# Effect of high-oil corn or added corn oil on ruminal biohydrogenation of fatty acids and conjugated linoleic acid formation in beef steers fed finishing diets

S. K. Duckett<sup>\*1</sup>, J. G. Andrae<sup>\*</sup>, and F. N. Owens<sup>†</sup>

\*The University of Georgia, Athens, 30602 and †Pioneer Hi-Bred International, Inc., Johnston, IA, 50131

**ABSTRACT:** Three Angus steers (410 kg) cannulated in the proximal duodenum were used in a replicated 3  $\times$  3 Latin square to evaluate the effects of dietary lipid level and oil source on ruminal biohydrogenation and conjugated linoleic acid (CLA) outflow. Dietary treatments included: 1) typical corn (TC; 79.2% typical corn), 2) high-oil corn (HOC; 79.2% high-oil corn), and 3) the TC diet with corn oil added to supply an amount of lipid equal to the HOC diet (OIL; 76.9% TC + 2.4% corn oil). Duodenal samples were collected for 4 d following 10d diet adaptation periods. Data were analyzed with animal, square, period, and treatment in the model and planned, nonorthogonal contrasts were used to test the effects of dietary lipid content (TC vs HOC and OIL) and oil source (HOC vs OIL) on ruminal biohydrogenation. Intake and duodenal flow of total long-chain fatty acids were increased (P < 0.05) by over 63% for diets containing more lipid regardless of oil source. Apparent ruminal dry matter and long chain fatty acid digestibilities were not altered (P > 0.05) by dietary lipid level or oil source. Ruminal biohydrogenation of total and individual 18-carbon unsaturated fatty acids was greater (P < 0.05) for diets with higher lipid content. Biohydrogenation of oleic acid was greater (P < 0.05)for HOC than OIL, but biohydrogenation of linoleic acid was lower (P < 0.05) for HOC than OIL. Duodenal flows of palmitic, stearic, oleic, linoleic, and arachidic acids were more than 30% greater (P < 0.05) for diets containing more lipid. Flow of all trans-octadecenoic acids was greater (P < 0.05) for diets containing more lipid. Corn oil addition increased (P < 0.05) the flow of trans-10 octadecenoic acid and the trans-10, cis-12 isomer of CLA by threefold compared to feeding high-oil corn. Feeding high-oil corn or adding corn oil to typical corn rations increased intake, biohydrogenation, and duodenal flow of unsaturated long-chain fatty acids. Compared with high-oil corn diets, addition of corn oil increased duodenal flow of trans-10, trans-12 and cis-12 isomers of octadecenoic acid and the trans-10, cis-12 isomer of CLA. The amount of cis-9, trans-11 isomer of conjugated linoleic acid flowing to the duodenum was less than 260 mg/d, a value over 20 times lower than flow of *trans*-11 vaccenic acid indicating the importance of tissue desaturation for enhanced conjugated linoleic acid content of beef.

Key Words: Beef, Biohydrogenation, High-Oil Corn

©2002 American Society of Animal Science. All rights reserved.

J. Anim. Sci. 2002. 80:3353-3360

### Introduction

Through selection, corn grain hybrids containing 7% ether extract, nearly twice the amount found in typical corn, have been developed and are commercially available. Previous research (Andrae et al., 2001) has shown that feeding high-oil corn to steers in finishing rations increased the percentages of linoleic ( $C_{18:2}$ ) and arachidonic ( $C_{20:4}$ ) acids in i.m. lipid. Because linoleate is not synthesized in the body, enrichment in tissue indicates that more linoleic acid reached the small intestine for

absorption and deposition when high-oil corn was substituted for typical corn. This increased intestinal supply of linoleic acid could be from reduced ruminal biohydrogenation of fatty acids, an increased dietary supply of unsaturated fat, or both.

Conjugated linoleic acid (**CLA**) and *trans*-octadecenoic acid are produced in the rumen as intermediates in the biohydrogenation of dietary linoleic acid to stearic acid (Bauman et al., 1999). Conjugated linoleic acid is an anticarcinogen, which reduced tumor proliferation when topically applied to mice with experimentally induced epidermal carcinogenesis (Ha et al., 1987). Chin et al. (1992) reported that CLA concentrations in ground beef are 3.8 to 4.3 mg/g of lipid with about 84% being *cis*-9, *trans*-11 CLA isomer. Concentrations of CLA in milk fat increase with dietary supplementation of unsaturated vegetable oils (McGuire et al., 1996;

 $<sup>^1\!</sup>Correspondence:$  Animal and Dairy Science Complex (E-mail: sduckett@arches.uga.edu).

Received April 16, 2002.

Accepted July 26, 2002.

Kelly et al., 1998) or oilseeds (Stanton et al., 1997; Lawless et al., 1998). However, little research is available on the flow of CLA to the duodenum in cattle fed high-concentrate diets. The hypothesis of this research was to determine if dietary lipid levels or sources increased the flow of unsaturated fatty acids to the small intestine. The objective of this research project was to determine the effect of dietary lipid level or oil source on ruminal biohydrogenation and CLA outflow in beef steers fed finishing diets.

#### Materials and Methods

Animals. Three Angus steers (410 kg) cannulated in the proximal duodenum were fed one of three diets in a replicated  $3 \times 3$  Latin square. The polyethylene Ttype cannulae (Ankom, Fairport, NY) were inserted by a clinical veterinarian into the proximal duodenum of steers while under general anesthesia. The Animal Care and Use Committee approved the surgical procedures and use of animals in this study. The experiment began after a 4-wk recovery period. Steers were housed in covered pens with concrete floors without bedding. Rubber mats were provided in each pen for animal comfort. Steers weighed 410 kg at the beginning of the first square and 480 kg at the beginning of the second square, which started 42 d later. All feeding conditions and animal management practices were similar for each square.

