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Influence of Feeding Soybean Oil on Conjugated Linoleic Acid Content in Beef

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Forty-eight steers were used to study the influence of feeding soybean oil (SO) on the conjugated linoleic acid (CLA) content of beef. Steers were fed either a control diet containing 954 g/kg of dry matter (DM) corn-based concentrate (CTL) or a control diet supplemented with SO at 20 (SO2) or 40 g/kg (SO4) of diet DM for 105 days. Adipose tissue samples were collected from the M. longissimus dorsi (LD) and from the M. semitendinosus (ST) on days 0 and 63 of the experiment. Adipose and muscle tissue samples were collected from the LD and ST immediately after slaughter. Feeding 40 g/kg of DM as SO increased the proportions of *trans*-$C_{18:1}$ in beef lipid as compared to CTL and SO2 treatments. The $C_{18:2}$ *cis*-9, *trans*-11 isomer of CLA as a proportion of total fat was not different in adipose and muscle across treatments. Supplementing SO increased $C_{18:2}$ *trans*-10, *cis*-12 CLA in adipose tissue of the LD. Supplementing high-grain finishing diets with SO is not an effective strategy to enhance the $C_{18:2}$ *cis*-9, *trans*-11 isomer of CLA in beef.

KEYWORDS: Beef; conjugated linoleic acid; soybean oil; fat; fatty acid

INTRODUCTION

Conjugated linoleic acid (CLA) is a group of fatty acid isomers that occur naturally in many foods. However, the principal dietary sources are milk, meat, and other foods derived from ruminant animals (*1*). The major isomer of CLA in milk and meat is $C_{18:2}$ *cis*-9, *trans*-11 (*1*−*3*).

Conjugated linoleic acid has been shown to have anticarcinogenic, antioxidative, and antidiabetic effects in animal models (*4*−*6*). If these potential health effects of CLA are found to occur in humans, an increase in the content of CLA in beef would enhance its nutritive and therapeutic values.

Feeding safflower oil, linseed oil, fish oil, or full-fat extruded soybean meal has increased the CLA content in lamb and beef (*7*−*9*). However, feeding soybean oil at 50 g/kg of diet dry matter (DM) (*10*) had no effect on $C_{18:2}$ *cis*-9, *trans*-11, but did increase the proportion of the $C_{18:2}$ *trans*-10, *cis*-12 in beef. Not only is CLA a product of the ruminal biohydrogenation process, but it has been shown that $C_{18:2}$ *cis*-9, *trans*-11 also originates from endogenous synthesis in tissues from $C_{18:1}$ *trans*-11 via the enzyme Δ^9-desaturase (*11*, *12*).

Nutritional and management factors that influence the CLA content of beef have not been studied extensively in growing and finishing cattle on high-grain diets. Our hypothesis is that providing feed sources rich in linoleic acid at <50 g/kg of diet DM (*10*) will provide substrate for lipid biohydrogenation without negatively affecting rumen fermentation. An increased supply of substrate and incomplete lipid biohydrogenation in the rumen will result in increased production of CLA and *trans*-$C_{18:1}$ fatty acid and will enhance the CLA content of beef. The objective of the present study was to examine the effect of feeding soybean oil at 20 and 40 g/kg of dietary DM to beef cattle during the finishing phase on the CLA content of beef.

MATERIALS AND METHODS

Animals and Treatments. Forty-eight beef steers weighing an average of 398 kg (range = 328−460 kg) were blocked into groups of three according to body weight (BW). Animals were then randomly assigned to one of three treatments. The animals were of mixed breed origin [Angus cross, Red Angus cross, Black Baldy (Angus × Hereford cross), Limousin cross, Hereford cross, and Salers crossbreeds]. Animals originated from two producers. An attempt was made to select 48 healthy animals with similar physical condition of a total of 60 animals. Because of extensive crossbreeding, it was not possible to block the animals according to breed. To the authors' knowledge there are no studies in the literature supporting differences in CLA content of different pure breeds. The age of the animals ranged from 9 to 11 months at the start of the experiment.

The experiment was conducted at the Johnson Research Facility (Parma, ID) from July to October. Animal care and procedures were approved and conducted under established standards of the Utah State University Institutional Animal Care and Use Committee. The animals were housed in three separate outdoor pens. The pens were 7.6 m × 21.4 m. The duration of the entire experiment was 105 days. All animals stayed healthy during the experiment.

