Milk Fatty Acid Composition of Cows Fed a Total Mixed Ration or Pasture Plus Concentrates Replacing Corn with Fat

G. F. Schroeder,*[†] J. E. Delahoy,[†] I. Vidaurreta,[‡] F. Bargo,[†]

G. A. Gagliostro,[‡] and L. D. Muller[†]

*Consejo Nacional de Investigaciones Científicas y Técnicas (CONICET), Argentina

[‡]Instituto Nacional de Tecnología Agropecuaria,

Estación Experimental Balcarce, CC 276, (7620), Balcarce, Argentina

[†]Department of Dairy and Animal Science, The Pennsylvania State University, University Park 16802

ABSTRACT

Thirty-one Holstein cows (six ruminally cannulated) were used to evaluate milk fatty acids (FA) composition and conjugated linoleic acid (CLA) content on three dietary treatments: 1) total mixed rations (TMR), 2) pasture (*Avena sativa* L.) plus 6.7 kg DM/d of corn-based concentrate (PCorn), and 3) pasture plus PCorn with 0.8 kg DM/d of Ca salts of unsaturated FA replacing 1.9 kg DM/d of corn (PFat). No differences were found in total (22.4 kg/d) or pasture (18.5 kg/d) dry matter intake, ruminal pH, or total volatile fatty acids concentrations. Fat supplementation did not affect pasture neutral detergent fiber digestion. Milk production did not differ among treatments (19.9 kg/d) but 4% fat-corrected milk was lower for cows fed the PFat compared to cows fed the TMR (16.1 vs. 19.5 kg/d) primarily because of the lower milk fat percentage (2.56 vs. 3.91%). Milk protein concentration was higher for cows fed the TMR than those on both pasture treatments (3.70 vs. 3.45%). Milk from the cows fed the PCorn had a lower content of short- (11.9 vs. 10.4 g/100 g) and medium-chain (56.5 vs. 47.6 g/100 g) FA, and a higher C18:3 percentage (0.07 vs. 0.57 g/100 g) compared with TMR-fed. Cows fed the PFat had the lowest content of short- (8.85 g/100 g) and medium-chain (41.0 g/100 g) FA, and the highest of long-chain FA (51.4 g/100 g). The CLA content was higher for cows in PCorn treatment (1.12 g/100 g FA) compared with cows fed the TMR (0.41 g/100 g FA), whereas the cows fed the PFat had the highest content (1.91 g/100 g FA). Pasture-based diets increased the concentrations of long-chain unsaturated FA and CLA in milk fat. The partial re-

placement of corn grain by Ca salts of unsaturated FA in grazing cows accentuated these changes. However, those changes in milk FA composition were related to a depression in milk fat.

(Key words: grazing dairy cow, fat supplementation, conjugated linoleic acid)

Abbreviation key: CLA = conjugated linoleic acid, CSFA = Ca salts of fatty acids, FA = fatty acids, INTA = National Institute of Agriculture Technology, IVDM = in vitro DM digestibility, MFD = milk fat depression, PCorn = pasture plus corn-based concentrate, PFat = pasture plus corn-based concentrate with Ca salts of fatty acids, WSC = water-soluble carbohydrates.

INTRODUCTION

High producing dairy cows fed pasture-based diets have lower DMI and milk production than do cows fed a nutritionally balanced TMR (Kolver and Muller, 1998; White et al., 2001; Bargo et al., 2002). This suggests that diets based on high quality pasture do not provide enough energy for dairy cows to express their high genetic potential (Kolver and Muller, 1998; Bargo et al., 2003). Concentrates containing starch-rich feed ingredients such as corn are supplemented to grazing cows to increase energy intake and milk production and to minimize losses of body condition and reproductive efficiency (Bargo et al., 2003). Partial replacement of cereal grains with fat supplements, which have a higher concentration of energy than starch-based supplements, is an alternative to increase energy intake and long-chain fatty acids (FA) in milk fat of grazing cows (Agenäs et al., 2002; Schroeder et al., 2002). However, few studies have compared animal performance between TMR and pasture-based diets supplemented with corn-based concentrates with or without fat addition, and there is limited information on the effect of fat supplementation on milk FA composition and spe-

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Corresponding author: G. A. Gagliostro; e-mail: ggagliostro@balcarce.inta.gov.ar.

[†]Present address: Kansas State University, Department of Animal Science and Industry, 108 Call Hall, Manhattan, KS, 66506-1600. E-mail: gfs10@ksu.edu.

cifically on conjugated linoleic acid (CLA) content of grazing dairy cows.

Compared with TMR diets, pasture-based diets have resulted in higher concentrations of unsaturated long-chain FA and CLA in milk (Kelly et al., 1998; White et al., 2001). Milk CLA results from both ruminal microbial biohydrogenation of dietary C18:2 and desaturation by mammary Δ^9 -desaturase of vaccenic acid, with more than 75% of total CLA originating from endogenous synthesis (Griinari and Bauman, 1999). Increasing the CLA content in milk has become an important objective of feeding strategies because of the potential benefits of CLA for human health including inhibition of carcinogenesis and atherosclerosis, alteration in lipid metabolism, stimulation of immune stimulation, and reduction in diabetes (Pariza, 1999). Supplementing the diets of dairy cows with specific unsaturated long-chain FA may enhance the beneficial effects of pasture intake on milk FA profile (Lawless et al., 1998; Kolver et al., 2002). However, the inclusion of unsaturated FA to ruminant diets has been also associated with negative effects on ruminal fiber digestion (Palmquist, 1988). Unsaturated long-chain FA supplementation as calcium salts (CSFA) may reduce the negative effects on microbial microorganisms and increase the absorption of unsaturated FA (Chalupa et al., 1986). The objective of this study was to compare the effects of feeding a TMR diet or pasture-based diets supplemented with two types of concentrates (corn-based or replacing part of corn with CSFA) on milk FA composition, animal performance, and ruminal digestion.

MATERIALS AND METHODS

Cows and Treatments

The experiment was conducted at the National Institute of Agriculture Technology (INTA) of Balcarce, Argentina, during the winter of 2000. The INTA Animal Committee approved all procedures involving dairy cows. Two months before the experiment (pretrial period), 31 multiparous Holstein cows (587 ± 58 kg BW; 57 ± 6 DIM; mean $\pm$ SD), six ruminally cannulated, were fed a TMR containing 18% corn grain, 59% corn silage, 22% sunflower meal, 0.5% urea, and 0.9% mineral-vitamin premix on DM basis (Table 1). The TMR composition was based on NRC (2001) recommendations. Cows were stratified in groups of three based on milk production in the pretrial period and randomly assigned for a period of 5 wk to one of three treatments: 1) same TMR fed during the pretrial period (TMR, $n = 11$), 2) winter oats (*Avena sativa* L.) pasture plus 6.7 kg DM/d of a corn-based concentrate (PCorn, $n = 10$, Table 1), and 3) winter oats (*Avena sativa* L.) pasture plus the same concentrate as in PCorn but replacing

Table 1. Feed ingredients for the TMR and the grazing treatments.

