

## Partial Replacement of Corn Grain by Hydrogenated Oil in Grazing Dairy Cows in Early Lactation

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### ABSTRACT

Thirty-six grazing dairy cows were used to determine milk production and composition, and dry matter and energy intake when corn grain was partially replaced by hydrogenated oil in the concentrate. Four additional cows, each fitted with a ruminal cannula, were used in a crossover design to evaluate effects of supplemental fat on rumen environment and pasture digestion. All cows grazed mixed pastures with an herbage allowance of 30 kg DM/cow per day. The control group was fed a concentrate containing corn grain (4.49 kg dry matter/cow per day) and fishmeal (0.37 kg dry matter/cow per day), whereas the other group (fat) received a concentrate containing corn grain (2.87 kg dry matter/cow per day), fishmeal (0.37 kg dry matter/cow per day) and fat (0.7 kg dry matter/cow per day). The fat was obtained by hydrogenation of vegetable oils (melting point 58 to 60°C, 30.3% C<sub>16:0</sub>, 34.9% C<sub>18:0</sub>, 21.8% C<sub>18:1</sub>, 3.3% C<sub>18:2</sub>). Supplemental fat increased milk production (control = 23.7 vs. fat = 25.0 kg/cow per day), fat-corrected milk (control = 22.5 vs. fat = 24.5 kg/cow per day), milk fat content (control = 3.64% vs. fat = 3.86%) and yields of milk fat (control = 0.86 vs. fat = 0.97 kg/cow per day) and protein (control = 0.74 vs. fat = 0.78 kg/cow per day). Milk percentages of protein, lactose, casein, cholesterol, and urea nitrogen were not affected. Pasture DMI and total DMI of pasture and concentrate and estimated energy intake were unchanged. No differences in loss of body weight or body condition score were detected. Plasma concentrations of nonesterified fatty acids, somatotropin, insulin, and insulin-like growth factor were not affected by supplemental fat. Concentrations of plasma triglyceride and total cholesterol were increased by supplemented fat, and no changes in plasma glucose and urea nitrogen were observed. The acetate-to-propionate

ratio was higher in rumen fluid of cows that consumed fat (fat = 3.39 vs. control = 3.27). In situ pasture NDF degradation was not affected. The partial replacement of corn grain with fat improved the productive performance of early-lactation cows grazing spring pastures. No negative effects of supplemental fat on ruminal fiber digestion were detected.

**(Key words:** hydrogenated oil, grazing, milk composition, ruminal digestion)

**Abbreviation key:** C2:C3 = acetate:propionate, **control** = no supplemental fat, **fat** = 0.73 kg/d of hydrogenated oil, **FCM** = 4% fat-corrected milk, **IVDMD** = in vitro DM digestibility, **J-M** = jugular-mammary differences, **ME** = metabolizable energy, **PUN** = plasma urea nitrogen, **SFD** = subcutaneous fat depth, **WSC** = water-soluble carbohydrates.

### INTRODUCTION

Early-lactating cows cannot consume enough dietary energy to match their requirements for milk production, and the energy balance of the cow becomes negative. Under grazing conditions, this negative energy balance may even be greater as the consequence of a lower DMI of cows compared with a TMR diet and the higher requirement over maintenance because of higher levels of activity (Muller and Fales, 1998). It has been described that cows fed only pastures produced less milk per day and lost substantially more BCS in early lactation compared with cows fed a nutritionally balanced ration, suggesting the need for energetic supplementation (Kolver and Muller, 1998; Schroeder et al., 2003). High quality pastures usually have low ratios of non-structural carbohydrates to N, which could lead to sub-optimal performance of the cows due to an impaired microbial protein synthesis (Schroeder et al., 2003). In this context, not only the amount but also the chemical nature of the energy supplemented to grazing cows needs to be considered.

In many countries that produce milk from pasture, the main source of supplemental energy used is nonfi-

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ber carbohydrates from cereal grains. When considerable amounts of these grains are fed to lactating dairy cows in combination with pastures of highly fermentable NDF, ruminal pH may decrease, resulting in a decrease in milk fat content (Schroeder et al, 2003).

Although in confinement systems milk production is often increased after fat supplementation (Schingoethe and Casper, 1991), the response in pasture-based diets is still poorly described (Schroeder et al., 2003). Besides, the response to nonfermentable energy supply may be different in grazing conditions because the specific nutrients that limit milk yield may not be the same (Schroeder et al., 2002). Supplemental fat can be used to increase the energy density of diets and substitute, in part, for cereal grains, although experimental data on effects of substitution of fermentable energy with rumen inert energy are scarce. Although the effect of feeding fat to dairy cows on milk production has been examined under many nutritional circumstances (Palmquist, 1984; Gagliostro and Chilliard, 1992; Wu and Huber, 1994), in this investigation the diets were based on pasture and were isocaloric, with starch substituted for fat. **[Au: previous sentence Ok as edited?]** When hydrogenated oil was added to a basal concentrate of grazing dairy cows in early lactation, the production of FCM and milk fat secretion were increased and pasture fiber digestion was not affected (Schroeder et al., 2002).

Although the hormone profile may play an important role in coordinating the partition of dietary fatty acids between milk fat secretion, deposition in adipose tissue, and body lipid mobilization (Palmquist, 1984), the effects of fat addition on plasma hormones are poorly understood in grazing dairy cows.

The objective of this experiment was to determine whether the partial replacement of energy-yielding substrates to ruminal microbes (corn grain) for nonfermentable energy (hydrogenated oil) may improve milk yield in early lactation cows. Because the control and fat supplied the same amount of energy to grazing cows, we examined the opportunity for fat to affect lactation performance when energy intake is held constant. **[Au: preceding sentence OK as edited?]** Effects of fat on milk composition, body condition parameters, plasma metabolites, and hormone concentrations were also studied. The effect of the hydrogenated fat on ruminal digestion was also evaluated in grazing dairy cows.

## MATERIALS AND METHODS

The experiment was conducted at the National Institute of Agricultural Technology (INTA) in Balcarce (37°45'S, 58°18'W), Argentina during the spring of 1999. Thirty-six multiparous Holstein cows (599 ± 49 kg

**Table 1.** Ingredient and nutrient composition of the experimental concentrates.

Ingredient, kg DM/ cow per day	Control	Fat
Ground corn	4.49	2.87
Fishmeal	0.37	0.37
Calcium chloride	0.02	0.02
Fat <sup>1</sup>	0	0.7
Mineral-vitamin mix <sup>2</sup>	0.12	0.12
Total offered	5.00	4.08
Nutrient		
DM, %	90	90.7
CP, % of DM	13.3	12.7
NDF, % of DM	11.0	8.6
Starch, % of DM	61.2	47.8
EE, % of DM <sup>3</sup>	6.7	22.5
NE <sub>L</sub> , <sup>4</sup> Mcal/kg MS	2.03	2.39

<sup>1</sup>Partially hydrogenated oil (95.9% DM). Melting point: 58 to 60°C. Fatty acid composition was: 30.3% C16:0, 34.9% C18:0, 21.8% C18:1, and 3.3% C18:2. Lipid metabolizable energy content (5.18 Mcal/kg DM) was calculated as previously described (Schroeder et al, 2002).

