)	-110	-857	-24	835	0.8760

[a, b] Means within row with different superscripts differ.
[1] SBO = Soybean oil 150 g/d; RP-CLA = Rumen protected conjugated linoleic acid 150 g/d.

Table 5. Fatty acid composition of milk fat from cows fed soybean oil and RP-CLA

Items	Treatments			SEM	p-value
	Control	SBO	RP-CLA		
	---------- mg/g fat----------				
C4:0	12.95[a]	10.81[b]	9.09[c]	0.52	0.0002
C6:0	9.72[a]	7.86[b]	4.28[c]	0.54	0.0001
C8:0	6.26[a]	4.70[b]	2.46[c]	0.41	0.0001
C10:0	13.69[a]	10.23[b]	5.98[c]	1.03	0.0002
C11:0	1.93[a]	1.24[b]	0.59[c]	0.14	0.0001
C12:0	47.74[a]	37.48[b]	29.63[c]	2.13	0.0001
C13:0	1.63[a]	1.11[b]	0.84[b]	0.11	0.0003
C14:0	80.66[a]	68.68[b]	56.64[c]	3.46	0.0004
C14:1	11.41[a]	8.10[b]	7.02[b]	0.68	0.0005
C15:0	5.22[a]	4.45[ab]	3.84[b]	0.29	0.0103
C16:0	197.67[a]	177.14[b]	149.51[b]	8.80	0.0036
C16: 1	20.45[a]	16.62[b]	14.23[b]	0.98	0.0009
C17: 1	1.73[a]	1.70[a]	1.22[b]	0.12	0.0102
C18:0	51.21[b]	69.40[a]	57.60[b]	3.79	0.0078
C18:1	212.88[ab]	231.48[a]	189.97[b]	8.46	0.0089
C18:2	15.58[b]	19.99[a]	12.12[b]	1.30	0.0014
C18:3	1.21[a]	1.38[a]	0.64[b]	0.12	0.0009
C20:0	0.82	0.98	0.83	0.05	0.0609
Others[2]	3.76[a]	3.92[a]	2.03[b]	0.18	0.0001
cis-9,*trans*-11 CLA[3]	5.57[b]	9.15[a]	5.22[b]	0.53	0.0001
trans -10, *cis* -12 CLA[3]	0.016[a]	0.044[b]	0.753[a]	0.03	0.0001
trans -9, *trans* -11 CLA[3]	0.22[b]	0.58[a]	0.54[a]	0.06	0.0010
Total CLA[4]	5.81[b]	9.77[a]	6.51[b]	0.53	0.0001

[a, b, c] Means within row with different superscripts differ.

[1] SBO = 150 g of soybean oil/cow/d; RP-CLA = 150 g of rumen protected conjugated linoleic acid/cow/d.

[2] Other = (Sum of C20:1,C20:2, C22:0, C20:3n6,C22:1n9+C20:3n3, C20:4n6, C20:5n3).

[3] CLA = Conjugated linoleic acid (*cis-9, trans-11 octadecadienoic acid*).

[4] Total CLA = (Sum of *cis*-9,*trans*-11 CLA; *trans* -10, *cis* -12 CLA; *trans* -9, *trans* -11 CLA).

concentration by 65% and 21% in milk fat when compared to control and RP-CLA treatments, respectively. RP-CLA significantly increased *trans*-10, *cis*-12 CLA concentration compared with control and SBO treatments. Increase in *trans*-10, *cis*-12 CLA concentration in milk fat seemed to be related with milk fat depression (Table 4). However, total CLA concentration was significantly increased ($p<0.001$) by SBO and RP-CLA addition compared with that of the control treatment.

Increase in *cis*-9, *trans*-11 CLA, in the present study, was due to a high linoleic acid and linolenic acid content in SBO (Table 1). The *cis*-9, *trans*-11 CLA in milk fat was probably formed by incomplete biohydrogenation of dietary linoleic acid in rumen and from *trans*-11 C18:1 vaccenic acids (the intermediate in biohydrogenation of linoleic acid, linolenic acid and oleic acid) which can endogenously synthesize *cis*-9, *trans*-11 CLA via Δ^9 desaturase in the mammary gland (Corl et al., 2001).

Similar results were previously reported; for example, Dhiman et al. (2000) found that feeding lipid sources of 2.0% and 4.0% SBO rich in linoleic acid and linolenic acid as free oils increased CLA concentration in milk fat by 77% and 188% respectively. Leonardi et al. (2005) showed that cows fed fish oil and sunflower oil increased total CLA concentration by 42 and 594%, respectively. Shingfield et al. (2005) summarized that concentration of *cis*-9, *trans*-11 CLA in milk fat was increased by dietary supplementation with fish and sunflower oil in the diet. Cows fed 5% SBO increased *cis*-9, *trans*-11 CLA by 97% (Zheng et al., 2005), while an increase of 318% was also found when fed with 4% SBO (Bu et al., 2007).

Previous studies with RP-CLA supplementation by Chouinard et al. (1999) reported that abomasal infusion of 50, 100 and 150 g/d RP-CLA increased ($p<0.01$) CLA content of milk fat by 246, 585 and 835%, respectively. Similarly, Perfield et al. (2004) found that the concentrations of *cis*-9, *trans*-11 CLA and *trans*-10, *cis*-12 CLA were increased by amide protected CLA (AP-CLA) and lipid encapsulated CLA (LE-CLA) supplements. Addition of calcium salts of CLA (Ca-CLA) also increased *cis*-9, *trans*-11 CLA and *trans*-10, *cis*-12 CLA concentration in milk fat (Giesy et al., 2002). Concentrations of total CLA in milk fat were linearly increased by dietary RP-CLA supplementation (Moore et al., 2004), while Viswanandha et al. (2003) showed that the proportion of *trans*-10, *cis*-12 CLA was linearly increased ($p<0.05$) by dose of CLA, but *cis*-9, *trans*-11 CLA concentration was not different, which was similar to the observation of Bernal-Santos et al. (2004).

Moreover, Piperova et al. (2004) reported that the proportion of total CLA and *trans*-10, *cis*-12 CLA was increased ($p<0.01$) by 60 and 164% respectively by Ca-CLA treatment, while *cis*-9, *trans*-11 CLA concentration in milk fat was decreased ($p<0.01$) by 12%. De Veth et al. (2005) showed that Ca-CLA and formaldehyde-protected CLA (FP-CLA) supplementation in the diet increased *trans*-10, *cis*-12 CLA concentration in milk fat, which is similar to the results of Castaneda-Cutierrez et al. (2005) who reported that the proportion of *cis*-9, *trans*-11 CLA and *trans*-10, *cis*-12 CLA was increased by 12% and 1,425% respectively by Ca-CLA treatment. Variation in results reported can be attributed to differences in basal diets containing different proportion of linoleic acid and linolenic acid and differences in the proportion of CLA isomers in the rumen-protected CLA supplement.

CONCLUSION

It can be clearly concluded from the present study that both SBO and RP-CLA supplementation in the diet increased CLA and ≥C18:0, but reduced <C16:0 fatty acids in milk fat, whereas these supplements had no effect on DMI and milk production of lactating dairy cows. In addition, RP-CLA reduced 3.5% FCM, fat yield, fat percentage and total solid percentage. It is suggested that SBO supplementation in the diet is superior to RP-CLA in accumulation of CLA in dairy cows' milk.

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