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The animals were fed either a corn-based basal diet (CTL) or a corn-based diet supplemented with soybean oil (SO) at 20 (SO2) or 40 g/kg of diet (SO4) on a DM basis. The SO replaced part of the grain in the control diet. The oil was mixed with small batches of grain and kept in a cold room (4 °C). Animals were group-fed. Diets were fed as a total mixed ration daily. Animals were gradually acclimated to full feed using a "step-up" grain feeding program over a period of 18 days prior to the beginning of the experiment. On day 1 of the experiment, animals were placed on experimental diets containing 46 g of forage and 954 g of concentrate/kg of DM. Animals were given ad libitum access to the feed. Daily feed offered and feed refused were recorded. Daily feed intake of individual animals was calculated by subtracting the weekly mean of feed refused from the weekly mean of feed offered divided by the number of animals in each pen.

Subcutaneous adipose tissue biopsies adjacent to the longissimus dorsi (LD) and semitendinosus (ST) were collected on days 0 and 63 of the experiment. Attempts were made on day 63 to collect tissue samples without contamination within 2.5 cm from the site at which they were collected on day 0. To collect adipose tissue biopsies, the steers were anesthetized at incision sites and tissue samples were surgically removed from the LD and ST on the right side of each animal. Samples were stored in the freezer until further analysis could be conducted.

Animals were weighed individually on two consecutive days on days 0, 63, and 105 of the experiment. Average body weights on day 105 were 602^{b}, 624^{a}, and $595^{b} \pm 7$ kg ($P = 0.01$) in CTL, SO2, and SO4 treatments, respectively. The average BW gains during the experiment (BW on day 105 minus BW on day 0) were 203, 226, and 201 ± 7 kg ($P = 0.02$) in CTL, SO2, and SO4 treatments, respectively. Animals were slaughtered on day 105 of the experiment at the Iowa Beef Processors slaughter facility (Boise, ID). Adipose tissue and muscle samples were dissected from outside the LD and ST on the left side of each carcass immediately after placement in the cold room. Tissue samples were stored in 20-mL glass scintillation vials at −20 °C until further analysis.

Feed Analyses. Representative samples of individual feed ingredients were collected monthly. The DM contents of the individual ingredients were determined by oven-drying at 60 °C for 48 h. Dried feed samples were ground through a Wiley mill using a 1-mm screen (Arthur H. Thomas, Philadelphia, PA) and analyzed for crude protein (CP) using a Leco nitrogen analyzer (model CHN-1000; Leco Corp., St. Joseph, MI). The forage samples were analyzed for neutral detergent fiber (NDF) and acid detergent fiber (ADF) using the Filter Bag Technology of ANKOM (ANKOM^{200} Fiber Analyzer, ANKOM Technology Corp., Fairport, NY). Samples were further dried at 105 °C for 8 h to determine the absolute DM. Chemical analyses were expressed on the basis of this final DM. The chemical composition of the total mixed ration was calculated from the chemical composition of individual ingredients of the diet.

The net energy (NE_g) content of the diets was estimated using NRC (*13*) NE_g values for dietary ingredients. Due to substitution of grain with oil in SO2 and SO4 treatments, the energy contents of these two diets were 0.17 and 0.33 MJ/kg of diet DM higher than that of the CTL diet. The CP, NDF, and ADF contents of the diets were similar. Ingredient and chemical compositions of the diets are given in **Table 1**.

Fatty acid analysis on individual ingredients of the diet was performed using a procedure described by Sukhija and Palmquist (*14*). Methylated fat samples were analyzed for fatty acids, including CLA in a gas chromatograph (model 6890, Hewlett-Packard Co., Wilmington, DE) fitted with a flame ionization detector. Gas chromatography conditions were the same as described by Dhiman et al. (*15*). Dietary fat in excess of 70−80 g/kg of diet DM has been shown to inhibit microbial activity and reduce feed digestibility (*16*). Total fatty acids in the SO4 treatment diet were 80 g/kg of DM and were close to the presumed safe limit for inclusion of fat in ruminant diets without decreasing nutrient utilization (**Table 2**). The diets were not balanced for total fatty acid content, as the objective of the experiment was to look solely at the effects of supplementing SO to the diet on the fatty acid profile and CLA content of the final meat product.