<sup>2</sup>Contained 21% Ca<sub>3</sub>(PO<sub>4</sub>)<sub>2</sub>, 4.6% P<sub>2</sub>O<sub>5</sub>, 6% MgO, 3% molasses, 0.15% Fe SO<sub>4</sub>, 0.4% CuSO<sub>4</sub>, 0.3% Zn SO<sub>4</sub>, 0.04% MnSO<sub>4</sub>, 0.02% CoSO<sub>4</sub>, 0.02% Na<sub>2</sub>SeO<sub>3</sub>, 0.01% I<sub>2</sub>, vitamin A (6,500,000 IU/kg), vitamin D<sub>3</sub> (1,600,000 IU/kg), vitamin E (12,500 IU/kg), and 64.46% excipient.

<sup>3</sup>EE = Ether extract.

<sup>4</sup>According to NRC (2001).

BW) were selected based on parity and milk production measured during the first 70 DIM for the previous lactation and randomly allocated to one of 2 dietary treatments. One group (**control**) received a concentrate based on ground corn grain and fishmeal, whereas in the concentrate fed to the other group (**fat**) part of the corn grain was substituted by partially hydrogenated oil (Table 1).

The concentrates were offered in 2 equal feedings during milking times (0600 and 1600 h) from 15 ± 5 d (mean ± SD) prior to the expected calving date to 75 DIM. As we desired to examine the effect of fat early in lactation (from the first week after calving) and a 2- or 3-wk lag time was observed before milk yield responded to dietary fat in early lactation (Schingoethe and Casper, 1991) the fat supplement was introduced prior to calving. After 75 DIM, all cows were fed the control concentrate for an additional period of 30 d to evaluate the residual effects of fat feeding on milk yield and composition. Intake of concentrate was daily and individually recorded by weighing the amounts offered and refused.

The fat was obtained by hydrogenation of vegetable oils. The hydrogenated fat was deodorized and solidified in flakes that were free of moisture (95.9% DM). The medium size of these particles was 2 to 3 mm, and the thickness about 0.3 mm. The flakes were composed of triglycerides of high melting point (58 to 60°C) that may remain solid at ruminal temperature (39 to 40°C).

The fatty acid composition of the partially hydrogenated oil was C<sub>14:0</sub> (2.4%), C<sub>14:1</sub> plus iso C<sub>15:0</sub> (0.7%), C<sub>15:0</sub> (0.9%), C<sub>15:1</sub> (0.4%), C<sub>16:0</sub> (30.3%), C<sub>16:1</sub> (0.2%), C<sub>17:0</sub> (1.2%), C<sub>17:1</sub> (0.1%), C<sub>18:0</sub> (34.9%), C<sub>18:1</sub> (21.8%), C<sub>18:2</sub> (3.3%), C<sub>18:3</sub> (0.9%), and others (2.9%) as previously described (Schroeder et al., 2002).

All cows grazed together on mixed pastures that contained (DM basis) alfalfa (*Medicago sativa*, 49%), orchardgrass (*Dactylis glomerata* L., 15%), bromegrass (*Bromus catharticus* L., 15%) and perennial ryegrass (*Lolium perenne* L., 21%). Cows had first access to the pasture in a vegetative stage of maturity of the grazed forage, and we tried to maintain it using a daily strip-grazing system. The area of the strip was regulated using a temporary electric fence to achieve an herbage allowance of about 30 kg DM/cow per day to allow a high pasture intake (Minson, 1990). This figure was calculated assuming an optimum herbage allowance of 45 g OM/kg BW (Minson, 1990), the OM content of the pasture (89.1% of DM) and the initial BW of cows that averaged 599 kg.

The cows were moved to a new strip every day, and after grazing each strip was clipped-out of nongrazed forage to about 6 cm to allow a clean and uniform pasture regrowth.

### Samples Collection and Analysis

After an adaptation period of 21 d (-14 to 15 DIM), milk production was measured at each milking daily. Milk samples were collected every 15 d at a.m. and p.m. milkings, composited according to the corresponding volume measured at each milking time and analyzed for fat, total protein, lactose, total solids, and SNF (AOAC 1997) by midinfrared spectrophotometry (Foss 605B Milko-Scan, Foss Electric, Hillerød, Denmark). Milk urea nitrogen (Wiener Lab., Rosario, Argentina; Clinical Chemistry, 18/8, 829-840, 1972) and cholesterol (Colestat, Wiener Lab., Rosario; Clinical Chemistry, 20/4, 470-475, 1974) were determined using commercial enzymatic kits as described in Schroeder et al. (2002). Casein was determined as proposed in AOAC (1997).

Total herbage mass was determined every week by cutting 16 quadrats (0.125 m<sup>2</sup> per quadrat) of pasture samples to ground level. Each sample was dried at 60°C in a forced-air oven. The quality of the grazed herbage was estimated from samples obtained by hand-plucking at random transects every 10 d. Samples of concentrates were collected every 30 d. Pasture and concentrates samples were dried (60°C in a forced-air oven), ground through a 1-mm screen (Wiley mill, Philadelphia, PA), and analyzed for OM, NDF, and ADF (Goering and Van Soest, 1970), CP (AOAC, 1997), in vitro DM digestibility (**IVDMD**) (Tilley and Terry, 1963), water-

soluble carbohydrate (**WSC**) (Morris, 1948), and ether extract (AOAC, 1997).

Pasture DMI was estimated on 9 cows per treatment from 45 to 60 DIM using Cr<sub>2</sub>O<sub>3</sub> as an indigestible fecal marker as previously described (Schroeder et al., 2002). After each milking, cows were dosed twice daily with 3 gelatin capsules containing 2 g of Cr<sub>2</sub>O<sub>3</sub> during a period of 15 d (12 g of Cr<sub>2</sub>O<sub>3</sub>/cow per day). Fecal grab samples were collected after milking on d 10 to 15. Total fecal DM production (kg/d) was estimated by dividing the total Cr dosed (g/d) by the Cr concentration in fecal DM (g/d) determined by absorption spectrophotometry. Fecal DM output due to concentrate was estimated as concentrate intake × (1 - concentrate IVDMD). This quantity was subtracted from the total fecal DM production and the remaining fecal DM material was attributed to pasture. Pasture DMI was calculated as the ratio between fecal DM yield due to pasture and pasture indigestibility (1 - IVDMD). Total energy intake was calculated from DMI of forage and concentrates and their NE<sub>L</sub> content estimated according to NRC (2001). Lipid metabolizable energy (**ME**) content (5.18 Mcal/kg DM) was calculated as proposed in Schroeder et al. (2002), assuming that FA digestion in the total tract was 56.5% for total C<sub>16</sub> and 63.6% for total C<sub>18</sub> (Pantoja et al., 1995).

Cows were weighed on 2 consecutive days after the a.m. milking at the start (3 and 4 DIM), in the middle (30 and 31 DIM), and at the end (74 and 75 DIM) of the period of fat supplementation. An additional record of BW was taken at the end of the residual period (104 and 105 DIM). On the same days, BCS and the subcutaneous fat depth (**SFD**) were also recorded. Body condition score was estimated by 2 independent observers using the 5-point scale (1 = thin to 5 = fat) and SFD was measured between the 12th and 13th ribs using a Pie Medical 480 scanner (Holland). The mean value of the 2 records was used to calculate changes in BW gain, BCS, and SFD.