Ingredients kg DM/cow per day	TMR	Pasture ¹	Concentrates ²	
			PCorn	PFat
Corn Silage	15.3	—	—	—
Ground corn	4.7	—	6.1	4.2
Fish meal	—	—	0.34	0.34
Sunflower meal	5.7	—	—	—
Urea	0.12	—	—	—
Mineral vitamin premix ³	0.24	—	0.25	0.25
Ca salts of FA ⁴	—	—	—	0.80
Total offered	26.0	29.0	6.7	5.6

¹Winter oats (*Avena sativa* L.).

²PCorn = pasture plus corn-based concentrate, PFat = pasture plus corn-based concentrate with Ca salts of fatty acids.

³Contained 21% Ca₃(PO₄)₂, 4.6% P₂O₅, 6% MgO, 3% molasses, 0.15% FeSO₄, 0.4% CuSO₄, 0.3% ZnSO₄, 0.04% MnSO₄, 0.02% CoSO₄, 0.02% Na₂SeO₃, 0.01% I₂, vitamin A (6,500,000 IU/kg), vitamin D₃ (1,600,000 IU/kg), vitamin E (12,500 IU/kg), 64.46% excipient.

⁴Ca salts of unsaturated fatty acid (Full Fat BP, Inagro, Buenos Aires, Argentina): C14:0 1.6%, C16:0 16%, C16:1 1.6%, C18:0 13.5%, C18:1 32%, C18:2 30%, C18:3 0.8%, and C20:0 0.3%.

1.9 kg DM/d of corn with 0.8 kg DM/d of Ca salts of FA (PFat, $n = 10$, Table 1). Both concentrates were formulated to daily provide a similar amount of energy (14.0 Mcal of NE_L/d, Table 2). At the beginning of the experimental period the cows were 117 ± 6 DIM (mean $\pm$ SD). Two cows fitted with rumen cannula were included in each treatment to describe corn silage and pasture digestion of the TMR treatment and grazing treatments and ruminal fermentation. The length of the experimental period (5 wk) was chosen to avoid changes in pasture FA composition and because milk

Table 2. Chemical composition and in vitro DM digestibility (IVDMD) of TMR, pasture, and concentrates fed during the experiment.

	TMR	Pasture ¹	Concentrates ²	
			PCorn	PFat
DM, %	40.8	21.8 $\pm$ 2.6	87.7	88.7
OM, %	92.6	89.4 $\pm$ 0.4	93.3	88.9
	% of DM			
IVDMD	73.6	77.6 $\pm$ 5.0	86.9	68.8
CP	16.5	18.0 $\pm$ 3.2	12.1	11.0
NDF	30.6	33.2 $\pm$ 3.9	8.7	9.2
Starch	16.9	—	61.4	49.9
WSC ³	14.3	20.2 $\pm$ 3.2	n.d. ⁴	n.d. ⁴
Ether extract	4.5	6.1 $\pm$ 1.1	6.2	19.6
NE _L Mcal/kg DM ⁵	1.45	1.63	2.10	2.56

¹Winter oats (*Avena sativa*, L.) sampled by hand-plucking (mean $\pm$ SD).

²PCorn = pasture plus corn-based concentrate, PFat = pasture plus corn-based concentrate with Ca salts of fatty acids.

³Water-soluble carbohydrates.

⁴Not determined.

⁵Estimated according NRC (2001).

FA composition changes can be detected after a 2-wk period (Dewhurst et al., 2001).

Cows on the TMR treatment were fed once daily after the a.m. milking to approximately 10% orts. Cows on the PCorn and PFat treatments started grazing the winter oats pasture (Table 1) on d 1 of the 5-wk experimental period, and the grazing time was gradually increased from 9 to 21 h over a period of 3 d. A temporary electric fence was used to adjust the area to assure an herbage allowance of approximately 29 kg DM/d per cow to allow an adequate pasture DMI and minimize substitution rate (Bargo et al., 2003). The concentrates were offered in two equal feedings during milking times (0600 and 1600 h), and concentrate DMI individually recorded by weighing the amounts offered and refused for each cow. The FA composition of the CSFA (Full Fat BP, Inagro, Buenos Aires, Argentina) was: 1.6% C14:0, 16% C16:0, 1.6% C16:1, 13.5% C18:0, 32% C18:1, 30% C18:2, 0.8% C18:3, and 0.3% C20:0, or 65% unsaturated FA and 31% saturated FA. The fat supplement was added to approximately provide 13% of the total metabolizable energy requirements (Palmquist, 1988).

Sample Collection and Analysis

Milk production was recorded daily. Milk samples were collected weekly (including the last week of pre-trial period) at a.m. and p.m. milking, composited by weight, and analyzed for fat, total protein, and lactose by infrared spectrophotometry (AOAC, 1990; Foss 605B Milko-Scan, Foss Electric, Hillerød, Denmark). The composition of FA was determined at The Pennsylvania State University on samples collected at the end of the pre-trial period (wk 0), and wk 2 and 5 of the experimental period. Milk FA were extracted and subsequently transmethylated as described by Baumgard et al. (2002). Fatty acid methyl esters were quantified by gas chromatography (Hewlett Packard 6890). Separations were made with the SP 2560 fuse silica capillary column (Supelco, Bellefonte, PA). The column was 100 m in length, with an inner diameter of 0.25 mm and a film thickness of 0.2 μ m. Oven temperatures were initially at 80°C and then ramped at 2°C/min until 180°C and held for 15 min. Helium was the carrier gas and flowed at 1.1 ml/min (17 cm/s, velocity). Airflow was set at 400 ml/min and the make gas, hydrogen was at 45 ml/min. Inlet and detector temperature were at 250°C. Retention times were determined with pure methyl ester standards (Nu Check Prep, Elysian, MN; GLC-60, *cis*-9, *trans*-11 CLA, and *trans*-10, *cis*-12 CLA). A butter oil reference standard (CRM 164; Commission of the European Community Bureau of References, Brussels, Belgium) was used to determine the efficiencies of recoveries and correction factors for individual FA as de-

scribed by Baumgard et al. (2002). Molar basis FA production (mmol/d) was estimated by dividing the yield (on a mass basis) by the molecular weight of each individual FA as described by Peterson et al. (2002).