Table 1. Ingredients and Chemical Composition of the Experimental Diets

	treatment[a]		
	CTL	SO2	SO4
ingredient (g/kg of DM)			
chopped alfalfa hay	46	46	46
ensiled high moisture ear corn	352	352	352
dry shelled corn	522	496	470
canola meal	36	42	48
Rumax[b]	44	44	44
soybean oil		20	40
chemical composition			
dry matter	810	794	777
net $energy_G$[c] (MJ/kg of DM)	4.27	4.44	4.60
crude protein	110	110	110
neutral detergent fiber	594	591	575
acid detergent fiber	88.1	89.8	89.3

[a] Animals were fed a basal diet containing 954 g/kg corn-based concentrate (CTL), or part of the concentrate in the basal diet was replaced by 20 g/kg soybean oil (SO2) or 40 g/kg soybean oil (SO4) on dry matter basis. [b] Rumax is a liquid supplement containing Monensin sodium at 441 g/metric ton of supplement (Rumensin is registered trademark of Elanco Corp., Lilly, Indianapolis, IN), Tylosin phosphate at 1.21 g/metric ton of supplement (Tylan is a registered trademark of Elanco Corp., Lilly, Indianapolis, IN), urea, ammonium polyphosphate, ammonium sulfate, minerals, and vitamins in beet soluble. The liquid supplement was manufactured by Agri-Beef Co., Animal Feeds Division (Nampa, ID). Guaranteed analysis (per kg): minimum CP = 400 g (this includes not more than 378.7 g of equivalent nonprotein nitrogen), salt as NaCl = 90 g, calcium = 120 g, phosphorus = 3 g, vitamin A = 100 000 IU, vitamin D_3 = 10 000 IU, vitamin E = 100 IU, selenium = 3.3 mg. [c] Estimated using NRC (1996) NE_G values for dietary ingredients.

Beef Tissue Fatty Acid Analyses. Lipid extraction and washing of the extract was conducted according to the method of Folch et al. (*17*) with some modifications. After homogenization, 5 mL of a 40 g/L KCl solution was added to the samples and the mixture was vortex-mixed. For the muscle tissue samples, another 5 mL of the 40 g/L KCl solution was added to precipitate the protein. Thirty milligrams of fat was derivatized to methyl esters by mixing with 5 mL of 40 mL/L HCl/MeOH (*1*). The methyl esters were extracted with 5 mL of hexane and 1 mL of distilled water. The hexane extract was washed twice with distilled water and dried over anhydrous sodium sulfate.

Gas chromatography conditions were the same as described by Dhiman et al. (*15*). Because $C_{17:0}$ is naturally present in beef, selected tissue samples were analyzed with and without heptadecanoic acid as an internal standard to ensure the accuracy and recovery. The CLA isomers reported are $C_{18:2}$ *cis*-9, *trans*-11 and $C_{18:2}$ *trans*-10, *cis*-12.

Statistical Analyses. Fatty acid data were analyzed as a randomized complete block design with repeated measures using the Proc Mixed Model procedures of SAS (*18*). Initially, a base model that included the independent variables of block, treatment, and block × treatment interaction terms was evaluated for each variable.

Treatment, sampling site (LD and ST), day, treatment × sampling site, treatment × day, sampling site × day, and treatment × sampling site × day interaction terms were evaluated within the base model. The statistical model used for analysis of the fatty acid profile of LD and ST muscle on day 105 included treatment, sampling site, and the treatment × sampling site interaction.

The data for fatty acids (*trans*-$C_{18:1}$ and $C_{18:2}$ *trans*-10, *cis*-12) that showed a significant treatment × sampling site interaction in adipose tissue were analyzed within sampling site classification using a model that included treatment, day, and treatment × day interaction. The data for *trans*-$C_{18:1}$ that showed a significant treatment × sampling site interaction in muscle on day 105 were evaluated within site using a similar model-building strategy as described previously without day as an independent variable. The effect of block was evaluated in all models as described above. Frequency of detection for the $C_{18:2}$ *trans*-10, *cis*-12 isomer was visually evaluated within each treatment group using the Proc Freq Procedure of SAS (*18*).

Table 2. Fatty Acid Composition and Total Fatty Acids of Experimental Diets

	fatty acids (wt % of total fatty acids)										total fatty acids[b]
treatment[a]	$C_{10:0}$	$C_{12:0}$	$C_{14:0}$	$C_{15:0}$	$C_{16:0}$	$C_{16:1}$	$C_{18:0}$	$C_{18:1}$	$C_{18:2}$	$C_{18:3}$	(g/kg of DM)
CTL	0.002	0.062	0.22	0.03	14.3	0.55	2.20	26.0	54.8	1.76	42
SO2	0.002	0.042	1.93	0.33	13.9	0.37	2.83	24.0	53.0	3.66	61
SO4	0.001	0.032	2.83	0.49	13.7	0.27	3.16	22.9	52.0	4.65	80

[a] Animals were fed a basal diet containing 954 g/kg corn-based concentrate (CTL), or part of the concentrate in the basal diet was replaced by 20 g/kg soybean oil (SO2) or 40 g/kg soybean oil (SO4) on a dry matter basis. [b] Sum of weight of $C_{10:0}$–$C_{18:3}$.