*Diets.* The three dietary treatments were: 1) typical corn (TC; 79.2% typical corn; 14% hay; 3.0% fatty acid content; DM basis), 2) high-oil corn (HOC; 79.2% highoil corn; 14% hay; 5.2% fatty acid content; DM basis), and 3) the TC diet in which corn oil replaced corn so that it contained the same amount of lipid as HOC (OIL; 76.9% typical corn, 14% hay, 2.4% corn oil, and 5.2% fatty acid content; DM basis). Corn was processed by dry rolling, which was similar to the previous feeding experiment of Andrae et al. (2000). Bromegrass hay was coarsely chopped to approximately 5 cm in length. The TC and HOC diets were identical except for the grain source. The typical corn and high-oil corn were supplied by Pioneer Hi-Bred International, Inc. (Johnston, IA) from a TopCross hybrid with the female (ear bearing) parent being the same as for the typical corn. Grain sources were grown in adjacent fields. Ingredient and chemical composition of the diets are presented in Table 1. Diets were designed to be isonitrogenous. No ionophore was included in diets because ionophores might reduce the benefit of added fat (Clary et al., 1993) and alter ruminal biohydrogenation (Dhiman et al., 1999). Total lipid content of typical and high-oil corn was 4.25% and 7.25%, respectively. The amount of corn oil added to the OIL diet was calculated to make the OIL diet equal in lipid to the HOC diet. Animals were individually fed twice daily at 0800 and 1700 with total daily intake being 95% of intake determined during a preliminary period in which feed supply was unlimited to eliminate refusals during the sampling period. Chro-

**Table 1.** Ingredient and chemical composition

 (percentage of diet dry matter) of diets fed to steers<sup>a</sup>

Dry matter, %	Typical corn	High oil corn	Typical corn + corn oil
Ingredients			
Typical corn <sup>b</sup>	79.20	_	76.90
High-oil corn <sup>b</sup>	_	79.20	_
Bromegrass hay	14.00	14.00	14.00
Soybean meal	4.50	4.50	4.50
Dicalcium phosphate	0.20	0.20	0.20
Ground limestone	0.96	0.96	0.96
Urea	0.69	0.69	0.69
Trace mineral salt	0.50	0.50	0.50
Corn oil <sup>c</sup>	_	_	2.37
Chemical composition <sup>d</sup>			
Dry matter	86.1	86.6	86.4
Organic matter	95.4	95.2	95.2
Crude protein	12.0	12.0	11.8
NDF	12.5	13.0	12.9
ADF	4.8	4.7	4.8
Total fatty acids	3.00	5.20	5.17

<sup>a</sup>Diets were fed as total mixed rations at 0800 and 1700.

<sup>b</sup>Typical and high-oil corn were dry rolled prior to feeding.

<sup>c</sup>Mazola corn oil, Bestfoods Foodservice, Franklin Park, IL. <sup>d</sup>NDF = neutral detergent fiber, ADF = acid detergent fiber.

NDF – neutral detergent iber, ibr – acid detergent ibe

mic oxide (5 g per animal per day) was added to the diets as an external marker for calculating duodenal flow.

Sample Collection and Analyses. Experimental periods lasted 14 d with 10 d for diet adaptation and 4 d for sample collection. Following the 10-d adaptation period, duodenal samples (200 mL) were taken 12 times over 4 d, with sampling at 8-h intervals and sampling times shifted ahead by 2 h each day, such that samples represented duodenal contents from 12 equally spaced intervals over a 24-h day. Feed samples were collected once daily during each sampling period. Feed samples were immediately frozen after collection. Duodenal samples were composited on an equal liquid volume basis for each steer within each period and frozen for later processing. Later, duodenal digesta and feed samples were thawed, lyophilized, ground through a Wiley mill (1-mm screen), and frozen at -20°C for subsequent analyses.

Prior to transmethylation of the samples, four CLA standards (free fatty acids; Matreya, Pleasant Gap, PA) were each methylated in triplicate using three methods to examine conversion of CLA isomers to methyl esters. The three methods tested were those of Sukhija and Palmquist (1988), Yurawecz et al. (1999), and Park and Goins (1994). Retention times of the CLA isomers were determined for methyl esters of CLA isomers obtained from Nu-Chek-Prep (Elysian, MN). Results from these method comparisons are presented in Table 2. The method of Sukhija and Palmquist (1988) resulted in lower recoveries of the specific CLA isomers than for the standard. Acid catalyzed transmethylation reactions can alter the *trans* and *cis* double bonds present in CLA, which in turn will invalidate results (Shantha et al., 1993; Yurawecz et al., 1999). The method of Yura-

**Table 2.** Comparison of transmethylation procedures for conversion conjugated linoleic acid (CLA) isomers to methyl esters (reported as a percentage of total fatty acids)

	5	/		
			Method	
	Standard <sup>a</sup>	$1^{\mathrm{b}}$	$2^{c}$	$3^{d}$
Matreya #1245,				
cis-9, trans-11 CLA	98.0	57.4	0.0	97.8
Matreya #1181,				
trans-9, trans-11 CLA	98.0	78.8	0.0	99.1
Matreya #1248,				
cis-9, cis-11 CLA	96.0	100.0	0.0	100.0
Matreya #1249,				
trans-10, cis-12 CLA	98.0	54.9	0.0	97.5

<sup>a</sup>Minimum CLA isomer percentages reported by the manufacturer (Matreya, Pleasant Gap, PA).

<sup>b</sup>Method 1: Sukhija and Palmquist (1988).

<sup>c</sup>Method 2: Yurawecz et al. (1999).