Blood samples were collected from the jugular vein immediately after the a.m. milking at 15, 30, 45, 60, 75, and 105 DIM. On the days of sampling, cows received the concentrates immediately after bleeding. On d 30, blood was simultaneously drawn from the external mammary abdominal vein to determine jugular-mammary (**J-M**) differences in metabolites and estimate apparent mammary uptake. Blood was collected in tubes containing EDTA (0.342 mol/L, pH 7.2, Wiener Laboratory, Rosario, Argentina), immediately placed on ice and plasma was obtained (2000 × g at 4°C for 10 min) and stored frozen (-24°C) until analysis. Commercial enzymatic kits were used for plasma urea nitrogen (**PUN**) (Wiener Laboratory Clinical Chemistry, 18/8, 829-840, 1972), NEFA (Wako Pure, Chemical Indus-



tries USA, Inc., Dallas, TX), glucose (Wiener Laboratory Clinical Chemistry, 21/12, 1754-1760, 1975), triglyceride (Wiener Laboratory, Clinical Chemistry, 28/10, 2077-2080, 1982) and total cholesterol (Wiener Laboratory; Clinical Chemistry, 20/4, 470-475, 1974) as described in Schroeder et al. (2002).

Plasma IGF-I was measured by radioimmunoassay with previous acid-ethanol extraction as previously described (Schroeder et al., 2002). Insulin-like growth factor-I antibody (UB2-495, Hormone Distribution Program of the NIDDK, National Institute of Diabetes and Digestive and Kidney Diseases, Rockville, MD) was used. The intraassay coefficient of variance was 7% and assay sensitivity was 60 pg/tube. Somatotropin (ST) and insulin were measured in the same samples. Concentration of somatotropin was determined by radioimmunoassay using an anti-ovine antibody as described in (Schoeder et al., 2002). The intraassay coefficient of variance was 7.2% and minimum detectable concentration was 0.8 ng. Insulin was measured by radioimmunoassay using anti-bovine insulin antibody (Sigma, St. Louis, MO) and standard human insulin provided by Laboratories Beta (Buenos Aires, Argentina). Minimum detectable concentration was 0.05 ng.

### Rumen Environment and in Situ Pasture NDF Degradability

The effects of fat supplementation on ruminal environment and parameters of pasture DM and NDF degradation were evaluated using 4 lactating Holstein cows in midlactation fitted with ruminal cannulas in a 2 × 2 crossover design with 15-d periods. These cows received the same treatments and were in a single herd under strip grazing conditions together with nonfistulated cows.

During the 15th day of each experimental period, samples of ruminal content were taken from the ventral rumen at 0 (0600 a.m.), 4, 8, 12, 16, and 20 h after the first sample. Ruminal fluid (200 mL) was obtained by straining through 4 layers of cheesecloth, and pH was measured immediately (Orion portable pH meter 250A, Orion Research Inc., Boston, MA). A sample (100 mL) was acidified with 1 mL of 1 N H<sub>2</sub>SO<sub>4</sub> and frozen (-24 °C). Samples were later thawed and centrifuged at 10,000 × g for 10 min (0°C), and the supernatants were analyzed for concentrations of ammonia nitrogen (NH<sub>3</sub>-N) (Autoanalyser Tecator, model Kjeltac 1030) and VFA.

The VFA were determined in a gas chromatograph (Shimadzu model GC-14) using a gas N<sub>2</sub> as a carrier at a flow rate of 50 mL/min and a glass column (2 m long × 2 mm of internal diameter) packed with 80/120 Carbowax B-DA/4% Carbowax 20 M (Supelco, Inc.,

Bellefonte, PA). Temperatures for the oven, injector port, and detector were 155, 185, and 190°C, respectively.

At the start of each experimental period pasture samples were obtained by hand-plucking and were cut to a final length of 1 cm. The wet material was placed (5 g of DM/bag) in Dacron bags (15.5 × 7.5 cm, 52-μm average pore size, Ankom, Fairport, NY). The bags were incubated in the ventral sac of the rumen by duplicate for 0, 4, 8, 12, 16, 20, 26, 32, 40, and 48 h. After incubation, the bags were rinsed in a pipette washer for 1 h and then hand-washed with cold tap water. Bags were squeezed until the water was clear and then oven dried at 60°C until constant weight. The residues from each bag were weighed, ground through a 1-mm screen, pooled within cow for each time of incubation and analyzed for DM and NDF content.

Kinetic parameters of ruminal DM degradation were estimated with the equation proposed by Ørskov and McDonald (1979):  $D = A + B(1 - e^{-kdt})$ , 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 of NDF was estimated as A + B. The effective degradation of DM was calculated with the following equation: effective degradation =  $A + B(kd/(kd + kp))$ , where kp = rate of passage (assumed to be 0.03, 0.05, and 0.07/h) (Ørskov and McDonald, 1979).

Kinetics parameters of NDF degradation were estimated with the equation proposed by Mertens and Lofton (1980):  $R = D e^{-k(t-L)} + U$ , where R = cell wall residue (at time after incubation = t), D = digestible fraction, k = digestion rate constant, L = lag time, and U = indigestible fraction. The effective degradation of NDF was calculated as: effective degradation =  $(D/100) * ((k/(k + kp)) * ((e^{-(kp/100)*L}))$ .

### Statistical Analysis

Milk production and composition, changes in BW, BCS, SFD, and plasma metabolite and hormone concentration were evaluated by the GLM procedure of SAS (1996) using the following model:

$$Y_{ijk} = \mu + T_i + A_{(ij)} + D_k + (TxD)_{ik} + \varepsilon_{ijk},$$

where:

$Y_{ijk}$  = the dependent variable,

$\mu$  = overall mean,

$T_i$  = treatment effects,

$A_{ij}$  = random effects of animal within treatments,

$D_k$  = effects of sampling date or time,  
 $(TxD)_{ik}$  = interaction effects of treatment and sampling date or time, and  
 $\varepsilon_{ijk}$  = the residual error associated with the  $ijk$  observation.

As no interaction between treatment and week of lactation was detected ( $P > 0.05$ ) for any parameter measured, milk yield and composition was also analyzed as a completed randomized design introducing data obtained over the early phase (7 to 17 DIM) of the previous lactation as the covariate. The model used was :

$$Y_{ij} = \mu + T_i + Cov + \varepsilon_{ij}$$

where:

$Y_{ij}$  = the dependent variable (average values over the first 75 DIM),  
 $\mu$  = overall mean,  
 $T_i$  = treatment effects,  
Cov = covariate (milk yield and composition over 7 to 17 DIM of the previous lactation), and  
 $\varepsilon_{ij}$  = residual error.

Data from DMI and J-M differences in metabolites were analyzed with the GLM procedure of the SAS (1996) program using the following model:

$$Y_{ij} = \mu + T_i + \varepsilon_{ij} ,$$

where:

$Y_{ij}$  = the dependent variable,  
 $\mu$  = overall mean,  
 $T_i$  = treatment effects, and  
 $\varepsilon_{ij}$  = residual error.

The rumen parameters were analyzed in a  $2 \times 2$  crossover design using the following model:

$$Y_{ijk} = \mu + T_i + A_{(ij)} + P_k + D_1 + (T)_{il} + \varepsilon_{ijkl}$$

where:

$Y_{ijk}$  = the dependent variable,  
 $\mu$  = overall mean,  
 $T_i$  = treatment effects,  
 $A_{ij}$  = random effects of animal within treatments,  
 $P_k$  = the effects of the experimental period,  
 $H_l$  = effects of hour of sampling,

$(TxH)_{ik}$  = interaction effects of treatment and hour, and  
 $\varepsilon_{ijkl}$  = the residual error associated with the  $ijkl$  observation.

Kinetics parameters of DM and NDF degradation were analyzed in a  $2 \times 2$  crossover using the following model:

$$Y_{ijk} = \mu + T_i + A_j + P_k + \varepsilon_{ijk}$$

where:

$Y_{ijk}$  = the dependent variable,  
 $\mu$  = overall mean,  
 $T_i$  = treatment effects,  
 $A_j$  = random effects of animal within treatments,  
 $P_k$  = the effects of the experimental period, and  
 $\varepsilon_{ijk}$  = the residual error associated with the  $ijk$  observation.