Table 3. Fatty Acid Composition of Adipose and Muscle Tissues from Longissimus and Semitendinosus on Days 0, 63, and 105 of the Experiment from Steers Fed Diets Containing Soybean Oil

	fatty acid[a] (wt % of total fatty acids)															
treatment[b]	$C_{12:0}$	$C_{14:0}$	$C_{15:0}$	$C_{16:0}$	$C_{16:1}$	$C_{17:0}$	$C_{17:1}$	$C_{18:0}$	*cis*-$C_{18:1}$	$C_{18:2}$	$C_{18:3}$	CLA[c]	$C_{20:3}$[d]	$C_{20:3}$[e]	SFA[f]	UFA[g]
								Adipose Tissue								
day 0																
CTL	0.07 a	3.79 b	1.04	30.8	6.40 a	1.20	1.10	12.2	38.7	1.10	0.48	0.70	ND[h]	ND	45.3	54.7
SO2	0.07 a	4.52 a	0.95	29.4	6.80 a	1.20	1.10	12.2	38.9	1.10	0.51	0.65	ND	ND	43.9	56.1
SO4	0.06 b	3.99 b	1.03	31.4	6.00 b	1.10	1.10	12.6	37.8	1.00	0.48	0.66	ND	ND	46.2	53.8
day 63																
CTL	0.06 b	3.68 b	1.35 a	28.6	4.70	2.10 a	1.60	11.0 b	40.9 a	1.40 b	0.23 b	0.50	ND	ND	43.1	56.9
SO2	0.07 a	4.10 a	1.14 b	27.5	4.90	1.80 b	1.60	11.4 ab	39.1 b	1.70 a	0.30 a	0.54	ND	ND	42.0	58.0
SO4	0.06 b	4.18 a	1.00 c	27.6	4.30	1.90 b	1.30	12.2 a	35.3 c	1.60 b	0.28 a	0.55	ND	ND	42.9	57.1
day 105																
CTL	0.05	3.68	0.74	27.3	5.30 a	1.70	1.60 a	10.9 b	42.8 a	1.50 b	0.19 c	0.45	0.020 a	0.020 a	40.7	59.3
SO2	0.06	3.99	0.79	26.4	4.90 ab	1.70	1.40 b	11.2 b	40.8 b	1.70 a	0.25 a	0.49	0.009 b	0.004 b	40.2	59.8
SO4	0.05	3.98	0.78	26.2	4.50 b	1.70	1.30 b	12.1 a	36.9 c	1.60 b	0.23 b	0.49	0.004 b	0.004 b	40.9	59.1
SEM[i]	<0.01	<0.01	0.05	0.70	0.20	<0.10	<0.10	0.30	0.50	0.10	0.01	0.02	0.002	0.002	0.60	0.60
								Muscle								
day 105																
CTL	0.03	2.88 b	1.35	26.7	4.90 a	1.60	1.50 a	9.7 b	40.0 a	6.00	0.50 b	0.23	0.52	1.84	39.4	60.8
SO2	0.04	3.18 a	1.30	26.8	4.60 a	1.40	1.30 b	10.2 b	37.6 b	6.80	0.53 b	0.29	0.41	1.31	39.8	60.2
SO4	0.03	3.27 a	1.55	26.0	3.90 b	1.60	1.20 b	11.0 a	33.4 c	7.40	0.62 a	0.31	0.48	1.58	40.2	59.9
SEM	<0.01	0.02	0.34	0.60	0.20	0.10	<0.10	0.30	0.70	0.50	0.04	0.03	0.07	0.18	0.50	0.50

[a] Means with different letters within a column, day, and tissue differ at $P < 0.05$. [b] Animals were fed a basal diet containing 954 g/kg corn-based concentrate (CTL), or part of the concentrate in the basal diet was replaced by 20 g/kg of soybean oil (SO2) or 40 g/kg of soybean oil (SO4) on a DM basis. [c] Conjugated linoleic acid ($C_{18:2}$ *cis*-9, *trans*-11). [d] $C_{20:3}$ *cis*-8,11,14. [e] $C_{20:3}$ *cis*-11,14,17. [f] Saturated fatty acids, sum of $C_{8:0}$, $C_{10:0}$, $C_{12:0}$, $C_{14:0}$, $C_{15:0}$, $C_{16:0}$, $C_{17:0}$, and $C_{18:0}$. [g] Unsaturated fatty acids, sum of $C_{16:1}$, $C_{17:1}$, *trans*-$C_{18:1}$, *cis*-$C_{18:1}$, $C_{18:2}$, $C_{18:3}$, $C_{18:2}$ *cis*-9, *trans*-11, $C_{18:2}$ *trans*-10, *cis*-12, and $C_{20:3}$. [h] Not detected. [i] Standard error of the least-squares mean.