Herbage mass was determined weekly by cutting 20 quadrats (0.1 m²/quadrat) of pasture to ground level and drying at 105°C in a forced-air oven. To estimate the quality of the pasture, herbage samples were collected by hand-plucking at random transects every 10 d. Samples of TMR and PCorn and PFat concentrates were collected weekly. Pasture, concentrates, and TMR samples were dried at 60°C in a forced-air oven, ground through a 1-mm screen (Wiley mill, Philadelphia, PA), and analyzed for DM (105°C in a forced-air oven for 24 h) OM (450°C for 8 h), NDF, and ADF (Goering and Van Soest, 1990), CP (AOAC, 1990), *in vitro* DM digestibility using the two-stage procedure (IVDMD; Tilley and Terry, 1963), water-soluble carbohydrate (WSC; AOAC, 1990), starch (MacRae and Armstrong, 1968), and ether extract (AOAC, 1990).

Dry matter intake was estimated during wk 4 and 5 of the experiment. On the TMR treatment, DMI was measured as the amount offered minus the amount refused. On the two grazing treatments, DMI of concentrate was also measured by difference between the amount offered and the amount refused. The DMI of pasture was estimated using Cr₂O₃ as fecal marker, which was dosed using controlled-release capsules of Captec (Nufarm Ltd., Auckland, New Zealand; Parker et al., 1989) with a release rate of 1.7 g of Cr/d. Cows were dosed 7 d before the start of fecal sampling. Fecal grab samples were collected twice daily after each milking during 9 d, and analyzed for Cr by atomic absorption spectrophotometry (Parker et al., 1989). Fecal Cr concentration was used to calculate total fecal output using the equation: total fecal output = (g of Cr dosed per day)/(g of Cr/g of fecal DM). Fecal output from the concentrate was estimated as: concentrate fecal output = concentrate DMI $\pm$ (1 - IVDMD of concentrate). Pasture DMI was determined as: pasture DMI = (total fecal output - concentrate fecal output)/(1 - IVDMD of pasture).

Cows on the TMR treatment were weighed after the a.m. milking on 2 consecutive days at the end of pre-trial period. To avoid confounding effects of ruminal fill, the initial BW on the two grazing treatments was determined on wk 1. All cows were also weighed as described on wk 5 at the end of the trial. The same day that BW was measured, BCS was determined by three independent observers using a five-point scale (1 = thin to 5 = fat). Preprandial blood samples were collected on the last day of the experimental period from the jugular vein into one tube containing sodium heparin (Abbot Laboratory, Argentina). Samples were centrifuged

(5000 $\times$ g for 10 min), plasma was collected and stored at -20°C . Plasma was analyzed for glucose (Wiener Laboratory, Argentina), plasma urea nitrogen (Wiener Laboratory), triacylglycerides (Wiener Laboratory), and NEFA (Wako Pure, Chemical Industries USA, Inc., Dallas, TX).

Rumen Fermentation

The last day of wk 4, rumen liquid from cannulated cows was obtained from the dorsal, ventral, and caudal areas of the rumen starting at 0600 h at 0, 3, 6, 9, 12, 16, and 20 h and squeezed through four layers of cheesecloth. The rumen samples were taken while the cannulated cows were grazing in a strip next to the rest of the cows to avoid alterations in grazing time and ruminal fermentation patterns. The pH of the filtered ruminal fluid was measured immediately (Orion portable pH meter 250A, Orion Research Inc., Boston, MA), a 50-ml aliquot was preserved with 0.5 ml of H_2SO_4 , and frozen at -20°C . Samples were subsequently centrifuged at 15,000 $\times$ g at 4°C for 15 min. These samples were analyzed for $\text{NH}_3\text{-N}$ and VFA concentration as described by Schroeder et al. (2002).

The ruminal digestion of the forage NDF of each diet (corn silage or pasture) was determined by the in situ technique. Corn silage and hand-plucked pasture samples were cut to approximately 1-cm pieces, and wet material representing approximately 5 g of DM/bag was immediately placed in duplicate Dacron bags (15.5 cm $\pm$ 7.5 cm; Ankom, Fairport, NY) with a mean pore size of 52 μm . The Dacron bags containing corn silage were incubated in the rumen cannulated cows on the TMR treatment, and the Dacron bags containing pasture were incubated in the ruminally cannulated cows on the two grazing treatments. All bags were placed in the ventral sac of the rumen at the same time and removed at 3, 6, 9, 12, 16, 20, 28, 48, and 72 h after incubation in the ventral sac of the rumen. Zero-hour bags were soaked in warm water and washed with the other bags. To evaluate the effects of the three dietary treatments on ruminal NDF digestion, additional Dacron bags containing a standard NDF ground through a 2-mm screen were incubated in all treatments during 9, 16, and 48 h. The NDF was extracted and prepared treating plant material (*Agropyrum elongatum* L.) with sodium lauryl sulfate to remove soluble material, washing with water and acetone, and drying at 65°C (Uden et al., 1980). All bags were washed in a pipette washer for 1 h and then hand-washed in cold tap water with gentle agitation and squeezed until the water was clear. The bags were dried at 60°C for 48 h, then were weighed and the residue was ground through a 1-mm screen. Duplicated residues at each time point were composited within cow

and analyzed for NDF. Kinetics of ruminal DM and NDF degradation were estimated using the model of Ørskov and McDonald (1979): $D = A + B(1 - e^{-kd \times t})$, where D = disappearance at time (t), A = soluble fraction (%), wash value at 0 h), B = insoluble potentially digestible fraction (%), kd = fractional rate of degradation (%/h), and t = time of incubation. Total potentially degradable fraction was estimated as $A + B$. All model parameters were estimated with a nonlinear model of SAS (1996) using the Marquardt iterative method.

Statistical Analysis

The mean of the milk production and composition data recorded during wk 4 and 5 of the experiment were analyzed using the PROC GLM of SAS (1996) as a completely randomized design adjusted using the production and composition of the pretrial period as covariates, according to the model: $Y_{ij} = \mu + T_i + \beta(x_{ij} - \bar{x}_{.}) + e_{ij}$, where Y_{ij} = dependent variable, μ = population mean, T_i = effect of the i th treatment, β = linear regression coefficient between Y_{ij} and x_{ij} , x_{ij} = value of covariate corresponding to Y_{ij} , and $\bar{x}$ = mean of x_{ij} and e_{ij} = residual error.