<sup>d</sup>Method 3: Park and Goins (1994).

wecz et al. (1999) did not convert free fatty acids to methyl esters. Others (Luddy et al., 1968; Glass, 1971; Park and Goins, 1994) have reported that base-catalyzed reactions can result in transesterification, but do not convert free fatty acids to methyl esters. The method of Park and Goins (1994) combines the use of sodium methoxide followed by boron trifluoride for a complete conversion of fatty acids to methyl esters. The percentages of the CLA isomers obtained after using the method of Park and Goins (1994) were similar to those reported by the supplier for each CLA standard. Kramer et al. (1997) reported similar results when comparing acid, base, or combination catalysts for methylation of milk and rumen fatty acids.

Fatty acid composition of feed and duodenal samples was determined by the direct transmethylation procedure of Park and Goins (1994). Fatty acid methyl esters (FAME) were analyzed using a HP6890 (Hewlett-Packard, San Fernando, CA) gas chromatograph equipped with a HP7673A (Hewlett-Packard, San Fernando, CA) automatic sampler. Separations were accomplished using a 100-m SP2560 (Supelco, Bellefonte, PA) capillary column (0.25 mm i.d. and 0.20  $\mu$ m film thickness). Column oven temperature was programmed to increase from 150 to 160°C at 1°C per min, from 160 to 167°C at 0.2°C per min, from 167 to 225°C at 1.5°C per min, and then held at 225°C for 5 min. The injector and detector were maintained at 250°C. Sample injection volume was 1 µL. Hydrogen was the carrier gas at a flow rate of 1 mL per min. In order to resolve both the individual trans- and cis-octadecenoic acids and CLA isomers, FAME had to be analyzed twice with GLC at different concentrations and split ratios. For the transand cis- octadecenoic fatty acids and the major longchain fatty acids, FAME with a concentration of 5 µg/ μL were injected into the gas-liquid chromotograph with the above temperature program and a split ratio of 1:20. For CLA isomers, FAME were concentrated to 50  $\mu$ g/ $\mu$ L by reducing solvent volume to 100  $\mu$ L in a limited vial insert and injected into the gas-liquid chromotograph with the above temperature program and a split ratio of 1:1. Peaks were identified by comparison to reference standards from Supelco. Conjugated linoleic acid isomers were identified by using available standards from Matreya and Nu-Chek-Prep, and by methylating Tonalin, a dietary supplement containing numerous CLA isomers at various levels, as described by Benito et al. (2001). Available standards (Sigma, St. Louis, MO) were used to identify *trans*- and *cis*-octadecenoic acids and compared to published chromatograms (Precht et al., 2001; Griinari et al., 1998). Fatty acids were quantified by incorporating an internal standard, methyl tricosanoate  $(C_{23:0})$  into each sample prior to methylation.

Chromium concentration in digesta was measured by inductively coupled plasma spectrometry (ICP; Thermo Jarrell Ash Corp., Franklin, MA) using the sample preparation method of Williams et al. (1962). Concentrations of Cr and fatty acids in the feed, refusals, and digesta were used to calculate digestibility and duodenal flow of fatty acids (Schneider and Flatt, 1975). The percent biohydrogenation of total and individual unsaturated 18-carbon fatty acids were calculated according to Wu et al. (1991). Data were analyzed using the GLM of SAS (SAS Inst., Inc., Cary, NC) with animal, square, period, and treatment included as class variables in the model. Preplanned, nonorthogonal contrasts were used to test the effects of dietary lipid level (TC vs HOC and OIL) and oil source (HOC vs OIL) on ruminal digestion, ruminal biohydrogenation, fatty acid intake, and flow of long-chain fatty acids to the small intestine.

### **Results and Discussion**

The fatty acid composition of typical corn, high-oil corn, and corn oil is presented in Table 3. High-oil corn contained a greater percentage of total fatty acids as oleic acid ( $C_{18:1}$ ), but a lower percentage as linoleic acid. These differences in fatty acid composition between

 Table 3. Fatty acid profiles of typical corn, high-oil corn, and corn oil

	0 ,		
Fatty acid, wt%	Typical corn	High-oi corn	l Corn oil
C <sub>14:0</sub>	0.04	0.05	0.00
C <sub>16:0</sub>	13.56	12.66	10.59
C <sub>16:1</sub> , c-9	0.14	0.11	0.10
$C_{18:0}$	1.81	2.38	1.96
C <sub>18:1</sub> , c-9	23.24	32.25	27.27
C <sub>18:2</sub> , c-9,c-12	57.55	49.39	57.47
C <sub>18:3</sub> , c-9,c-12,c-15	1.84	1.14	0.97
$C_{20:0}$	0.40	0.45	0.43
C <sub>20:1</sub> , c-9	0.23	0.23	0.24
$C_{22:0}$	0.19	0.14	0.14
$C_{24:0}$	0.34	0.20	0.18
Unidentified	0.00	0.68	0.22

high-oil corn and typical corn are similar to those reported by Crum and Stilborn (1997). Corn oil had a similar oleic acid percentage, but was richer in linoleic acid and poorer in palmitic acid ( $C_{16:0}$ ) concentrations than typical corn.

Dry matter intake for all three diets was similar (P> 0.05) and averaged 10.57 kg per animal per day (2.2%) BW; Table 4). Similarly, Eibs et al. (2000) found no reduction in DMI when feeding HOC to finishing steers. Zinn et al. (2000) reported that intakes by finishing cattle were not changed when 2 to 6% yellow grease or 2 to 4% formaldehyde-protected fat was added to the diet. In contrast, Andrae et al. (2000) reported that DMI was lower for cattle fed HOC than when cattle were fed TC. Daily intake of total long-chain fatty acids was 78% greater (P < 0.05) for high-lipid (HOC and OIL) diets compared to low-lipid (TC) diets, but intake was not different (P > 0.05) between oil sources (HOC and OIL). Dry matter flow of duodenal digesta and apparent ruminal dry matter digestibility were not different (P > 0.05) between dietary lipid levels or oil sources. Long chain fatty acid flow (g/d) at the duodenum was 63% greater (P < 0.05) with the higher lipid diets, but was not different (P > 0.05) for HOC and OIL diets. Apparent ruminal long chain fatty acid digestibility was similar (P > 0.05) between dietary lipid levels and oil sources. These values for apparent ruminal digestibility of longchain fatty acids in the rumen are within the range of those reported by other researchers, as summarized by Jenkins (1993).