Estimates of treatment group least-squares means are reported. Differences among treatment groups were evaluated by Tukey's *T* test on the differences of least-squares means. Significance was declared at $P < 0.05$ unless otherwise noted. Significance at $P = 0.01$ or less is mentioned as $P < 0.01$ to simplify the tables and text.

RESULTS AND DISCUSSION

Daily average feed intakes were 11.0, 11.1, and 10.3 kg in CTL, SO2, and SO4 treatments, respectively. Because animals were group-fed, the feed intake data were not analyzed statistically.

Treatment × sampling site interactions were not significant for the majority of fatty acids in adipose tissue and muscle (13 of 15 fatty acids). Treatment × day interactions were significant for 8 of the total 15 fatty acids. Therefore, for presentation and interpretation, data from the two sites (LD and ST) were combined and treatment comparisons were made within measurement days (0, 63, and 105 days; **Table 3**) for tissue fatty acid proportions. The data for fatty acids that showed significant treatment × sampling site interactions within adipose tissue ($C_{18:2}$ *trans*-10, *cis*-12) and muscle (*trans*-$C_{18:1}$) are discussed separately for each site.

There were no trends in treatment differences for fatty acids $C_{12:0}$–$C_{16:0}$ (**Table 3**) except that $C_{14:0}$ content was higher in muscle from steers in the SO2 and SO4 treatments at the time of slaughter compared with those in the CTL (**Table 3**). Other differences reported but not discussed specifically in the text are likely due to differences of fatty acid composition among the three diets. Adipose tissue and muscle from steers in the SO4 had lower proportions of $C_{16:1}$ compared with CTL and SO2. The decrease in $C_{16:1}$ content is probably a reflection of its lower proportion in the diets containing SO (**Table 2**). Engle et al. (*19*) observed a similar decrease in $C_{16:1}$ content in muscle and adipose tissues and no change in $C_{16:0}$ fatty acids in LD muscle of finishing steers fed diets containing 40 g/kg SO. A decrease in $C_{17:1}$ was observed on day 105 in adipose and muscle tissues from steers in SO2 and SO4 treatments compared with steers in CTL.

Feeding 40 g/kg SO to steers for 63 and 105 days in the present study increased (10%) the proportions of $C_{18:0}$ in adipose and muscle tissues at slaughter compared with CTL and SO2 treatments (**Table 3**). Similar increases in $C_{18:0}$ of adipose tissue obtained from LD and ST were observed in steers fed high-grain diets supplemented with 19 or 39 g/kg SO through extruded full-fat soybean meal (*9*). Beaulieu et al. (*10*) observed an increase in $C_{18:0}$ of the mesenteric and perirenal adipose tissue, but not in subcutaneous adipose tissue of beef heifers fed a high-frain diet supplemented with 50 g/kg SO. In contrast, feeding 40 g/kg SO to finishing steers resulted in no change in $C_{18:0}$ of the LD muscle (*19*).

Feeding 20 and 40 g/kg SO resulted in 5 and 15% decreases in the proportions of *cis*-$C_{18:1}$ in adipose and muscle tissues

Table 4. *trans*-$C_{18:1}$ and $C_{18:2}$ *trans*-10, *cis*-12 in Adipose Tissue on Days 0, 63, and 105 and in Muscle on Day 105 of the Experiment from Longissimus and Semitendinosus of Beef Cattle Fed Diets Containing Soybean Oil

			treatment[a] (wt % of total fatty acids)									
			M. longissimus					M. semitendinosus				
tissue	fatty acid	day	CTL	SO2	SO4	SEM[b]	*P*[c]	CTL	SO2	SO4	SEM	*P*
adipose	*trans*-$C_{18:1}$	0	2.99	2.98	2.98	0.27	1.0	2.25	2.13	2.51	0.24	0.99
		63	4.24 b	7.05 b	11.12 a	0.92	<0.01	3.11 b	4.05 b	7.15 a	0.66	<0.01
		105	4.88 b	7.55 b	12.69 a	1.15	<0.01	2.81 b	4.90 b	7.52 a	0.83	0.05
muscle	*trans*-$c_{18:1}$	105	2.95 b	5.01 b	9.28 a	0.74	<0.01	1.95 c	3.39 b	6.30 a	0.38	0.02
adipose	$C_{18:2}$ *trans*-10, *cis*-12	0	ND[d]	ND	ND	–[e]	–	ND	ND	ND	–	–
		63	ND	0.019 b	0.043 a	0.008	<0.01	ND	ND	0.014	0.007	–
		105	0.009 c	0.054 b	0.091 a	0.008	<0.01	ND	0.019	0.036	0.007	0.22