The changes in milk fat percentage and yield, milk protein percentage, and milk fatty acid composition over time were analyzed as a repeated measures design using the PROC MIXED of SAS (1996) according to the model: $Y_{ijk} = \mu + T_i + A_{(ij)} + W_k + (T + W)_{ik} + \varepsilon_{ijk}$, where Y_{ijk} = mean of response variable, μ = population mean, T_i = effect of treatment, A_{ij} = random effect of animal within the treatments, W_k = effect of wk sampled, $(T + W)_{ik}$ = effect of interaction of treatment and time sampled, and ε_{ijk} = experimental error. Orthogonal contrasts were used when the interactions were significant. The orthogonal contrasts were TMR treatment versus grazing treatments and PCorn versus PFat treatments.

The DMI, changes in BW and BCS, metabolite concentrations, and rumen parameters were analyzed as a completely randomized design according to the model: $Y_{ij} = \mu + T_i + \varepsilon_{ij}$, where Y_{ij} = mean of response variable, μ = population mean, T_i = effect of treatment, and ε_{ij} = experimental error.

Differences were declared significant at $P < 0.05$ unless otherwise noted, and the separations of means were done using the test of Duncan.

RESULTS AND DISCUSSION

Feed Quality and Dry Matter Intake

The pregrazing and postgrazing herbage mass averaged 1445 ± 222 kg DM/ha and 1044 ± 126 kg of DM/ha (mean $\pm$ SD), respectively. Herbage allowance was 29 ± 6 kg of DM/d per cow. The chemical composition

Table 3. Total diet composition of the TMR and the two pasture-based diets supplemented with concentrates based on ground corn (PCorn) or partially replacing corn with Ca salts of FA (PFat).

	Treatment ¹		
	TMR	PCorn	PFat
DM, %	40.8	33.0	31.5
CP, % of DM	16.5	16.9	17.0
NDF, % of DM	30.6	28.6	29.7
EE, % of DM	4.5	6.1	8.1
NE _L , Mcal/kg of DM ²	1.45	1.70	1.76

¹Total diet composition calculated from nutrient composition (Table 2) and DMI (Table 4).

²Estimated according NRC (2001).

and IVDMD of pasture, TMR and concentrates are presented in Table 2. The estimated chemical composition for the total diet of each treatment is presented in Table 3.

The NDF concentration of pasture (Table 2) was below those considered typical for well-managed cool season pastures (Muller and Fales, 1998). The CP content of the pasture was in the range of 15 to 25% proposed by Minson (1990) to obtain high forage digestibility. The pasture had a higher content of ether extract (6.1%) compared to the values reported by Schroeder et al. (2002), although in the normal range (3 to 8%) for high quality pastures (Harfoot, 1981). Total ether extract content in the diet of both grazing treatments (6 and 8% in PCorn and PFat, respectively, Table 3) was in the range considered to theoretically maximize efficiency of nutrient utilization and minimize negative effects on ruminal digestion (Palmquist, 1988). The high quality of the pasture used (Table 2), and the supplementation with corn-based concentrates (Table 1) determined that total energy concentration were 17 and 21% higher on the PCorn and PFat treatments than on the TMR treatment (Table 3).

Dry matter and nutrient intakes are shown in Table 4. No significant differences were found in total DMI among the treatments (22.4 kg/d), or in pasture DMI between the grazing treatments (18.5 kg/d). Previous studies have reported lower DMI for dairy cows fed only pasture (Kolver and Muller, 1998) or pasture plus concentrate (Bargo et al., 2002) when compared with cows fed a TMR. In this study, pasture DMI was estimated using Cr₂O₃ as indigestible marker and it has been reported that this method overestimated DMI (Bargo et al., 2002). A possible overestimation in pasture DMI could explain the higher body reserve mobilization observed in grazing cows (Table 5). However, total DMI estimated in grazing cows was not higher (Table 4) than that recorded on TMR cows (estimated by difference between individual feeding and refusals). Besides, the same DMI (Table 4) also agreed with the similar DM and NDF content of the TMR and pasture-based diets (Table 3). Concentrate DMI tended ($P < 0.07$) to be lower on the PFat than on the PCorn treatment (4.3 vs. 3.1 kg/d, Table 4), because the amount of concentrate offered (Table 1) was lower on the PFat treatment (5.6 vs. 6.7 kg/d) to provide a similar energy intake by the supplements. Concentrate refusal was not different between the two grazing treatments (2.1 and 2.5 kg/d for the PCorn and PFat treatments, respectively). The high refusal of the concentrate for both treatments could be related to the inclusion of fish meal (Table 1; Husein and Jordan, 1991) and the limited time provided for cows to consume the concentrate (King et al., 1990). Based on the intake of concentrate for the PFat group (Table 4), the total intake of CSFA was 433 $\pm$ 85 g/d (mean $\pm$ SD).

Ruminal Digestion

Parameters of ruminal fermentation on the three treatments are shown in Table 6. The ruminal pH fluctuated

Table 4. Dry matter and nutrient intake of dairy cows fed a TMR or grazing pasture supplemented with concentrates based on ground corn (PCorn) or partially replacing corn with Ca salts of FA (PFat).

	Treatment			SEM	$P <$
	TMR	PCorn	PFat		
DMI, ¹ kg/d					
Pasture	—	18.6	18.4	2.42	0.95
Concentrate	—	4.3	3.1	0.41	0.07
Total	23.7	22.9	21.5	2.05	0.64
DMI, %BW	3.9	4.0	3.7	0.39	0.83
F:C ²	59:41 ^a	84:16 ^b	85:15 ^b	1.57	0.01
CP intake, kg/d	3.9	3.8	3.7	0.35	0.82
NDF intake, kg/d	7.2	6.5	6.4	0.64	0.46
NDF intake, % BW	1.2	1.2	1.1	0.13	0.76

^{a,b}Least square means in the same row with different superscripts differ ($P < 0.05$).

¹Determined on wk 4 and 5 by difference between offered and refused on TMR cows and using Cr₂O₃ in the grazing cows.

²Forage to concentrate ratio (DM basis).

Table 5. Ruminal fermentation parameters and NDF disappearance of dairy cows fed a TMR or grazing pasture and supplemented with concentrates based on ground corn (PCorn) or partially replacing corn with Ca salts of FA (PFat).