Differences were considered significant with  $P < 0.10$  unless otherwise stated. All results are reported as least square means.

## RESULTS

### Pasture Characteristics

The average value ( $\pm$  standard deviation) of herbage mass in the pregrazing strips was  $2039 \pm 1033$  kg DM/ha, and the average herbage allowance obtained was  $30.4 \pm 2.5$  kg DM/cow per day during the trial. Values for the chemical composition of the forage apparently consumed by cows across the entire study are shown in Table 2. Pasture CP, NDF, and ether extract composition for every 10 d is shown in Figure 1.

### Milk Production and DMI

Yield of FCM was greater for cows receiving the concentrate containing fat (Figure 2). An interaction between treatment and week of lactation was not detected. Peak yield of FCM occurred at wk 4 of lactation (control = 23.7; fat = 25.6 kg/cow per day, Figure 2).

During the first 75 DIM, the fat supplemented cows produced more milk (+5.5%) and more FCM (+9.1%) compared with control cows. Milk fat content (+6%), milk fat production (+12.8%), and milk protein yields (+5.4%) were higher when hydrogenated oil replaced corn grain in the concentrate (Table 3). Milk composition of protein, casein, lactose, SNF, MUN, and total cholesterol were not affected by fat supplementation (Table 3).

**Table 2.** Nutrient composition of spring pastures during the experiment.<sup>1,2</sup>

	% DM	% of DM						
		OM	IVDMD	NDF	ADF	CP	WSC	EE
Average	29.5	89.1	74.8	32.7	20.2	19.1	13.4	3.6
SD	4.7	1.2	5.8	11.0	5.5	3.1	4.2	0.7
Maximum	37.7	90.8	81.6	51.6	30.8	23.9	21.6	4.8
Minimum	21.7	86.8	60.9	22.2	13.5	13.5	8.2	2.1

<sup>1</sup> Samples were obtained by hand-plucking from August to February.

<sup>2</sup>IVDMD = In vitro dry matter digestibility, WSC = water-soluble carbohydrates, EE = ether extract.

Since fat replaced ground corn in the concentrate on an equal energy basis, a higher amount of concentrate was offered to the cows from the control treatment (+0.92 kg DM/cow per day). This fact and the lack of differences in concentrate refusals explained the higher concentrate DMI observed in cows from the control treatment (Table 4). Pasture and total DMI were not affected by fat feeding. Changes in energy intake from pasture or total energy were not observed (Table 4).

During the period when all cows were fed the control concentrate, no residual effects of fat supplementation were detected for any of the parameters measured (Table 5). Total FCM produced over this period averaged 584 (control) and 596 kg (fat) without significant differences between treatments ( $P < 0.35$ ).

### Changes in BCS, SFD, and BW

No significant differences were observed in the changes of BCS (Figure 3), BW gain or SFD (Table 6)

during the period of fat supplementation. After the end of fat feeding at 75th day of lactation, the cows began to gain BCS, (Figure 2), BW, and SFD in both treatments (Table 6).

### Plasma Metabolites and Hormones Concentration

No interaction between treatment and week of lactation was detected for plasma metabolite and hormone concentrations over the first 75 DIM (Table 7). Average plasma levels of glucose, NEFA, and PUN were not affected by fat feeding. Concentration of plasma triglyceride and total cholesterol was higher in fat supplemented cows compared with controls (Table 7). Average plasma levels of IGF-I tended to be lower ( $P < 0.11$ ) in cows that received the supplemental fat, whereas plasma somatotropin and insulin concentrations did not change (Table 7). The highest plasma somatotropin concentrations were detected at 15 DIM in control cows (3.66 ng/mL) and at 30 DIM in fat-supplemented cows

**Table 3.** Milk production and composition in grazing dairy cows supplemented with 0 (control) and 0.73 kg/d (fat) of partially hydrogenated oil during the first 75 d of lactation.

Item	Treatment <sup>1</sup>		SEM	$P <^2$	
	Control	Fat		Treat	Cov
Milk, kg/d	23.7	25.0	0.40	0.02	0.01
FCM, kg/d	22.5	24.5	0.38	0.01	0.01
Milk fat					
kg/d	0.86	0.97	0.02	0.01	0.01
%	3.64	3.86	0.04	0.01	0.01
Milk protein					
Kg/d	0.74	0.78	0.01	0.03	0.01
%	3.12	3.14	0.02	0.55	0.01
Casein, <sup>3</sup> % of total N	72.75	72.33	1.08	0.83	
Fat/protein ratio	1.17	1.24	0.01	0.01	0.01
Lactose, %	4.71	4.70	0.03	0.84	0.01
Total solids, %	12.3	12.5	0.04	0.01	0.01
SNF, %	8.64	8.63	0.03	0.81	0.01
MUN, mg/dL	11.42	11.85	0.28	0.29	0.01
Cholesterol, mg/dL	33.05	32.97	1.36	0.97	0.01

<sup>1</sup>Results are expressed as least square means adjusted by covariate (milk production during the early phase [7 to 17 DIM] of the previous lactation).

<sup>2</sup>Effects of treatment (Treat) and covariate (Cov). No interactions between treatment and week of lactation were detected for any parameter measured.

<sup>3</sup>Data obtained during the first 45 DIM.

**Table 4.** Concentrate, pasture, and total DM and energy intake in grazing dairy cows supplemented with 0 (control) and 0.73 kg/d (fat) of partially hydrogenated oil during the period of pasture intake measurements (45 to 60 DIM).

Item	Treatment <sup>1</sup>		SEM	P <
	Control	Fat		
<b>Concentrate</b>				
Offered, kg DM/cow per day	5.00	4.08		
Refused, kg DM/cow per day	0.31	0.27	0.03	0.29
DM Intake, kg/cow per day	4.69	3.81	0.03	0.01
NE <sub>L</sub> intake, Mcal/cow per day <sup>2</sup>	9.51	9.10	0.06	0.01
<b>Pasture</b>				
DMI, kg/cow per day	15.57	15.83	0.78	0.82
NE <sub>L</sub> intake, Mcal/cow per day <sup>2</sup>	20.58	21.30	1.01	0.62
<b>Total</b>				
DMI, kg/cow per day	20.26	19.64	0.80	0.59
% BW	3.70	3.67	0.16	0.91
CP intake, kg/d	2.65	2.54	0.10	0.49
NDF intake, kg/d	8.74	8.73	0.42	0.98
NDF intake, % BW	1.60	1.63	0.08	0.75
NE <sub>L</sub> intake, Mcal/cow per day	30.09	30.40	1.03	0.83
Energy balance, Mcal/d <sup>2</sup>	2.00	0.36	1.04	0.28
FCM/DMI	1.10	1.30	0.05	0.01
FCM/NE <sub>L</sub> intake	0.741	0.834	0.03	0.03

<sup>1</sup>Results are expressed as least square means.

<sup>2</sup>Estimated according to NRC (2001) using data obtained during the period of pasture intake measurements (45 to 60 DIM). The calculated ME content of fat was 5.18 Mcal/kg DM (Schroeder et al., 2002).