[a] Animals were fed a basal diet containing 954 g/kg corn-based concentrate (CTL), or part of the concentrate in the basal diet was replaced by 20 g/kg soybean oil (SO2) or 40 g/kg soybean oil (SO4) on adry matter basis. [b] Standard error of the least-squares mean. [c] Means with different letters within a row and site differ significantly at the *P* value mentioned in the last column. [d] Not detected. [e] Value not determined.

compared with CTL, respectively, on days 63 and 105 (**Table 3**). Other researchers have reported 4−10% decreases in *cis*-$C_{18:1}$ of beef adipose tissue and muscle by feeding SO or feed sources rich in SO (*9*, *10*, *19*).

Adipose tissue from steers fed 20 g/kg SO showed higher proportions of $C_{18:2}$ compared with CTL and SO4 at days 63 and 105 (**Table 3**). The $C_{18:2}$ content of muscle was not different among treatments on day 105. Interestingly, the average proportion of $C_{18:2}$ fatty acid for all three treatments was 300% higher in muscle (6.7 vs 1.6 wt % of total fatty acids) than adipose tissue on day 105. Lower proportions of $C_{18:2}$ were also observed in adipose tissue than in muscle (2.0 vs 3.6 wt %) of lambs (*20*) and beef heifers (2.9 vs 5.0 wt %; *10*), irrespective of dietary treatment. This observation is probably due to the fact that fat in muscle tissue is more reflective of membrane structure. The larger proportion of $C_{18:3}$ in adipose tissue from steers fed SO at days 63 and 105 as compared to those of the CTL is probably a reflection of its higher supply through the diet (**Table 2**). In muscle tissue, $C_{18:3}$ was higher for steers in the SO4 as compared to either of the other two treatments.

The $C_{18:2}$ *cis*-9, *trans*-11 contents in adipose and muscle were not different among treatments on days 0, 63, and 105 (**Table 3**). The $C_{20:3}$ fatty acids were not detected in adipose tissues on days 0 and 63 of the experiment. The proportions of $C_{20:3}$ were decreased in adipose tissue from steers fed diets containing 20 or 40 g/kg SO as compared to CTL at day 105, suggesting reduced activity of the elongase enzyme in those two treatment groups. A decrease in $C_{20:3}$ was observed in muscle from the forequarter but not from the LD and hindquarter of steers fed 50 g/kg SO as compared to control diet (*10*). The proportions of $C_{20:3}$ fatty acids were considerably higher in muscle than in adipose tissue (highest level, 1.84 vs 0.02 wt % of total fatty acids; **Table 3**). Beaulieu et al. (*10*) also observed lower proportions of $C_{20:3}$ fatty acids in adipose tissue compared with muscle, liver, or intestine.

As mentioned earlier, $C_{18:2}$ *cis*-9, *trans*-11 is the major contributor to total CLA (600−900 g/kg) in adipose tissue (*1*−*3*) and is an isomer responsible for the desirable anticarcinogenic effects of CLA (*21*). Feeding sources rich in $C_{18:2}$ to dairy cows increases $C_{18:2}$ *cis*-9, *trans*-11 in milk fat (*22*, *23*). It was assumed in the present study that feeding increasing amounts of sources rich in $C_{18:2}$ would increase the ruminal and endogenous production of $C_{18:2}$ *cis*-9, *trans*-11 and enhance its concentration in tissues. Surprisingly, we did not observe an effect of dietary SO on the proportion of $C_{18:2}$ *cis*-9, *trans*-11 in either adipose or muscle. Beaulieu et al. (*10*) fed SO at 50 g/kg of the diet to beef heifers and also observed no change in $C_{18:2}$ *cis*-9, *trans*-11 content in muscle tissue.