	Treatment			SEM	P <
	TMR	PCorn	PFat		
pH	5.60	5.73	5.59	0.07	0.39
NH ₃ -N, mg/dl	12.5	10.3	11.3	1.42	0.56
VFA, mmol/L					
Total	117.9	124.3	122.3	4.91	0.65
Acetate (A)	67.8	69.8	68.2	2.59	0.86
Propionate (P)	25.6	26.4	25.9	1.13	0.90
Isobutyrate	3.41	3.48	3.38	0.19	0.92
Butyrate	15.1 ^b	18.9 ^a	18.8 ^a	1.02	0.02
2-Methyl butyrate	0.59 ^a	0.26 ^b	0.29 ^b	0.06	0.01
Isovalerate	3.23	3.16	3.05	0.19	0.77
Valerate	2.10	2.39	2.68	0.18	0.09
A:P	2.66	2.67	2.66	0.07	0.99
Standard NDF disappearance, ¹ %					
After 9 h in rumen	6.2	4.4	6.8	0.99	0.26
After 16 h in rumen	11.3	10.6	15.0	2.41	0.49
After 48 h in rumen	35.0	34.8	33.9	6.55	0.99

^{a,b}Least square means in the same row with different superscripts differ ($P < 0.05$).

¹From plant material (*Agropyrum elongatum* L.).

tuations over a 24-h period were not affected by treatments ($P > 0.10$; Figure 1). For most of the sampling times, ruminal pH was below 6.0 in the three treatments. Mean ruminal pH (5.64) did not differ among treatments (Table 6). Total VFA (121.5 mmol/L) and NH₃-N concentration (11.4 mg/dl) as well as acetate/propionate ratio (2.66) did not differ among treatments (Table 6). These results agreed with the similar content of NDF and CP in the total diet of the TMR and the two grazing treatments that averaged 29.6 and 16.8%, respectively (Table 3).

Fat supplements, mainly those containing unsaturated FA, have been associated with negative effects on ruminal fiber digestion due to an inhibition on microbial activity of cellulolytic and methanogenic microorganisms (Palmquist, 1988). When ruminal digestion is affected by fat feeding, a reduction in total VFA production and a lower acetate/propionate ratio may be expected. Although CSFA are ruminally inert (Chalupa et al., 1986), rumen pH is generally low when feeding high quality pasture, which could result in the disassociation of part of the Ca and long-chain FA (Chalupa

Table 6. Milk production and composition of dairy cows fed a TMR or grazing pasture and supplemented with concentrates based on ground corn (PCorn) or partially replacing corn with Ca salts of FA (PFat).

	Treatment ¹			SEM	P <
	TMR	PCorn	PFat		
Milk, kg/d	20.2	19.2	20.2	0.65	0.43
Milk fat					
%	3.91 ^a	3.45 ^b	2.56 ^c	0.20	0.01
kg/d	0.76 ^a	0.67 ^b	0.52 ^c	0.03	0.01
4%FCM, kg/d	19.5 ^a	17.8 ^{ab}	16.1 ^b	0.63	0.01
Efficiency					
kg 4%FCM/kg DMI	0.89	0.71	0.84	0.08	0.35
Milk total protein					
%	3.70 ^a	3.49 ^b	3.41 ^b	0.05	0.01
kg/d	0.75	0.67	0.70	0.03	0.13
Protein:fat ratio	0.95 ^b	1.01 ^a	1.33 ^a	0.05	0.01
Lactose					
%	5.06	4.88	4.79	0.09	0.12
kg/d	1.03	0.94	0.99	0.05	0.36

^{a,b,c}Least square means in the same row with different superscripts differ ($P < 0.05$).

¹Least squares means of the data recorded during the last 14 d of experimental period and adjusted using the data from pre-trial period as covariate.

Figure 1. Daily variations in ruminal pH of dairy cows fed a TMR (▲) or grazing pasture and supplemented with concentrates based on ground corn (PCorn; ●) or partially replacing corn with Ca salts of FA (PFat; □). Black arrows indicate the moment of the day when the PCorn and PFat cows received the concentrate and the white arrow time of the day when the TMR cows were fed. Vertical brackets represent standard error. Treat ($P < 0.39$) Hours ($P < 0.01$) Treat $\times$ hours ($P < 0.01$).

et al., 1986). Although ruminal pH was below 6 for the most part of the 24-h period (Figure 1), neither total VFA, acetate/propionate ratio, pasture NDF digestion, nor the ruminal degradation of the standard NDF differed between the PCorn and PFat treatments (Table 6). These results suggest that the CSFA were not dissociated or the degree of dissociation was not important enough to affect the ruminal digestion of grazing cows.

The kinetics of NDF disappearance of the corn silage of the TMR treatment and the pasture of the two grazing treatments is shown in Figure 2. The NDF of the corn silage had a lag time of 6.0 h and a slow rate of degradation (2.8%/h). The NDF of the pasture had no

Figure 2. Curves of NDF disappearance of the corn silage fed to dairy cows on a TMR (▲) or of the pasture grazed by cows supplemented with concentrates based on ground corn (PCorn; ●) or partially replacing corn with Ca salts of fatty acids (PFat; □).

lag time and a faster degradation rate (5.2 %/h). The high quality of the pasture used in the present study (Table 2) and the high level of ruminal digestion of the pasture NDF (Figure 2) likely explains the similar DMI and ruminal environment among treatments (Table 6) even though the forage:concentrate ratio was higher for the cows on pasture (85:15 vs. 59:41, Table 4). Although ruminal NDF digestion could be potentially affected by adding fat to the diet, NDF disappearance of pasture did not differ between the two grazing treatments, indicating that NDF digestion was not affected by replacing corn with CSFA.

Milk Production and Composition, BW, BCS, and Plasma Metabolites

Milk production did not differ among treatments and averaged 19.9 kg/d (Table 7). Previous studies comparing TMR with pasture-based diets have reported lower milk production by dairy cows in midlactation grazing pastures with White et al. (2001), Agenäs et al. (2002), and Bargo et al. (2002) or without concentrate supplementation (Auldist et al., 2002). The similar DM and nutrients intake (Table 4), the relatively low production level and genetic merit of the cows used (Table 7), the short length of the experimental period (5 wk), and the increase in BW on the TMR treatment and BCS losses on the grazing treatments (Table 5) may explain the similar milk production among treatments (Table 7).