Intakes of long-chain fatty acids are shown in Table 5. Steers consuming the two diets with higher lipid levels had greater (P < 0.05) intakes for each of the fatty acids measured. Intake of several of the saturated fatty acids (myristic [ $C_{14:0}$ ], stearic [ $C_{18:0}$ ], and arachidic [ $C_{20:0}$ ] acids) and oleic acid was greater (P < 0.05) for HOC than OIL, whereas intakes of other saturated (palmitic) and unsaturated fatty acids (palmitoleic [ $C_{16:1}$ ], linoleic, and linolenic [ $C_{18:3}$ ] acids) were lower (P < 0.05) for HOC than OIL. Diets containing higher lipid levels from substituting with high-oil corn or supplementing

corn oil increased (P < 0.05) dietary intake of unsaturated fatty acids by 193 g/d compared to the low-lipid diet. Intake of total unsaturated fatty acids did not differ (P > 0.05) between oil sources.

Ruminal biohydrogenation was calculated simply by comparison of dietary intake with duodenal flow of individual 18-carbon fatty acids according to Wu et al. (1991). By this calculation, ruminal biohydrogenation of total and individual 18-carbon unsaturated fatty acids was greater (P < 0.05) for diets with higher lipid levels (Table 6). Bolte et al. (2001) also reported that ruminal biohydrogenation of total 18-carbon fatty acids was increased when high-oil corn was fed. Oil source did not alter (P > 0.05) overall 18-carbon unsaturated fatty acid biohydrogenation level, but differences were observed for the individual fatty acids by oil source. Biohydrogenation of oleic acid was greater (P < 0.05) for HOC than OIL diets, but biohydrogenation of linoleic acid was lower (P < 0.05) for HOC than OIL diets. Linolenic acid biohydrogenation tended to be greater (P > 0.05) for OIL than HOC diets. Aldrich et al. (1997) suggested that the seedcoat of canola may physically protect lipids from ruminal biohydrogenation. However, similar overall biohydrogenation levels of unsaturated 18-carbon fatty acids between HOC and OIL diets suggests that the seedcoat of dry-rolled high-oil corn did not offer any protection against biohydrogenation.

Biohydrogenation percentages were highest for linolenic (91%), intermediate for linoleic (80%), and lowest for oleic (70%) acids. Scollan et al. (2001) reported similar levels of biohydrogenation for the linolenic and oleic acids in cannulated steers given free choice access to silage with a barley/sugarbeet supplement (40 to 60% of DMI) containing rumen-protected lipid, linseed oil, or fish oil. Zinn et al. (2000) reported biohydrogenation percentages for cattle consuming wheat-based finishing diets containing 2, 4, or 6% yellow grease that were quite similar to our values. Elizalde et al. (1999) reported higher values (82%) for ruminal biohydrogenation with diets containing white grease (2.5 or 5%) and 52% roughage. Zinn et al. (2000) reported lower biohy-

<b>Table 4.</b> Dry matter and long-chain fatty acid intake and ruminal digestion by steers
fed typical corn (TC), high-oil corn (HOC), or typical corn plus corn oil (OIL)

					Contrasts <sup>a</sup>	
	D16	etary Treatme	ents		L ow vs	HOC WS
Item	TC	HOC OIL		SEM	high lipid	OIL
Intake, g/d						
Dry matter	10,387	10,683	10,639	132.5	0.12	0.82
Fatty acids	310	555	550	5.6	0.01	0.51
Duodenal, g/d						
Dry matter	7364	7786	8276	689.9	0.45	0.63
Fatty acids	345	538	584	52.2	0.01	0.42
Apparent ruminal digestion, %						
Dry matter	28.98	27.24	29.06	5.50	0.91	0.83
Fatty acids	-10.83	3.12	2.81	7.04	0.14	0.95

 $^{\rm a}{\rm Contrasts}$  low- (TC) vs high- (average of HOC and OIL treatments) lipid diets and oil source (HOC vs OIL treatments).

					$Contrasts^{a}$	
	Die	Dietary Treatments			Low vs	HOC vs
Fatty acid	TC	HOC	OIL	SEM	high lipid	OIL
$C_{12:0}$	0.15	0.27	0.27	0.01	0.01	0.48
C <sub>14:0</sub>	0.49	0.92	0.86	0.01	0.01	0.01
C <sub>16:0</sub>	45.25	76.85	79.64	0.82	0.01	0.03
C <sub>16:1</sub> , c-9	0.49	0.74	0.86	0.01	0.01	0.01
C <sub>18:0</sub>	6.88	14.78	12.18	0.14	0.01	0.01
C <sub>18:1</sub> , c-9	63.97	153.89	113.67	1.45	0.01	0.01
C <sub>18:2</sub> , c-9,c-12	162.36	254.14	287.13	2.73	0.01	0.01
C <sub>18:3</sub> , c-9,c-12,c-15	21.02	34.46	37.06	0.37	0.01	0.01
C <sub>20:0</sub>	1.43	2.76	2.54	0.03	0.01	0.01
Unidentified	8.80	16.81	15.56	0.05	0.01	0.01

**Table 5.** Long-chain fatty acid intake (g/d) by steers fed diets containing typical corn (TC), high-oil corn (HOC) or corn oil (OIL)

<sup>a</sup>Contrasts low- (TC) vs high- (average of HOC and OIL treatments) lipid diets and oil source (HOC vs OIL treatments).

drogenation values for steers fed protected fat sources with high-concentrate diets. These biohydrogenation levels for the individual 18-carbon fatty acids indicate that oils rich in oleic acid have the greatest potential for altering tissue composition due to its lower level of biohydrogenation.