(3.07 ng/mL). Concentrations of somatotropin decreased gradually in both treatments until 75 d postpartum where the lower levels of the hormone were observed (control = 1.62 ng/mL and fat = 1.33 ng/mL). Plasma IGF-I gradually increased from 15 d postpartum (control = 128.8 ng/mL and fat = 94.9 ng/mL) until 60 d postpartum (control = 164 ng/mL and fat = 136 ng/mL). Plasma insulin increased from 15 d postpartum (control = 0.31 ng/mL and fat = 0.31 ng/mL) until 45 d postpartum in fat supplemented cows (0.87 ng/mL) and 60 d postpartum in control cows (0.80 ng/mL).

Cows that received the partially hydrogenated oil over the first 75 DIM showed the higher concentration of plasma insulin during the residual period (76 to 105 DIM, Table 8). No residual effects of fat supplementation were detected for the somatotropin/insulin ratio or for any other parameter measured (Table 8).

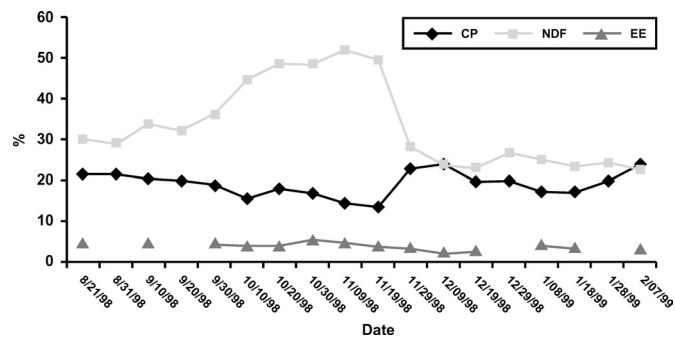
Differences between jugular and mammary vein plasma metabolites observed on d 30 of lactation were significantly different from zero (Student's *t* test for pairwise observations) for glucose, NEFA, and triglycerides but not for total cholesterol (Table 9). A higher

**Table 5.** Milk production and composition recorded from 76 to 105 DIM in grazing dairy cows supplemented with 0 (control) and 0.73 kg/d (fat) of partially hydrogenated oil during the first 75 d of lactation.

Item	Treatment <sup>1</sup>		SE	P < 2		
	Control	Fat		Treat	Wk	Treat × Wk
Milk, kg/d	20.40	20.62	0.13	0.62	0.01	0.71
4%FCM, kg/d	19.49	19.86	0.26	0.38	0.03	0.63
<b>Milk fat</b>						
kg/d	0.75	0.77	0.01	0.35	0.10	0.74
%	3.67	3.72	0.06	0.59	0.85	0.98
<b>Milk protein</b>						
kg/d	0.63	0.65	0.01	0.20	0.01	0.62
%	3.07	3.14	0.03	0.26	0.23	0.93
Fat/protein ratio	1.21	1.19	0.02	0.75	0.64	0.89
Lactose, %	4.62	4.67	0.02	0.51	0.01	0.14
Total solids, %	12.2	12.3	0.07	0.13	0.02	0.60
Solids non fat, %	8.49	8.60	0.04	0.19	0.01	0.38
MUN, mg/dL	15.15	13.99	0.35	0.27	0.04	0.83
Cholesterol, mg/dL	32.30	32.24	0.01	0.99	0.01	0.58

<sup>1</sup>Results are expressed as least square means.

<sup>2</sup>Effects of treatment (Treat), week of lactation (Wk) and Treat × Wk interaction.



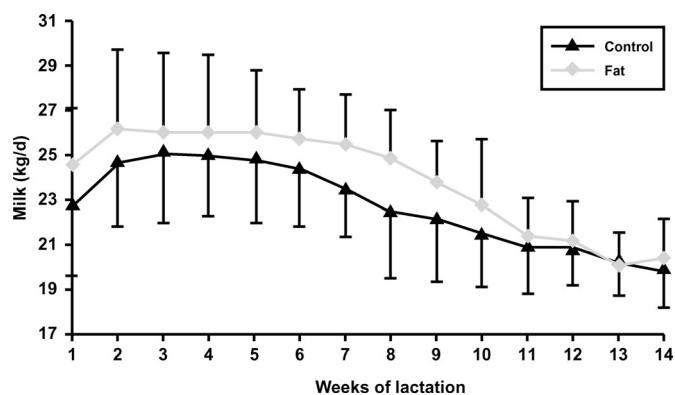
**Figure 1.** Pasture CP, NDF, and ether extract (EE) composition across the experiment.

apparent mammary uptake for triglycerides was detected in cows supplemented with hydrogenated oil.

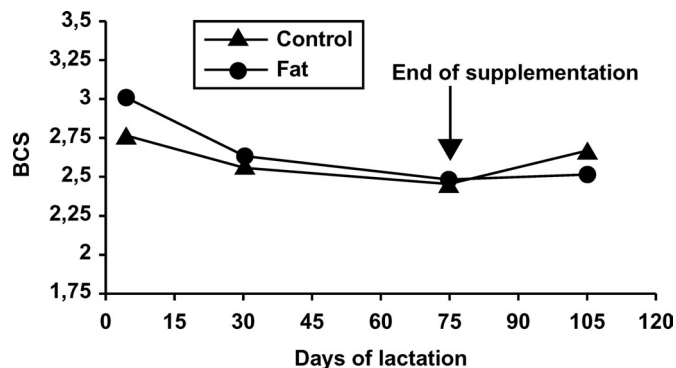
### Ruminal Environment and Pasture NDF Disappearance

The overall means for total or individual VFA concentrations, for the concentrations of  $\text{NH}_3\text{-N}$  or the pH values were not affected by fat supplementation (Table 10). The acetate:propionate (**C2:C3**) ratio tended to be higher ( $P < 0.06$ ) in the cows that received the hydrogenated oil. The highest pH values were detected at 0600 a.m. (control = 6.23 and fat = 6.40) and the lowest were observed at 1400 p.m. (control = 5.54 and fat = 5.56).

The partially hydrogenated oil did not affect the degradable fractions, the digestion rate or the effective degradability of the DM and NDF fractions of the pasture (Table 11). The partial hydrogenation of the oil demonstrated no effect on ruminal digestion.



**Figure 2.** Yield of 4% FCM in grazing dairy cows supplemented with 0 (control) and 0.7 kg/d (fat) of partially hydrogenated oil during the first 75 d of lactation. Treatment  $\times$  week of lactation interaction =  $P < 0.75$ , day =  $P < 0.0001$ , and treatment  $P < 0.001$ .



**Figure 3.** Changes in BCS in grazing dairy cows supplemented with 0 (control) and 0.73 kg/d (fat) of partially hydrogenated oil in early lactation. Treatment  $\times$  week of lactation interaction =  $P < 0.24$ , wk =  $P < 0.0001$ , and treatment  $P < 0.43$ .

## DISCUSSION

### Diet Characteristics and Feed Intake

Pasture management was adjusted to avoid restrictions in forage intake to adequately evaluate the effects of supplemental fat on pasture DM and energy intake. A daily strip-grazing system was used to adjust areas of sufficient size that may provide 30 kg DM of pasture per day. This target herbage allowance was in the range of 21 to 39 kg DM/d suggested by Minson (1990) in order to obtain ad libitum pasture intake. In grazing conditions, pasture intake should be maximal when the offer is about 45 g of OM pasture /kg BW (Minson, 1990). From the average BW of cows (578 kg) and the OM content of pastures (89.1%, DM basis) it can be calculated that optimal herbage allowance (29.2 kg DM/cow per day) was close to that utilized in the present experiment ( $30.4 \pm 2.5$  kg DM/cow per day). The forage NDF content is one of the nutritional factors that may reduce the voluntary intake of cows (Mertens, 1994). The average pasture NDF content (Table 2) was below the values (40 to 50%) considered as adequate for well-managed pastures (Minson, 1990; Muller and Fales, 1998) and close to the value of 34 to 36%, which would not affect voluntary DMI due to rumen fill (Mertens, 1994). The pasture NDF also showed a high rate and extent of rumen digestion (Table 11), suggesting a good quality of the forage apparently consumed by cows. Average pasture CP content (Table 2) was in the range of 15 to 25% proposed by Minson (1990) to obtain high values of forage DM digestibility.