The 4% FCM production was lower on the PFat treatment than on the TMR treatment (16.1 vs. 19.5 kg/d, Table 7). This result was related to the significant milk fat depression (MFD) on the PFat treatment compared with the TMR treatment (3.91 vs. 2.56%, Table 7) and not to a lower DMI or milk production. The progress of milk fat percentage and yield for the three treatments during the 5 wk of experiment are shown in Figures 3 and 4, respectively. After 2 wk on pasture, grazing cows had significantly lower milk fat percentage and yield than TMR cows (Figures 3 and 4). At the end of the experimental period, cows on the PCorn treatment had a significantly lower milk fat percentage than the TMR treatment (3.45 vs. 3.91%), but the depression was not as severe as it was on the PFat treatment (Table 7; Figure 3).

Milk fat depression in dairy cows on pasture-based diets has been previously reported (Kolver and Muller, 1998; White et al., 2001; Bargo et al., 2002) and associated with the high digestibility of the fiber in pasture (NRC, 2001). In this study, the ruminal digestion of the NDF of the pasture was higher and faster than that of the NDF of the corn silage in the TMR (Figure 2); however, this did not result in changes on ruminal pH (Figure 1) nor VFA concentration (Table 6). Bauman and

Table 7. Body weight, BCS, and plasma metabolite concentration of dairy cows fed a TMR or grazing pasture and supplemented with concentrates based on ground corn (PCorn) or partially replacing corn with Ca salts of FA (PFat).

	Treatment			SEM	P <
	TMR	PCorn	PFat		
BW, kg					
Initial	585	581	594	22	0.90
Final	609	575	584	23	0.57
Change	23 ^a	-6 ^b	-10 ^b	4.6	0.01
BCS, 1 to 5					
Initial	2.63	2.57	2.69	0.14	0.84
Final	2.73	2.41	2.54	0.11	0.16
Change	0.10 ^a	-0.17 ^b	-0.16 ^b	0.06	0.01
Plasma metabolites ¹					
Glucose, mg/dl	66.2	70.0	70.6	1.81	0.19
PUN, ² mg/dl	18.9 ^b	26.3 ^a	24.5 ^{ab}	2.11	0.05
Triacylglycerides, mg/dl	119.3 ^b	159.0 ^a	152.2 ^a	5.74	0.01
NEFA, μ eq/L	348.6	289.1	311.0	50.5	0.71

^{a,b}Least square means in the same row with different superscripts differ ($P < 0.05$).

¹Samples taken on wk 5.

²Plasma urea nitrogen.

Griinari (2001) have suggested that MFD was related to the direct action on the mammary gland of specific FA isomers derived from the ruminal metabolism of dietary FA. Recently, Mackle et al. (2003) observed a marked MFD in pasture-fed cows receiving abomasal infusion of a CLA mixture. Baumgard et al. (2002) reported that *trans*-10, *cis*-12 CLA could be the main isomer responsible for the reduction in de novo FA synthesis and the expression of genes encoding key enzymes involved in uptake, transport, and synthesis of FA in mammary gland. Although in the present study the concentration of *trans*-10, *cis*-12 CLA in milk was not detected, the MFD seemed to be related to the in-

crease in CLA content (Table 8) rather than to negative effects of CSFA on ruminal NDF digestion (Table 6). The identification of those specific FA that may be responsible for the MFD in grazing cows has not been determined.

The changes of milk total protein percentage for the three treatments during the pretrial period (wk 0) and the 5 wk of experiment is shown in Figure 5. Milk protein concentration was higher on the TMR treatment than on the two grazing treatments (3.70 vs. 3.45%), but milk protein yield did not differ among treatments (0.71 kg/d; Table 7). These results agree

Figure 3. Evolution in milk fat percentage of dairy cows fed a TMR (▲) or grazing pasture and supplemented with concentrates based on ground corn (PCorn; ●) or partially replacing corn with Ca salts of FA (PFat; □). Vertical brackets represent standard error. **Figure 4.** Evolution in milk fat yield of dairy cows fed a TMR (▲) or grazing pasture and supplemented with concentrates based on ground corn (PCorn; ●) or partially replacing corn with Ca salts of FA (PFat; □). Vertical brackets represent standard errors.

Table 8. Fatty acid (FA) profile in milk fat of dairy cows fed with a TMR diet or grazing pasture and supplemented with concentrates based on ground corn (PCorn) or partially replacing corn with Ca salts of FA (PFat)¹.