The effect of diet on duodenal flow (g/d) of long-chain fatty acids is shown in Table 7. Duodenal flows of palmitic, stearic, oleic, linoleic, and arachidic acids were more than 30% greater (P < 0.05) for diets containing more lipid. Similarly, Bolte et al. (2001) reported greater duodenal flow of stearic, oleic, linoleic, and linolenic acids for steers consuming high-oil corn diets compared to typical corn. Andrae et al. (2000) reported that tissue concentrations of linoleic and arachidonic acids were increased when steers were fed high-oil corn. Presumably, the greater percentages of the essential fatty acids in adipose tissue of cattle fed high-oil corn previously observed is the result of greater intake and flow of these unsaturated fatty acids to the small intestine and not a result of reduced biohydrogenation.

Biohydrogenation of dietary linoleic acid to stearic acid is sometimes incomplete, yielding several interme-

diates, including various CLA isomers and trans- or cisoctadecenoic acids (Bauman et al., 1999). Duodenal flow of all *trans*-octadecenoic acids was greater (P < 0.05)for diets containing greater lipid content. Flow of cisoctadecenoic acids did not differ (P > 0.05) by dietary lipid level. High-oil corn feeding increased (P < 0.05) the flow of trans-11 vaccenic acid by 73% and cis-11 octadecenoic acid by 39% compared to OIL. The trans-10 octadecenoic acid flow was 3.4-fold greater (P < 0.01) for OIL than HOC. Duodenal flows of trans-12 and cis-12 octadecenoic acids were greater (P < 0.05) for OIL than HOC. Similarly, Griinari et al. (1998) reported increased levels of trans-10 octadecenoic acid in milk fat from cows fed low-fiber diets containing 4% corn oil. Beaulieu et al. (2002) reported increased levels of transoctadecenoic acids in rumen contents of steers fed highconcentrate diets supplemented with increasing soybean oil levels.

The predominant CLA isomers identified in duodenal contents were *trans*-9, *trans*-11/*trans*-10, *trans*-12; *cis*-9, *trans*-11; and *trans*-10, *cis*-12 isomers, which accounted for more than 77% of the total CLA present. Duodenal flow of *trans*-10, *cis*-12 CLA isomer and total

**Table 6.** Effects of high-oil corn (HOC) or added corn oil (OIL) on ruminal

 biohydrogenation (%) of individual and overall unsaturated 18-carbon fatty acids

	D				Contrasts <sup>a</sup>	
Fatty acid	TC	HOC	OIL	SEM	Low vs high lipid	HOC vs OIL
	$69.23 \\ 65.77$	73.18 78.90	$72.58 \\ 69.60$	$1.11 \\ 2.77$	0.02 0.03	0.71 0.04
C <sub>18:2</sub> , c-9,c-12 <sup>c</sup> C <sub>18:3</sub> , c-9,c-12,c-15 <sup>c</sup>	77.82 89.33	79.23 91.32	$83.67 \\ 92.85$	$\begin{array}{c} 1.19\\ 0.60\end{array}$	$\begin{array}{c} 0.03 \\ 0.01 \end{array}$	$\begin{array}{c} 0.02\\ 0.10\end{array}$

<sup>a</sup>Contrasts low- (typical corn) vs high- (average of HOC and OIL treatments) lipid diets and oil source (HOC vs OIL treatments).

<sup>b</sup>Total C18 unsaturated fatty acid (USFA) biohydrogenation,  $\% = 100 - [100 \times (\text{total } C_{18} \text{ USFA/total } C_{18} \text{ in digesta})/(\text{total } C_{18} \text{ USFA/total } C_{18} \text{ in feed})].$ 

<sup>c</sup>Individual C<sub>18</sub> USFA biohydrogenation,  $\% = 100 - [100 \times (\text{individual C}_{18} \text{ USFA/total C}_{18} \text{ in digesta})/(\text{individual C}_{18} \text{ USFA/total C}_{18} \text{ in feed})].$ 

					Contr	asts <sup>a</sup>
	Die	tary treatme	ents		T	HOO
Item	TC	TC HOC OIL		SEM	high lipid	OIL
Long-chain fatty acids						
$C_{12:0}$	1.68	1.46	1.73	0.13	0.66	0.18
$C_{14:0}$	5.86	5.06	6.61	0.61	0.98	0.10
$C_{14:1}$	2.46	2.42	2.56	0.28	0.93	0.74
$\mathrm{C}_{15:0}$	2.27	2.26	2.73	0.26	0.54	0.23
$C_{16:0}$	51.24	75.75	78.20	4.50	0.01	0.73
C <sub>16:1</sub> , c-9	0.85	0.79	0.87	0.10	0.90	0.57
$C_{18:0}$	180.63	319.28	342.27	23.70	0.01	0.51
C <sub>18:1</sub> , c-9	21.95	35.92	31.75	2.00	0.01	0.17
C <sub>18:2</sub> , c-9,c-12	30.74	42.34	38.21	2.96	0.02	0.35
C <sub>18:3</sub> , c-9,c-12,c-15	1.58	1.89	1.61	0.16	0.42	0.23
$C_{20:0}$	1.98	3.52	3.44	0.24	0.01	0.80
Biohydrogenation intermediates						
C <sub>18:1</sub> , t-9	0.74	1.91	3.22	0.51	0.01	0.10
C <sub>18:1</sub> , t-10	3.43	4.10	17.91	1.80	0.01	0.01
C <sub>18:1</sub> , t-11	5.02	9.04	5.22	0.66	0.02	0.01
C <sub>18:1</sub> , t-12	0.50	0.65	3.46	0.34	0.01	0.01
C <sub>18:1</sub> , c-11	6.22	8.94	6.42	0.66	0.18	0.02
C <sub>18:1</sub> , c-12	0.63	0.59	1.63	0.24	0.13	0.01
C <sub>18:2</sub> , c-9,t-11	0.185	0.228	0.256	0.049	0.40	0.70
C <sub>18:2</sub> , c-11,t-13	0.037	0.039	0.101	0.025	0.36	0.12
C <sub>18:2</sub> , t-10,c-12	0.040	0.072	0.320	0.039	0.01	0.01
C <sub>18:2</sub> , c-9,c-11	0.089	0.082	0.058	0.022	0.55	0.46
C <sub>18:2</sub> , c-11,c-13	0.000	0.000	0.012	0.004	0.33	0.07
C <sub>18:2</sub> , t-11,t-13/t-8,t-10	0.021	0.008	0.042	0.019	0.86	0.23
C <sub>18:2</sub> , t-9,t-11/t-10,t-12	0.258	0.283	0.456	0.054	0.16	0.05
Total CLA	0.630	0.714	1.245	0.121	0.05	0.01
Unidentified	24.79	32.50	28.10	3.05	0.89	0.10