The partial replacement of corn grain with hydrogenated oil increased the energy density of the concentrate (+18%) and reduced its starch content (-22%, Table 1). The energy provided by the concentrate was slightly higher in the control treatment (+4.5%, Table



**Table 6.** Changes in BW gain (BWG) and subcutaneous fat depth (SFD) in grazing dairy cows supplemented with 0 (control) and 0.73 kg/d (fat) of partially hydrogenated oil during the first 75 d of lactation.

Item	Treatment <sup>1</sup>		SE	P <sup>2</sup> <
	Control	Fat		
BWG, kg/d				
4 to 30 DIM	-0.65	-0.69	0.17	0.87
30 to 75 DIM	-0.40	-0.42	0.17	0.93
75 to 105 DIM	0.22	0.15	0.21	0.81
SFD, $\mu\text{m}$				
4 to 30 DIM	-310	-316	0.25	0.98
30 to 75 DIM	-167	-172	0.12	0.97
75 to 105 DIM	110	113	0.14	0.98

<sup>1</sup>Results are expressed as least square means.

<sup>2</sup>BWG: day of lactation (Time) =  $P < 0.21$  and treatment (Treat)  $\times$  Time interaction =  $P < 0.96$ ; SFD: Time =  $P < 0.49$  and Treat  $\times$  Time =  $P < 0.99$ .

4) and the main difference between treatments was the nature of this energy being fermentable (starch) in control and nonfermentable (fatty acids) in fat concentrate (Table 1).

In fat-supplemented cows, microbial protein synthesis may be reduced as the consequence of a lower intake of fermentable energy from the concentrate (Clark et al., 1992). However, providing hydrogenated fat might also allow the microbes to use preformed fatty acids, which might improve efficiency of growth (D. J. Schingoethe, personal communication). In this experiment, the inclusion of fishmeal as a source of RUP could prevent a decrease in milk protein content observed when supplemental fat is supplied to lactating dairy cows (Gagliostro and Chilliard, 1992; Wu and Huber, 1994). A soluble source of Ca was added to the concentrate to compensate for any potential loss in Ca digestibility due to fat supplementation (Palmquist, 1984).

### Milk Production and Composition

Among all the previous grazing experiments conducted at INTA Balcarce on the effects of fat feeding

on lactating dairy cows performance, this was the first one in which nonfermentable energy partially replaced starch in the concentrate and total energy intake was held constant. The results showed that some starch may be replaced with fat increasing milk yield in dairy cows grazing spring pastures (Table 3). This may contribute to prevent ruminal acidosis and reduction in milk fat content often observed with excessive amounts of fermentable energy in the diet.

The increase in milk yield over the first 75 DIM in fat-supplemented cows (+1.3 kg/d, Table 3) was close to the mean effect observed (about 1.0 kg/d) when grazing experiments were reviewed and fat supplementation ranged from 0.2 to 1.0 kg/cow per day (Salado, 2000; Schroeder et al 2003). It was suggested that positive effects of fat feeding would be mainly obtained in high yielding dairy cows owing to their higher requirements of energy and long-chain fatty acids (Gagliostro and Chilliard, 1992). In the present experiment (replacing starch by fat) and in our previous work (adding fat to the concentrate, Schroeder et al., 2002), a positive response was detected using cows of moderate milk

**Table 7.** Plasma metabolites and hormones in grazing dairy cows supplemented with 0 (control) and 0.73 kg/d (fat) of partially hydrogenated oil in early lactation.

	Treatment <sup>1</sup>			P < <sup>2</sup>		
	Control	Fat	SE	Treat	Time	Treat $\times$ Time
Glucose, mg/dL	61.8	63.3	0.7	0.32	0.01	0.91
NEFA, $\mu\text{eq/L}$	659.1	690.9	13.4	0.59	0.01	0.99
PUN, <sup>3</sup> mg/dL	7.0	7.5	0.2	0.21	0.01	0.65
Triglycerides, mg/dL	250.9	267.2	5.3	0.02	0.01	0.93
Cholesterol, mg/dL	177.0	205.5	1.8	0.001	0.01	0.59
Somatotropin, ng/mL	2.52	2.44	0.17	0.8	0.01	0.42
Insulin, ng/mL	0.52	0.55	0.03	0.69	0.01	0.24
Somatotropin/insulin	8.27	7.54	0.88	0.64	0.01	0.66
IGF-I, ng/mL	140.1	112.5	5.6	0.11	0.01	0.99

<sup>1</sup>Results are expressed as least square means.

<sup>2</sup>Effects of treatment (Treat), day of lactation (Time) and Treat  $\times$  Time interaction.

<sup>3</sup>PUN = Plasma urea nitrogen.

**Table 8.** Plasma metabolites and hormones in grazing dairy cows after supplementation with 0 (control) and 0.73 kg/d (fat) of partially hydrogenated oil (75 to 105 DIM).

	Treatment <sup>1</sup>		SE	P <
	Control	Fat		
Glucose, mg/dL	61.5	62.4	1.29	0.65
NEFA, $\mu$ eq/L	351.7	371.1	19.5	0.49
PUN <sup>2</sup> , mg/dL	10.6	10.9	0.5	0.72
Triglycerides, mg/dL	224.8	230.3	8.8	0.66
Cholesterol, mg/dL	183.9	184.9	5.6	0.90
Somatotropin, ng/mL	2.28	2.61	0.40	0.56
Insulin, ng/mL	0.66	1.13	0.10	0.01
Somatotropin/insulin	3.73	2.91	0.72	0.43
IGF-I, ng/mL	151.4	111.0	21.8	0.17

<sup>1</sup>Results are expressed as least square means.

<sup>2</sup>PUN = Plasma urea nitrogen.

production potential (26 to 27 kg/d at peak of lactation). An increase in milk yield in early lactation (+11.3%) was also detected when 0.4 kg/d of calcium salts of saturated fatty acids were fed to grazing cows of moderate genetic merit (22 to 24 kg of milk/d) for milk production (Gagliostro, 1998). The difference between fat and control cows for FCM (+2 kg/d, Table 3) can be mainly explained by an increase in milk fat production (+12.8%) rather than in milk yield (+5.5%, Table 3). The effect of fat supplementation on milk and FCM production in grazing dairy cows also depends on the degree of saturation of the fat. Whereas supplementation with unsaturated FA sources did not significantly increase milk or FCM production, both parameters

**Table 9.** Differences between jugular and mammary vein (J-M) plasma metabolites in grazing dairy cows supplemented with 0 (control) and 0.73 kg/d (fat) of partially hydrogenated oil in early lactation.<sup>1</sup>

	Treatment <sup>1</sup>		SE	P <
	Control	Fat		
Glucose, mg/dL				
Jugular	54.3	54.4	1.5	0.95
J-M	10.8*	10.6*	1.91	0.94
(%) <sup>2</sup>	19.9	19.5		
NEFA, $\mu$ eq/L				
Jugular	723.4	756.3	55.2	0.68
J-M	98.3*	105.0*	28.6	0.87
(%)	13.6	13.9		
Triglycerides, mg/dL				
Jugular	179.9	192.4	6.4	0.18
J-M	2.65*	5.11*	0.78	0.03
(%)	1.47	2.66		
Cholesterol, mg/dL				
Jugular	162.4	191.2	7.5	0.01
J-M	2.9	2.1	4.8	0.91
(%)	1.82	1.12		

<sup>1</sup> Samples were obtained on d 30 postpartum. Results are expressed as least square means.