FA, g/100 g FA	Pre-trial period					Experimental period wk 2					Experimental period wk 5				
	Treatment			SEM	<i>P</i> <	Treatment			SEM	<i>P</i> <	Treatment			SEM	<i>P</i> <
	TMR	PCorn	PFat			TMR	PCorn	PFat			TMR	PCorn	PFat		
C4:0	4.54	4.13	3.98	0.23	0.22	4.63	4.71	4.73	0.18	0.91	4.17	3.26	4.21	0.30	0.09
C6:0	2.75	2.46	2.51	0.11	0.16	2.81 ^a	2.53 ^a	1.98 ^b	0.10	0.01	2.46 ^a	2.12 ^{ab}	1.85 ^b	0.12	0.01
C8:0	1.70	1.53	1.61	0.07	0.24	1.65 ^a	1.38 ^b	0.94 ^c	0.07	0.01	1.45 ^a	1.33 ^a	0.92 ^b	0.07	0.01
C10:0	3.92	3.67	3.99	0.24	0.59	3.69 ^a	2.88 ^b	1.77 ^c	0.20	0.01	3.81 ^a	3.40 ^a	1.88 ^b	0.26	0.01
C12:0	4.50	4.43	4.90	0.36	0.55	4.19 ^a	3.14 ^b	1.97 ^c	0.23	0.01	3.07 ^{ab}	3.74 ^a	2.09 ^b	0.39	0.02
Total short-chain	12.9	11.8	12.1	0.45	0.22	12.8 ^a	11.5 ^a	9.42 ^b	0.43	0.01	11.9 ^a	10.4 ^b	8.85 ^b	0.61	0.01
C14:0	12.0	11.9	12.5	0.58	0.72	11.7 ^a	9.99 ^b	7.03 ^c	0.45	0.01	11.4 ^a	10.8 ^a	7.31 ^b	0.33	0.01
C14:1	1.02	1.08	1.21	0.11	0.41	1.04 ^a	0.94 ^a	0.58 ^b	0.11	0.02	1.01 ^{ab}	1.15 ^a	0.84 ^b	0.07	0.05
C14:0/C14:1 ratio	12.1	11.0	10.7	1.03	0.55	11.7 ^b	10.9 ^b	15.7 ^a	1.50	0.08	11.4	9.47	9.46	0.68	0.11
C16:0	27.6	30.7	29.1	2.04	0.59	28.4 ^a	25.1 ^b	22.4 ^c	0.65	0.01	27.3 ^a	24.4 ^b	20.6 ^c	0.70	0.01
C16:1	1.14	1.36	1.45	0.14	0.31	1.33	1.57	1.29	0.09	0.11	1.16 ^b	1.54 ^a	1.15 ^b	0.11	0.04
C16:0/C16:1 ratio	25.7	23.9	20.5	2.90	0.38	22.3 ^a	16.4 ^b	17.9 ^b	1.40	0.02	25.1 ^a	16.7 ^b	19.5 ^{ab}	2.26	0.05
Total medium-chain	59.4	61.2	60.9	2.43	0.85	59.0 ^a	52.4 ^b	49.4 ^b	1.82	0.01	56.5 ^a	47.6 ^b	41.0 ^c	2.01	0.01
C18:0	12.9	12.0	11.4	1.24	0.66	12.0 ^b	12.6 ^{ab}	14.6 ^a	0.71	0.04	13.3 ^a	10.5 ^b	13.2 ^a	0.62	0.01
C18:1	21.8	21.1	21.0	1.37	0.91	24.1 ^b	29.0 ^a	32.4 ^a	1.14	0.01	23.8 ^b	24.9 ^b	30.6 ^a	1.03	0.02
C18:0/C18:1 ratio	0.59	0.59	0.55	0.07	0.86	0.50	0.44	0.46	0.03	0.37	0.56 ^a	0.42 ^b	0.44 ^b	0.03	0.01
C18:2	2.59	2.10	2.37	0.26	0.43	2.53 ^b	2.21 ^b	5.20 ^a	0.42	0.01	2.17 ^b	2.10 ^b	4.75 ^a	0.24	0.01
CLA ²	0.37	0.49	0.43	0.03	0.07	0.52 ^c	0.80 ^b	1.29 ^a	0.08	0.01	0.41 ^c	1.12 ^b	1.91 ^a	0.10	0.01
C18:3	0.07	0.06	0.04	0.03	0.82	0.30	0.41	0.49	0.13	0.58	0.07 ^b	0.57 ^a	0.65 ^a	0.05	0.01
Others	1.68	1.89	2.09	0.80	0.93	0.42 ^b	1.90 ^{ab}	2.94 ^a	0.62	0.03	2.95 ^b	3.48 ^b	5.73 ^a	0.63	0.01
Total long-chain	38.1	36.1	35.6	2.04	0.64	39.5 ^c	45.2 ^b	54.0 ^a	1.33	0.01	40.1 ^b	39.5 ^b	51.4 ^a	1.31	0.01
Saturated: unsaturated FA ratio	71:27	72:26	71:27	1.81	0.98	70:30 ^a	63:35 ^b	56:41 ^c	1.27	0.01	69:29 ^a	64:32 ^b	53:42 ^c	1.29	0.01

¹Least square means in the same row (within date of sampling) with different superscripts differ (*P* < 0.05).²CLA content tended to be different in the pre-trial period, therefore experimental period data were adjusted by covariate.

Figure 5. Evolution in milk protein percentage of dairy cows fed a TMR (▲) or grazing pasture and supplemented with concentrates based on ground corn (PFCorn; ●) or partially replacing corn with Ca salts of FA (PFCorn; □). Vertical brackets represent standard errors.

with those previously reported (Kolver and Muller, 1998; Bargo et al., 2002) comparing TMR versus pasture-based diets. Although in the present study the DM and CP intake were similar among treatments (Table 4), the lower milk protein percentage in grazing cows could be related to the higher forage:concentrate ratio (Table 4) and the higher dietary fat content (Table 3).

Cows on the two grazing treatments lost BW (−8 kg) and BCS (−0.17), whereas cows on the TMR treatment gained BW (23 kg) and BCS (0.10; Table 5). However, the concentration of NEFA determined on wk 5 was not affected by treatments and averaged 316.2 μ Eq/L (Table 5).

Plasma glucose concentration was not affected by treatments (68.9 mg/dl, Table 5). Plasma urea nitrogen did not differ between the PFCorn and PFCorn treatments but was significantly higher on the grazing treatments compared to the TMR treatment (25.4 vs. 18.9 mg/dl, Table 5). These results may support the hypothesis that the higher forage:concentrate ratio of pasture-based diets increased the losses of highly degradable N of the pasture and explain the lower milk protein concentration (Figure 5). The concentration of plasma triacylglycerides was significantly higher on the grazing treatments than on the TMR treatment (119.3 vs. 156.1 mg/dl), which can be associated with the high level of ether extract in the pasture (6.1%, Table 2) resulting in a higher level of ether extract in the total diet on the two grazing treatments than in the TMR treatment (6.1 and 8.1 vs. 4.5%, Table 3).

Milk FA Composition and CLA Content

The FA composition of milk for the pretrial (wk 0) and experimental periods (wk 2 and 5) is shown in Table

8. During the pretrial period, all the cows were fed the same TMR, resulting in no differences among treatments ($P > 0.10$). On wk 2, the cows fed the PFCorn and PFCorn treatments had lower percentage of medium-chain FA (C12 to C16), and a higher percentage of long-chain FA ($> C18$) compared with the TMR treatment, with the biggest differences resulting from the PFCorn treatment. On wk 5, the PFCorn treatment had lower percentage of short- (11.9 vs. 10.4 g/100 g FA) and medium-chain (56.5 vs. 47.6 g/100 g FA) FA, and a higher C18:3 percentage (0.65 vs. 0.07 g/100 g FA) compared with the TMR treatment and the pretrial period. High milk content of C18:3 is frequently observed in cows consuming high-quality pastures because this is the most abundant FA in forages (Dewhurst et al., 2001; Mackle et al., 2003). The partial replacement of corn by CSFA in the concentrate resulted in a marked reduction in short (C4 to C12) and medium-chain FA and increase in total long-chain FA. When milk fat composition on wk 5 was analyzed on molar basis (data not presented), the production (mmol/d) of $< C16$, C16 plus C16:1, and $> C16$ FA were −13.0, −20.3, and −13.1% lower on the PFCorn treatment compared with the TMR treatment. On the PFCorn treatment, these differences compared with the TMR treatment were −48.9, −40.9, and 0% of $< C16$, C16 plus C16:1, and $> C16$ FA, respectively. These results may suggest that on the PFCorn treatment, de novo synthesized and preformed FA contributed in a similar magnitude to the reduction in milk fat percentage, whereas on PFCorn treatment this reduction was primarily associated with a reduction in the de novo synthesized FA. The reduction on short- and medium-chain FA on the PFCorn treatment could be the result of the dissociation of part of CSFA in the rumen and a posterior isomerization into *trans*-10, *cis*-12 CLA, which inhibits milk fat synthesis (Baumgard, et al., 2002). Larger mobilization of body reserves, showed by the loss of BW and BCS (Table 5), could also provide C16:0 and C18:0 to mammary gland (Agenäs et al., 2002); however, no differences in NEFA concentration were detected (Table 5) and both FA were significantly lower on the PFCorn with no differences in C18:0 on the PFCorn (Table 8). Total saturated:unsaturated FA ratio was lower on the PFCorn treatment compared with the TMR treatment (69:29 vs. 64:32), with the PFCorn treatment having the lowest saturated:unsaturated ratio (53:42, Table 8). Reducing the saturated FA content and increasing the long-chain unsaturated FA have been associated with increased healthfulness of milk (Pariza, 1999).