**Table 7.** Flow (g/d) of long-chain fatty acid and biohydrogenation intermediates at the<br/>duodenum of steers fed diets containing typical corn (TC),<br/>high-oil corn (HOC), or added corn oil (OIL)

<sup>a</sup>Contrasts low- (TC) vs high- (average of HOC and OIL treatments) lipid diets and oil source (HOC vs OIL treatments).

CLA was greater (P < 0.05) for diets with higher lipid content. Dietary lipid level did not alter (P > 0.05) the flow of other CLA isomers including the *cis*-9, *trans*-11 isomer. Similarly, Beaulieu et al. (2002) reported no change in *cis*-9, *trans*-11 CLA isomer level in ruminal contents or tissues of finishing cattle fed varying levels of soybean oil. McGuire et al. (1998) and Dhiman et al. (1999) found no change in beef i.m. fat or milk fat concentrations of *cis*-9, *trans*-11 CLA when high-oil corn was fed to finishing steers or lactating dairy cows, respectively. In contrast, Bolte et al. (2001) reported greater duodenal flow of *cis*-9, *trans*-11 CLA when finishing steers were fed dry-rolled high-oil corn.

Corn oil addition increased (P < 0.05) the flow of *trans*-10, *cis*-12 isomer of CLA by 3.4-fold compared to feeding high-oil corn. Beaulieu et al. (2002) also reported linear increases in *trans*-10, *cis*-12 CLA isomer of ruminal contents from finishing steers supplemented with increasing levels of soybean oil. Others (Griinari et al., 1998; 1999) have shown that feeding low-fiber diets containing unsaturated fats to dairy cows increased the proportion of *trans*-10, *cis*-12 CLA isomer in milk fat. Griinari and Bauman (1999) have proposed pathways

for the formation of *trans*-10, *cis*-12 CLA. Flow of *trans*-9, *trans*-11/*trans*-10, *trans*-12 CLA isomers and total CLA was greater (P < 0.05) for OIL than HOC. Oil addition also tended (P = 0.07) to increase the flow of *cis*-11, *cis*-13 isomer of CLA to duodenum. Flows of other CLA isomers (*cis*-9, *trans*-11; *cis*-11, *trans*-13; *cis*-9, *cis*-11; *trans*-11, *trans*-13/*trans*-9, *trans*-10) were unchanged (P > 0.05) by oil source. Duodenal flow of *trans*-10, *trans*-12 and *cis*-12 octadecenoic acids and *trans*-10, *cis*-12 CLA isomers were over threefold greater for OIL than HOC diets suggesting that oil source may alter microbial populations that favor the *trans*-10 pathway of linoleic acid biohydrogenation.

Flow of *trans*-octadecenoic acids (*trans*-10 and *trans*-11) and CLA isomers (*trans*-10, *cis*-12 and *cis*-9, *trans*-11) as a percentage of linoleic acid intake by dietary treatment is shown in Figure 1. Flow of *cis*-9, *trans*-11 CLA and *trans*-11 vaccenic acids as a percentage of linoleic acid intake did not differ (P > 0.05) for diets with higher lipid content. Flow of *cis*-9, *trans*-11 CLA as a percentage of linoleic acid intake did not differ (P > 0.05) by oil source. Duodenal flow of *trans*-10, *cis*-12 CLA and *trans*-10 octadecenoic acid as a percentage of



**Figure 1.** Flow of *trans*-octadecenoic acids (*trans*-10 and -11) and conjugated linoleic acid (CLA; *trans*-10, *cis*-12 and *cis*-9, *trans*-11) isomers by dietary treatment as a percentage of linoleic ( $C_{18:2}$ ) acid intake.

linoleic acid intake was greater (P > 0.05) for OIL than HOC. Percentage of dietary linoleic acid flowing to the duodenum as trans-11 vaccenic acid was greater (P <0.05) for HOC than OIL. More importantly, duodenal flow of cis-9, trans-11 or trans-10, cis-12 CLA isomer was less than 0.12% of linoleic acid intake, whereas flow of trans-10 or trans-11 octadecenoic acid ranged from 1.5 to 6.3% of linoleic acid intake. The percentage of linoleic acid intake flowing to the duodenum as trans-11 vaccenic acid was 20- to 39-fold greater, depending on dietary treatment, than that of the cis-9, trans-11 isomer of CLA. Santora et al. (2000) have shown that feeding trans-11 vaccenic acid to rodents increases accumulation of cis-9, trans-11 CLA isomer in adipose tissue, which suggests an important contribution of  $\Delta^9$ desaturase to CLA tissue accumulation. In lactating dairy cows, infusion of trans-vaccenic acid into the abomasum also increased the concentrations of cis-9, trans-11 CLA in milk fat (Griinari et al., 2000), whereas infusion of sterculic acid, an inhibitor of  $\Delta^9$ -desaturase enzyme, reduced cis-9, trans-11 levels in milk fat. From these experiments, Griinari et al. (2000) estimated that 64% of the *cis*-9, *trans*-11 CLA present in milk fat came from the desaturation of infused trans-vaccenic acid by  $\Delta^9$ -desaturase enzyme in mammary tissue. Bovine adipose tissues also contain  $\Delta^9$ -desaturase and can convert stearic acid to oleic acid (St. John et al., 1991). The very small duodenal flow of cis-9, trans-11 CLA suggests that conversion of *trans*-11 vaccenic acid to *cis*-9, *trans*-11 CLA in adipose tissue by  $\Delta^9$ -desaturase also serves as major source of CLA in beef fat.