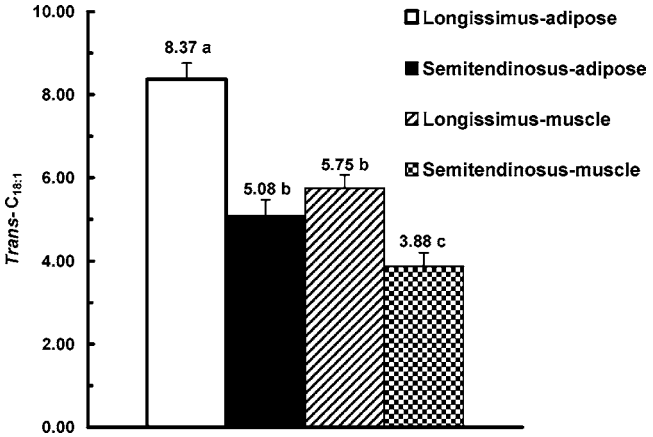

Figure 1. Total *trans*-$C_{18:1}$ fatty acid weight percent of total fatty acids in adipose tissue and muscle from longissimus and semitendinosus of beef steers fed diets containing 0, 20, or 40 g/kg soybean oil on a DM basis. Means with different letters differ at $P < 0.01$.

In the present study, an acidic catalyst was used for fatty acid methyl ester preparation. Studies have shown that recovery of $C_{18:2}$ *cis*-9, *trans*-11 is lower when using an acid catalyst as compared to a base catalyst (*24*, *25*). Although the values reported in this study are lower than would be determined using a basic catalyst for methylation, relative comparisons among treatments should still be valid. Treatment differences in CLA levels in muscle were not affected by use of acidic methylation catalysts (*25*).

Feeding SO at 40 g/kg of the diet increased total *trans*-$C_{18:1}$ content in adipose tissue from LD and ST on days 63 and 105 by 130−168% (**Table 4**). Increases of 31−40% in the proportion of *trans*-$C_{18:1}$ fatty acids in beef fat were observed in other studies when beef cattle were fed diets containing 50 g/kg SO (*10*) or 19−39 g/kg SO through full-fat extruded soybean meal (*9*). Enser et al. (*7*) reported an increase of 166% in the proportion of *trans*-$C_{18:1}$ in muscle by feeding 60 g/kg linseed and fish oil to Charolais steers. In the present study, feeding finishing diets containing 20 and 40 g/kg of SO to beef steers increased *trans*-$C_{18:1}$ of the LD muscle by 70 and 215%, respectively, compared with CTL steers.

Interestingly, the LD had a higher *trans*-$C_{18:1}$ content than ST ($P < 0.03$) within adipose tissue and muscle (**Figure 1**), suggesting that there is a preferential deposition of *trans*-$C_{18:1}$ fatty acids in LD or that the activities of the Δ^9-desaturase

enzyme could be slightly different in LD and ST. The Δ^9-desaturase enzyme activity has been shown to vary among animals, organs, and tissues (*26*, *27*). However, if the activities of Δ^9-desaturase were substantially different in LD and ST, there would probably be a difference in the proportion of $C_{18:2}$ *cis*-9, *trans*-11 in those tissues.

The endogenous synthesis of $C_{18:2}$ *cis*-9, *trans*-11 from *trans*-11 $C_{18:1}$ has been observed in milk fat (*28*). The majority of $C_{18:2}$ *cis*-9, *trans*-11 in milk fat originates via the Δ^9-desaturase enzyme. The Δ^9-desaturase enzyme is present in adipose tissue (*29*) where CLA is synthesized endogenously (*30*). However, an animal's diet may influence the activity of the Δ^9-desaturase enzyme. The abundance of mRNA and the enzyme activity of Δ^9-desaturase are also influenced by hormone balance, physiological state, insulin level, and other activating and inhibiting factors (*27*, *31*). A decline in insulin resulted in a decrease in Δ^9-desaturase gene expression in adipose tissue (*32*), suggesting that the energy level of the diet may influence the activity of Δ^9-desaturase in adipose tissue. In addition, it has been reported that certain isomers of CLA, particularly the $C_{18:2}$ *trans*-10, *cis*-12 isomer, may inhibit the activity and/or expression of the Δ^9-desaturase enzyme, which would result in less endogenous synthesis of $C_{18:2}$ *cis*-9, *trans*-11 (*33*, *34*).