<sup>2</sup>Expressed as % of jugular concentration.

\*Significantly different from zero ( $P < 0.01$ , Student's  $t$  test for pairwise observations). [Au: OK as edited?]

were increased by saturated FA supplements (Schroeder et al., 2003).

As cows had similar total NE<sub>L</sub> intake (Table 4), the increase in milk and in FCM yields (Table 3) after fat feeding were not apparently explained by higher energy absorption (Table 4). The supplementation with hydrogenated oil did not induce a different pattern of variation in BW gain, SFD (Table 6) or plasma NEFA concentrations (Table 7). These results suggest that body tissue mobilization did not seem to contribute in a different way to milk energy between fat supplemented and control cows. As was observed here (Table 4), our previous study also showed a higher efficiency (expressed as FCM production per kilograms of total DMI or per Mcal of NE<sub>L</sub> consumed) in grazing dairy cows receiving the hydrogenated oil (Schroeder et al., 2002). It has been proposed that maximum efficiency of milk production occurs when about 12 to 16% of total ME requirements are supplied as dietary fat (Brumby et al, 1978). In the present experiment, ME provided by supplemental fat was estimated to be about 8.2%, and in the previous experiment it represented about 11% (Schroeder et al. 2002). Although the 2 estimations were below the range proposed as the most efficient for milk production, positive milk responses were detected.

It has been suggested that supplemental fat in early lactation may improve the persistency of milk and milk fat production through a carryover effect that may occur after the end of fat feeding (Palmquist, 1984; Schingoethe and Casper, 1991). In the present study, fat supply was stopped at 75 DIM and no residual effects of fat supplementation were detected (Table 5, Figure 2) as previously observed by Schroeder et al. (2002). The lack of a positive residual effect in the present experiment should be taken with care because the pasture quality decreased towards the end of the experiment.

The observed increase in milk fat content after fat feeding (+0.22 g/100 g, Table 3) was close to the mean response of 0.17 g/100 g ( $P < 0.01$ ) observed when grazing experiments were reviewed by Salado (2000) but lower than that observed in nongrazing experiments (+0.40 g/100 g,  $P < 0.01$ ) using saturated fat (Gagliostro and Chilliard, 1992). The response in total milk fat output (+110 g/d,  $P < 0.01$ , Table 3) was higher than the mean response obtained in grazing experiments (+85 g/d,  $P < 0.01$ ) (Salado 2000). Concentration and yield of milk fat depends on the balance between the increase in exogenous fatty acid transfer to the mammary gland and the decrease in de novo synthesis. A reduction of fatty acid synthesis within the mammary gland may be expected when supplemental fat is added to the diet of lactating dairy cows (Chilliard, 1993). The reduction may be mediated at the ruminal level through

**Table 10.** Parameters of rumen environment in grazing dairy cows supplemented with 0 (control) and 0.73 kg/d (fat) of partially hydrogenated oil.

	Treatment <sup>1</sup>		SE	P < <sup>2</sup>		
	Control	Fat		Treat	Hour	Treat × Hour
VFA, mmol/L						
Total	137.3	140.6	5.59	0.84	0.01	0.59
Acetate (C2)	86.2	88.4	3.59	0.83	0.01	0.63
Propionate (C3)	26.8	26.9	1.04	0.99	0.01	0.59
Isobutyrate	2.68	2.77	0.12	0.52	0.02	0.24
Butyrate	17.5	18.3	0.77	0.72	0.01	0.54
Isovalerate	2.05	2.11	0.10	0.72	0.01	0.44
Valerate	1.82	1.86	0.09	0.85	0.01	0.53
Caproate	0.24	0.29	0.02	0.40	0.01	0.47
C2:C3 ratio	3.27	3.39	0.04	0.06	0.01	0.56
NH <sub>3</sub> -N, mg/dL	10.05	11.46	0.97	0.14	0.06	0.35
PH	5.73	5.75	0.03	0.21	0.01	0.36

<sup>1</sup>Results are expressed as least square means.

<sup>2</sup>Effects of treatment (Treat), hour of sampling (Hour) and treatment × hour interaction.

a lower rumen acetate and butyrate production or through the inhibitory effect of long-chain FA on mammary lipogenic enzymes (Palmquist, 1984; Chilliard, 1993). In our experiment, production rates of acetate and butyrate were not measured. As changes in the acetate and butyrate proportions were not observed after fat feeding (Table 10) and pasture DMI was not decreased (Table 4), the production of acetate and butyrate was probably not affected. The saturated source of fat used in the experiment could also contribute to maintain the secretion of short- and medium-chain fatty acids, because the inhibition of de novo mammary synthesis is more sensitive to unsaturated fatty acid supply (Palmquist, 1984). The higher apparent mammary uptake of triglycerides in the fat treatment (Table

9) was also consistent with the observed increase in milk fat output (Table 3). According to the results obtained here, when the hydrogenated oil (1 kg/d) was supplied to early lactation cows fed spring pastures, milk fat content (+0.34 g/100 g), and milk fat output (+160 g/d) were increased (Schroeder et al. 2002). The results were explained by a higher yield of C<sub>16:0</sub>, C<sub>18:0</sub>, and C<sub>18:1</sub> FA with no changes in the secretion of short and medium-chain FA (Schroeder et al. 2002).

Replacement of starch with fat (Table 1) decreases the amount of energy that is available for growth of ruminal microorganisms and may reduce microbial protein synthesis (Clark et al., 1992). As pasture DMI was not decreased by fat (Table 4) and the WSC content in the grazed forage was relatively high (13.4%, Table 2), microbial protein synthesis in the rumen was probably not decreased. Concentration of MUN (11.6 mg/dL, Table 3) was in the range of 10 to 16 mg/dL proposed by Jonker et al. (1998), which may reflect a high N efficiency and a low N excretion. This fact and the inclusion of fishmeal as a source of RUP probably contributed to preventing a possible decrease in amino acid availability to the mammary gland. Other results obtained in grazing conditions (King et al, 1990; Gagliostro 1998; Schroeder et al., 2002) also showed no decrease in milk protein content when saturated fat was added to the diet. In grazing experiments (n = 14), the overall effect of fat feeding on milk protein content (0.53 ± 0.17 kg fat/cow) was near zero in 86% of the studies reviewed by Salado (2000), and reductions in milk protein content were only detected in 7% of the experiments. Decreases in the casein N in milk is often observed when fats are fed and a simultaneous increase in NPN was also reported (DePeters and Cant, 1992). Both in this study (Table 3) and in our previous one (Schroeder et al.,

**Table 11.** Parameters of forage DM and NDF degradation in grazing dairy cows supplemented with 0 (control) and 0.73 kg/d (fat) of partially hydrogenated oil.