## Implications

Feeding high-oil corn or adding corn oil to typical corn rations increased both intake and duodenal flow of unsaturated long-chain fatty acids. Higher lipid diets, from added corn oil or high-oil corn increased ruminal biohydrogenation of 18-carbon unsaturated fatty acids. Compared with high-oil corn diets, the addition of corn oil to a typical corn diet increased linoleic acid biohydrogenation and duodenal flow of specific isomers of octadecenoic and conjugated linoleic acids. However, the amount of *cis*-9, *trans*-11 isomer of conjugated linoleic acid flowing to the duodenum was less than 260 mg/d, a value more than 20 times lower than the flow of *trans*-11 vaccenic acid, indicating the importance of tissue desaturation for enhanced conjugated linoleic acid content of beef.

#### Literature Cited

- Aldrich, C. G., N. R. Merchen, J. K. Drackley, S. S. Gonzalez, G. C. Fahey Jr., and L. L. Berger. 1997. The effects of chemical treatment of whole canola seed on lipid and protein digestion by steers. J. Anim. Sci. 75:502–511.
- Andrae, J. G., S. K. Duckett, C. W. Hunt, G. T. Pritchard, and F. N. Owens. 2001. Effects of feeding high-oil corn to beef steers on carcass characteristics and meat quality. J. Anim. Sci. 79:582–588.
- Andrae, J. G., C. W. Hunt, S. K. Duckett, L. R. Kennington, P. Feng, F. N. Owens, and S. Soderlund. 2000. Effect of high-oil corn on growth performance, diet digestibility, and energy content of finishing diets fed to beef cattle. J. Anim. Sci. 78:2257–2262.
- Bauman, D. E., L. H. Baumgard, B. A. Corl, and J. M. Griinari. 1999. Biosynthesis of conjugated linoleic acid in ruminants. Proc. Am. Soc. Anim. Sci. Available: http://www.asas.org/jas/journal.asp? Society=asas&month=0&year=0. Accessed March 3, 2001.
- Beaulieu, A. D., J. K. Drackley, and N. R. Merchen. 2002. Concentrations of conjugated linoleic acid (*cis-9 trans-11* octadecadienoic acid) are not increased in tissue lipids of cattle fed a high concentrate diet supplemented with soybean oil. J. Anim. Sci. 80:847–861.
- Benito, P., G. J. Nelson, D. S. Kelley, G. Bartolini, P. C. Schmidt, and V. Simon. 2001. The effect of conjugated linoleic acid on plasma lipoproteins and tissue fatty acid composition in humans. Lipids 36:229–236.
- Bolte, M. R., E. J. Scholljegerdes, B. W. Hess, J. Gould, D. C. Rule, and F. N. Owens. 2001. Long-chain fatty acid flow in and digestion by beef steers fed dry-rolled or high-moisture typical or high-oil corn diets. J. Anim. Sci. 79(Suppl. 2):98 (Abstr.).
- Chin, S. F, W. Lui, J. M. Storkson, Y. L. Ha and M. W. Pariza. 1992. Dietary sources of conjugated dienoic isomers of linoleic acid, a newly recognized class of anticarcinogens. J. Food Comp. Anal. 5:185–197.
- Clary, E. M., R. T. Brandt, Jr., D. L. Harmon, and T. G. Nagaraja. 1993. Supplemental fat and ionophores in finishing diets: Feedlot performance and ruminal digesta kinetics in steers. J. Anim. Sci. 71:3115–3123.
- Crum, R. C., and H. L. Stilborn. 1997. Valuing high-oil corn. Feed Mange. 48:16–20.
- Dhiman, T. R., G. R. Anand, L. D. Satter, and M. W. Pariza. 1999. Conjugated linoleic acid content of milk from cows fed difference diets. J. Dairy Sci. 82:2146–2156.
- Eibs, M. S., B. J. Johnson, D. M. Wulf, B. D. Rops, and F. N. Owens. 2000. Effects of high-oil corn on feedlot performance, carcass characteristics and meat quality. Page 76 in Proc. Plains Nutr. Counc. Spring Conf. Texas A&M Research and Education Center, Amarillo.