The $C_{18:2}$ *trans*-10, *cis*-12 isomer was undetectable in tissue samples on day 0 and had low detection rates in samples collected on days 63 and 105. The frequencies of detection were 30 and 49% across treatments and muscles on days 63 and 105, respectively. On day 105, the frequency of detection was higher in the SO4 (72%) followed by SO2 (63%) and only 12% in CTL. Low detection rates of $C_{18:2}$ *trans*-10, *cis*-12 were also reported by Beaulieu et al. (*10*). The $C_{18:2}$ *trans*-10, *cis*-12 isomer has been shown to inhibit body fat accretion in mice (*35*) and decrease milk fat synthesis in dairy cows (*36*).

The $C_{18:2}$ *trans*-10, *cis*-12 content in adipose and muscle tissues ranged from 1.0 to 22.5 wt % of total CLA (sum of $C_{18:2}$ *cis*-9, *trans*-11 and $C_{18:2}$ *trans*-10, *cis*-12) across treatments on day 105 (**Tables 3** and **4**). Supplementing SO increased the proportion of $C_{18:2}$ *trans*-10, *cis*-12 in adipose tissue, but not in muscle from LD on day 105, and feeding SO up to day 105 resulted in higher proportions of $C_{18:2}$ *trans*-10, *cis*-12 than feeding up to 63 days (**Table 4**). The proportions of $C_{18:2}$ *trans*-10, *cis*-12 in ST were not different among treatments. Similar increases in $C_{18:2}$ *trans*-10, *cis*-12 were observed in adipose and muscle tissues from beef heifers fed 50 g/kg SO (*10*). In another study, Madron et al. (*9*) detected no significant amounts of $C_{18:2}$ *trans*-10, *cis*-12 in adipose or muscle by feeding 19 or 39 g of SO.

Beaulieu et al. (*10*) demonstrated that feeding 50 g/kg SO resulted in no change in $C_{18:2}$ *cis*-9, *trans*-11 content in the rumen fluid, but did increase the content of $C_{18:2}$ *trans*-10, *cis*-12. Assuming that some of this $C_{18:2}$ *trans*-10, *cis*-12 could be digested, absorbed, and eventually stored in the tissue, this may partially explain the increase in $C_{18:2}$ *trans*-10, *cis*-12 of beef muscle tissue observed in the present study and others. The results on $C_{18:2}$ *trans*-10, *cis*-12 must be interpreted with caution due to smaller proportions and low detection levels as suggested by Beaulieu et al. (*10*).

It seems likely that the acidic ruminal pH in finishing beef cattle alters the microbial population involved in lipid biohydrogenation, thereby influencing the ruminal synthesis of CLA isomers. Acidic conditions in the rumen may decrease the biohydrogenation of $C_{18:2}$ *trans*-10, *cis*-12 and *trans*-$C_{18:1}$, thereby increasing the availability of these two groups. Kalscheur et al. (*37*) demonstrated a direct effect of rumen pH on lipid biohydrogenation by adding buffers to high-concentrate diets, thereby reducing the duodenal flow of *trans* fatty acids in dairy cows. Results from several studies (*10*, *37*, *38*) suggest that high-grain diets resulting in low ruminal pH may cause a shift in the ruminal environment, favoring the production of $C_{18:2}$ *trans*-10, *cis*-12 and *trans*-$C_{18:1}$ fatty acids. This, in turn, would result in higher concentrations of these fatty acids in the beef muscle tissue. In muscle, *trans*-$C_{18:1}$ is present; however, all may not be converted to $C_{18:2}$ *cis*-9, *trans*-11 because of the presence of *trans* fatty acids that are not *trans*-11 $C_{18:1}$. Also, as mentioned earlier, the $C_{18:2}$ *trans*-10, *cis*-12 isomer has been shown to inhibit the activity and gene expression of Δ^9-desaturase (*33*, *34*), which could result in a reduction in the endogenous synthesis of $C_{18:2}$ *cis*-9, *trans*-11.

Results from this study and that of Beaulieu et al. (*10*) suggest that supplementing high-grain, corn-based finishing diets with soybean oil at 20−50 g/kg of the dietary DM is not an effective strategy to enhance $C_{18:2}$ *cis*-9, *trans*-11 content of beef. Supplementing soybean oil increases *trans*-$C_{18:1}$ and $C_{18:2}$ *trans*-10, *cis*-12 contents in beef. The potential nutritive and subsequent health implications of increased proportions of *trans*-$C_{18:1}$ and $C_{18:2}$ *trans*-10, *cis*-12 in beef merit further investigation.

ABBREVIATIONS USED

ADF, acid detergent fiber; BW, body weight; CLA, conjugated linoleic acid; CP, crude protein; DM, dry matter; LD, longissimus dorsi; NE_g, net energy; NDF, neutral detergent fiber; ST, semitendinosus; SO, soybean oil.

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