	Treatment <sup>1</sup>		SE	P <
	Control	Fat		
DM, %				
Soluble fraction	32.96	34.84	1.80	0.53
Degradable fraction	53.14	54.42	2.80	0.78
Rate of digestion, %/h	8.42	6.83	0.66	0.23
Effective degradability				
Kp <sup>2</sup> = 5%/h	65.94	65.63	0.49	0.70
Kp <sup>2</sup> = 7%/h	61.65	61.17	0.71	0.68
NDF, %				
Digestible fraction	68.03	79.76	5.28	0.26
Rate of digestion, %/h	5.22	3.68	0.65	0.23
Lag time, h	1.66	1.60	0.85	0.96
Effective degradability				
Kp <sup>2</sup> = 5%/h	31.77	30.83	1.05	0.59
Kp <sup>2</sup> = 7%/h	25.81	24.45	1.44	0.57

<sup>1</sup>Results are expressed as least square means.

<sup>2</sup>kp = Rates of passage assumed according to Van Vuuren et al (1992).

2002), casein N content and MUN were not affected by fat feeding.

### Changes in BCS and BW

As it was observed here (Table 6), dietary fat did not seem to decrease the loss of BW or lipid mobilization in early lactation cows (Palmquist, 1984; Chilliard, 1993, Komaragiri et al., 1998). Similar results were observed in grazing dairy cows in early lactation (Gagliostro, 1998; Schroeder et al., 2002).

Total energy intake was not increased (Table 4), and a higher response in milk yield was observed in fat fed cows (Table 3). As the basal NEFA concentrations remained unchanged (Table 7), it seems that energy balance was probably not decreased in fat supplemented cows throughout the experiment. During the period of intake measurements (45 to 60 DIM), calculated energy balance was similar between treatments (Table 4). Agreeing with the results of this experiment, when the hydrogenated oil was added to the concentrate of grazing cows, a higher response in milk yield was observed, but changes in BCS, BW, total energy intake, or basal NEFA concentration were not detected (Schroeder et al., 2002). Komaragiri et al. (1998) suggested that the hormonal profile may play a more important role than the type of diet (fat) in regulating body lipid mobilization in the early-lactation dairy cow. In our study (Table 7) and in the previous experiment by Schroeder et al. (2002), supplemented fat did not change the plasma hormone profile of cows or the somatotropin/insulin ratio. (Table 4). The improvement in the efficiency of energy utilization for lactation when fat is added to the diet has been attributed to a lower energy loss as methane, a greater efficiency arising from the direct use of long-chain FA for milk fat secretion and a higher efficiency of ATP generation from long-chain FA rather than acetate (Chilliard, 1993).

### Plasma Concentration of Metabolites and Hormones

Fat supplementation had no consistent effects on circulating glucose and insulin concentrations (Chilliard, 1993). However, as fat replaced starch in the concentrate, a decrease in the entry of propionic acid to maintain hepatic glucose synthesis and pancreatic insulin secretion could be expected. The absence of negative effects of fat on pasture DMI and the high WSC content in the grazed forage (14.2%, Table 2) probably contributed to maintain ruminal propionate, plasma glucose, and insulin levels. Fatty acids absorbed from the hydrogenated oil may have also contributed to maintaining glycemia by reducing total (CO<sub>2</sub>) or partial (to NADPH<sub>2</sub>) glucose oxidation (Chilliard, 1993).

Hormones may play an important role in coordinating the partition of dietary fatty acids between milk fat secretion, deposition in adipose tissue, and body lipid mobilization (Palmquist, 1984). Enhanced adipose tissue lipolysis is also associated with a higher plasma somatotropin/insulin ratio (Vernon, 1988). The lack of fat feeding effects on basal NEFA was in accordance with the absence of variations in the body parameters and with the similar somatotropin/insulin ratio. Plasma urea levels did not change in fat-supplemented cows in agreement with the similar MUN observed in both treatments, and the lack of fat effects on rumen NH<sub>3</sub>-N, and may be due to the similar pasture (and hence RDP) intake.

The higher apparent mammary uptake of triglycerides agrees with the higher milk fat content and yield as jugular concentration and apparent uptake are closely linked (Gagliostro et al., 1991). On the other side, in spite of the increase in plasma cholesterol levels in fat-supplemented cows, the apparent mammary uptake and milk cholesterol content were not affected as previously observed by Schroeder et al. (2002), suggesting a reduced mammary uptake of this metabolite (Christie, 1981).

In cattle, the main hormone stimulating milk production is somatotropin by direct and indirect mechanisms, including the involvement of the IGF system and interactions with metabolic hormones such as insulin and T4 (Vernon, 1989; Cohick, 1998). Circulating concentrations of these hormones are all regulated by food intake and/or nutritional status. No clear effects of dietary fat have been found on circulating somatotropin in lactating dairy cows (Wu and Huber, 1994). In this study and in the previous experiment (Schroeder et al., 2002), somatotropin concentrations were not affected after hydrogenated oil feeding to grazing dairy cows in early lactation, suggesting that the increase in milk production was not mediated by this hormone.

The somatotropin-IGF-1 axis is functional in lactating cows only when nutrient availability is not restricted (Cohick, 1998). In early-lactation cows, McGuire et al. (1998) reported that plasma IGF-I concentration was positively correlated with energy intake and plasma somatotropin levels. The lack of a significant effect of fat on IGF-I concentrations in the present experiment is consistent with the similar somatotropin levels observed, in accordance with the adequate nutritional level provided throughout the experiment (discussed above), and with the similar energy intake of cows.

### Ruminal Environment and Forage NDF Digestion

When supplemental fat has negative effects on ruminal digestion, a reduced VFA production and a lower



C2:C3 ratio may be expected (Jenkins, 1993). In this experiment, concentration of total or individual VFA were not affected (Table 10), suggesting that the elevation of the melting point above the ruminal temperature was effective in preventing any negative action of the hydrogenated oil on ruminal digestion as proposed by Chalupa (1986). Although the C2:C3 ratio tended to be higher in oil-supplemented cows (Table 10), the result was probably better explained by the replacement of corn grain with fat rather than a direct effect of the supplemented oil because parameters of pasture NDF fermentation were not affected (Table 11).

It was pointed out that replacement of starch with fat may increase ruminal  $\text{NH}_3\text{-N}$  concentration owing to the higher RDP/nonstructural carbohydrate ratio (DePeters and Cant, 1992), but this result was not observed here (Table 10). The hydrogenated oil did not seem to affect ruminal forage CP degradation as reported in other grazing experiments (King et al., 1990).

Parameters of pasture NDF digestion were not affected by the hydrogenated oil (Table 11) and showed a high rate and extent of ruminal fiber digestion. The result is consistent with the good quality of the forage offered to cows (Table 2) and agree with previous studies concluding that negative effects were minimal in diets with a high proportion of forage (Palmquist, 1984). The high rate of passage generally observed when dairy cows are grazing high-quality pastures and the adequate levels of calcium of these pastures could alleviate some possible negative effects of fat supplements (Schroeder et al, 2003). More research including unsaturated fat sources and different types of pastures is needed before definitive conclusions can be obtained.

## CONCLUSIONS

This experiment examined the opportunity for fat to affect lactation performance of grazing dairy cows when total energy intake was held constant. In the isocaloric diets of the trial, nonfermentable energy in the form of saturated fat was successfully used to increase milk yield even when it replaced energy-yielding substrates to ruminal microbes in grazing production systems. Although supplemental fat did not increase total energy intake and milk yield was enhanced, higher BW loss or plasma NEFA was not observed. Concentrations of regulatory hormones were not sensitive to saturated fat feeding in grazing cows, suggesting that mechanisms by which milk yield was improved need to be further investigated. The use of a partially hydrogenated oil did not modify parameters of forage DM and NDF degradation or rumen environment in grazing dairy cows.

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