- Elizalde, J. C., C. G. Aldrich, D. W. LaCount, J. K. Drackley, and N. R. Merchen. 1999. Ruminal and total tract digestibilities in steers fed diets containing liquefied or prilled saturated fatty acids. J. Anim. Sci. 77:1930–1939.
- Glass, R. G. 1971. Alcoholysis, saponification and the separation of fatty acid methyl esters. Lipids 6:919–924.
- Griinari, J. M., and D. E. Bauman. 1999. Biosynthesis of conjugated linoleic acid and its incorporation into meat and milk in ruminants. Pages 180–200 in Advances in Conjugated Linoleic Acid Research. Vol. 1. M. P. Yurawecz, M. M. Mossoba, J. K. G. Kramer, M. W. Pariza, and G. J. Nelson, ed. AOCS Press, Champaign, IL.
- Griinari, J. M., B. A. Corl, S. H. Lacy, P. Y. Chouinard, K. V. V. Nurmela, and D. E. Bauman. 2000. Conjugated linoleic acid is synthesized endogenously in lactating dairy cows by  $\Delta^9$ -desaturase. J. Nutr. 130:2285–2291.
- Griinari, J. M., D. A. Dwyer, M. A. McGuire, D. E. Bauman, D. L. Palmquist, and K. V. V. Nurmela. 1998. *Trans*-octadecenoic acids and milk fat depression in lactating dairy cows. J. Dairy Sci. 81:1251–1261.
- Griinari, J. M., K. Nurmela, D. A. Dwyer, D. N. Barbano, and D. E. Bauman. 1999. Variation of milk fat concentration of conjugated linoleic acid and milk fat percentage is associated with a change in ruminal biohydrogenation. J. Anim. Sci. 77(Suppl. 1):117– 118 (Abstr.).
- Ha, Y. L., N. K. Grimm and M. W. Pariza. 1987. Anticarcinogens from fried ground beef: Heat-altered derivatives of linoleic acid. Carcinogenesis 8:1881–1887.
- Jenkins, T. C. 1993. Lipid metabolism in the rumen. J. Dairy Sci. 76:3851–3863.
- Kelly, M. L., J. R. Berry, D. A. Dwyer, J. M. Griinari, P. Yvan Chouinard, M. E. Van Amburgh, and D. E. Bauman. 1998. Dietary fatty acid sources affect conjugated linoleic acid concentrations in milk from lactating dairy cows. J. Nutr. 128:881–885.
- Kramer, J. K. G., V. Fellner, M. E. R. Dugan, F. D. Sauer, M. M. Mossoba, and M. P. Yurawecz. 1997. Evaluating acid and base catalysts in the methylation of milk and rumen fatty acids with special emphasis on conjugated dienes and total trans fatty acids. Lipids 32:1219–1228.
- Lawless, F., J. J. Murphy, D. Harrington, R. Devery, and C. Stanton. 1998. Elevation of conjugated *cis-9*, *trans-11-octadecadienoic* acid in bovine milk because of dietary supplementation. J. Dairy Sci. 81:3259–3267.
- Luddy, F. E., R. A. Barford, S. F. Herb, and P. Magidman. 1968. A rapid and quantitative procedure for the preparation of methyl esters of butteroil and other fats. J. Am. Oil Chem. Soc. 45:549-551.
- McGuire, M. A., S. K. Duckett, J. G. Andrae, J. G. Giesy, and C. W. Hunt. 1998. Effect of high-oil corn on conjugated linoleic acid (CLA) in beef. J. Anim. Sci. 76(Suppl. 1):301 (Abstr.)

- McGuire, M. A., M. K. McGuire, M. A. Guy, W. K. Sanchez, T. D. Shultz, L. Y. Harrison, D. E. Bauman, and J. M. Griinari. 1996. Short-term effect of dietary lipid concentration on content of conjugated linoleic acid (CLA) in milk from dairy cattle. J. Anim. Sci. 74(Suppl. 1):266 (Abstr.).
- Park, P. W., and R. E. Goins. 1999. In situ preparation of fatty acid methyl esters for analysis of fatty acid composition in foods. J. Food Sci. 59:1262–1266.
- Precht, D., J. Molkentin, F. Destaillats, and R. L. Wolff. 2001. Comparative studies on individual isomeric 18:1 acids in cow, goat, and ewe milk fats by low-temperature high-resolution capillary gas-liquid chromatography. Lipids 36:827–832.
- Santora, J. E., D. L. Palmquist, and K. L. Roehrig. 2000. Transvaccenic acid is desaturated to conjugated linoleic acid in mice. J. Nutr. 130:208–215.
- Schneider, B. H., and W. P. Flatt. 1975. Pages 168–178 in The Evaluation of Feeds Through Digestibility Experiments. Univ. of Georgia Press, Athens.
- Scollan, N. D., M. S. Dhanoa, N. J. Choi, W. J. Maeng, M. Enser, and J. D. Wood. 2001. Biohydrogenation and digestion of longchain fatty acids in steers fed on different sources of lipid. J. Agric. Sci. 136:345–355.
- Shantha, N. C., E. A. Decker, and B. Hennig. 1993. Comparison of methylation methods for the quantification of conjugated linoleic acid isomers. J. Assoc. Off. Anal. Chem. Int. 76:644–649.
- St. John, L. C., and S. B. Smith. 1987. Fatty acid elongase and desaturase activities in bovine liver and adipose tissue microsomes. J. Anim. Sci. 64:1441–1447.
- Stanton, C., F. Lawless, G. Kjellmer, D. Harrington, R. Devery, J. F. Connolly, and J. Murphy. 1997. Dietary influences on bovine milk *cis*-9, *trans*-11-conjugated linoleic acid content. J. Food Sci. 62:1083–1086.
- Sukhija, P. S., and D. L. Palmquist. 1988. Rapid method for determination of total fatty acid content and composition of feedstuffs and feces. J. Agric. Food Chem. 36:1202–1206.
- Williams, C. H., D. J. David, and O. Iismaa. 1962. The determination of chromic oxide in faeces samples by atomic absorption. J. Agric. Sci. 59:381–385.
- Wu, Z., O. A. Ohajuruka, and D. L. Palmquist. 1991. Ruminal synthesis, biohydrogenation, and digestibility of fatty acids by dairy cows. J. Dairy Sci. 74:3025–3034.
- Yurawecz, M. P., J. K. G. Kramer, and Y. Ku. 1999. Methylation procedures for CLA. Pages 64–82 in Advances in Conjugated Linoleic Acid Research. Vol. 1. M. P. Yurawecz, M. M. Mossoba, J. K. G. Kramer, and M. W. Pariza, ed. AOCS Press, Champaign, IL.
- Zinn, R. A, S. K. Gulati, A. Plascencia and J. Salinas. 2000. Influence of ruminal biohydrogentation on the feeding value of fat in finishing diets for feedlot cattle. J. Anim. Sci. 78:1738–1746.