The change of CLA content in milk over time is shown in Table 8. The CLA content tended ($P < 0.07$) to be higher during the pretrial period on the PFCorn treatment (Table 8), therefore the CLA content for wk 2 and

5 were adjusted using the pretrial period CLA content as covariate. After 2 wk on pasture and fed the experimental concentrates, CLA content was 1.5- and 2.5-fold higher on the PCorn and PFat treatments than on the TMR treatment (Table 8). The rates of increase in CLA concentration between the pretrial period and wk 2 of the experimental period were 0.09, 0.17, and 0.41 g of CLA/100 g of FA per week ($P < 0.01$) for the TMR, PCorn, and PFat treatments, respectively. The CLA content was 2.7- and 4.7-fold higher on the PCorn and PFat treatments at wk 5, respectively, compared with the TMR treatment (Table 8). The rates of increase between wk 2 and 5 were -0.06, 0.10, and 0.24 g CLA/100 g FA per week ($P < 0.01$) on the TMR, PCorn, and PFat treatments, respectively. Summarizing four previous studies (Timmen and Patton, 1988; Kelly et al., 1998; White et al., 2001; Auldist et al., 2002), milk CLA concentration was 2.3-fold (range 1.2- to 5.0-fold) higher for dairy cows fed pasture compared with cows fed TMR. The 2.7-fold increase observed on the PCorn treatment (Table 8) was similar to the mean of those six previous studies. The intake of 433 g/d of CSFA on the PFat treatment resulted in a milk CLA content of 1.91 g/100 g FA compared to 0.41 g/100 g of FA for the TMR treatment (Table 8). This 4.7-fold increase was similar to the highest increase (5.0-fold) reported for grazing cows (Timmen and Patton, 1988), although in this study the content of CLA was higher than that previous study (1.91 vs. 1.34 g/100 g FA). In agreement with previous studies (Kelly et al., 1998; White et al., 2001), there was more individual variation in the milk CLA content in grazing cows supplemented with corn-based concentrates (CV = 35%) than in cows fed TMR (CV = 19%; Table 8). In our study, the partial replacement of corn by CSFA in the concentrate supplemented to grazing cows did not only increase CLA content, but also reduced the individual variation in CLA content (CV = 11%; Table 8).

The increase in CLA content could be a result of an increased synthesis in the mammary gland. The high content of C18:3 in high quality pastures (40 to 80 g/100 g FA; Dewhurst et al., 2001) may increase the ruminal production of the *trans*-11 C18:1 (vaccenic acid). In addition, C18:3 may reduce the conversion of *trans*-11 C18:1 to C18:0, which is the rate-limiting step in biohydrogenation of FA (Griinari and Bauman, 1999) resulting in large amounts of *trans*-11 C18:1 absorbed postruminally in grazing cows. The subsequent desaturation of *trans*-11 C18:1 to CLA by the enzyme Δ^9 -desaturase in mammary gland could explain the higher CLA content in milk reported in dairy cows on pasture-based diets (Griinari and Bauman, 1999). After 5 wk on trial, the ratio of C18:0 to C18:1 was lower (59:37 vs. 69:29; Table 8) on the two grazing treatments compared

with the TMR treatment, suggesting that the activity and/or expression of the mammary gland enzyme Δ^9 -desaturase may have been higher in those two treatments. The effects of fat supplementation on CLA content in grazing dairy cows have not been well documented. In this study, the CSFA could have been partially disassociated in the rumen because of the low ruminal pH (Figure 1), and part of the FA could have been biohydrogenated by ruminal microorganisms generating additional CLA and vaccenic acid. For CSFA of palm oil, Klusmeyer and Clark (1991) found that 33 to 57% of unsaturated C18 was biohydrogenated in rumen. Besides, the increase in unsaturated long-chain FA concentration in the rumen may also decrease the rate of biohydrogenation of other dietary FA (Chalupa et al., 2001) increasing the amount of CLA and its precursor (vaccenic acid) in the rumen. Recently, Kolver et al. (2002) reported that supplementation with C18:2 or C18:3 FA to high-quality pastures increased the ruminal content of CLA by 15- and 5-fold and *trans* vaccenic acids by nine- and fourfold, respectively. Regardless of which physiological mechanisms were involved in the increase of CLA content with CSFA supplementation (Table 8), feed CSFA seemed to maximize the effects of pasture-based diets.

CONCLUSIONS

Cows on the two pasture-based diets had MFD compared with the cows on the TMR, which was accentuated by the partial replacement of corn by CSFA. Grazing cows had a reduction in the content of de novo synthesized FA (C4 to C14) and an increased content of C18:3 and CLA. The partial replacement of corn by CSFA in the concentrate supplemented to the grazing cows accentuated the decrease of medium-chain FA, and the increase of long-chain FA and CLA content compared with the TMR treatment. The reduction in the de novo synthesized FA and milk fat percentage was likely associated with a direct effect of some isomer of CLA on the mammary gland rather than associated with negative effects of CSFA on NDF ruminal digestion. Partial replacement of corn grain by CSFA in a pasture-based diet seemed to be an effective strategy for maximizing CLA and unsaturated FA content in milk enhancing its nutritional and health value; however, milk fat percentage was depressed. A better identification of specific FA related to MFD and longer-term studies considering the possible beneficial effects of MFD on milk production, body reserved mobilization, and reproductive performance in grazing cows are needed.

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