

Effect of Supplemental Tallow on Performance of Dairy Cows Fed Diets with Different Corn Silage:Alfalfa Silage Ratios

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ABSTRACT

A study was conducted to investigate the response to supplemental tallow of lactating cows fed basal diets with different alfalfa silage:corn silage ratios. We postulated that supplemental tallow will have decreasing negative effects on rumen fermentation, dry matter intake (DMI), and milk fat percentage as the dietary ratio of alfalfa silage:corn silage is increased. Eighteen Holstein cows averaging 134 ± 14 d in milk were used in a replicated 6×6 Latin square design with 21-d periods. Treatments were arranged as a 2×3 factorial with 0 or 2% tallow (DM basis) and three forage treatments: 1) 50% of diet DM as corn silage, 2) 37.5% corn silage and 12.5% alfalfa silage, and 3) 25% corn silage and 25% alfalfa silage. Cows were allowed ad libitum consumption of a total mixed ration. Diets were formulated to contain 18% crude protein and 32% neutral detergent fiber. No fat \times forage treatment interactions were observed. Fat supplemented cows had lower DMI and produced more milk with less milk fat content relative to non-supplemented cows. Concentration of *trans*-octadecenoic acids was higher in milk fat of tallow-supplemented cows. Tallow supplementation had no effect on ruminal pH and acetate:propionate ratio, but tended to decrease total volatile fatty acid (VFA) concentration in the rumen. Increasing the proportion of alfalfa silage increased DMI, milk fat percentage, and milk fat yield regardless of the fat content of the diet. Total VFA concentration and acetate:propionate ratio in the rumen were increased in response to higher levels of alfalfa in the diets. These results suggest that replacing corn silage with alfalfa silage did not alleviate the negative response of dairy cows to tallow supplementation at 2% of diet DM.

(**Key words:** tallow, corn silage and alfalfa silage, milk fat and rumen fermentation)

Abbreviation key: A:P = ruminal acetate to propionate ratio, AS = alfalfa silage, CLA = conjugated linoleic acid, CS = corn silage, FA = fatty acid, MFD = milk fat depression, NFC = nonfibrous carbohydrate; SNF = solids not fat.

INTRODUCTION

A compilation of data from lactation studies (Smith and Harris, 1993) suggested that the likelihood for decreased milk yield or milk fat percentage, or both, when extruded soybeans, whole cottonseeds, and rendered animal fats were fed was greater with corn silage-based diets than with alfalfa-based diets. In other words, the influence of a ruminally active fat source on rumen fermentation may vary according to the feed ingredients of the basal diet. In contrast, response to fats designed to be ruminally inert were not affected or inconsistently affected by forage type (Smith and Harris, 1993), suggesting that diet \times fat interactions on lactation performance are related to ruminal events.

Smith et al. (1993) indicated that the depression in milk production and milk fat percentage caused by supplementation of 2.5% tallow or 12% cottonseed, or both (DM basis), to corn silage-based diets was alleviated by replacing 25 to 50% of the corn silage with alfalfa hay. Examination of nutrient digestibility data led the authors to speculate that ruminally active fats may have a more negative effect on rumen fermentation when diets are based on corn silage as the only forage source. Ben Salem et al. (1993) observed that 7% rapeseed oil can be supplemented to hay-based diets without disturbing rumen digestion, but it depressed fiber digestibility when supplemented to corn silage-based diets.

It was unknown whether the fat \times forage interactions observed by Smith et al. (1993) could be replicated using diets commonly fed in the Midwestern United States.

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In a previous study (Onetti et al., 2001), we showed that supplementing tallow or choice white grease at 2 or 4% of the diet DM to diets containing 50% concentrate and 50% corn silage as the sole forage source (DM basis) had negative effects on DMI, milk production, and milk fat percentage, and rumen fermentation of high producing dairy cows in midlactation. Based on these findings, the objective of the present study was to determine whether replacing corn silage (CS) made from high grain-yielding corn silage with alfalfa silage (AS) would alleviate the negative effects of feeding supplemental tallow at 2% of the diet DM on rumen fermentation, DMI, and milk fat percentage. A major difference between our study and that of Smith et al. (1993) is the use of alfalfa silage versus hay. Although we postulated that the negative effects of tallow on corn silage-based diets will be alleviated by replacement with AS, it may be presumptive that results will be similar. For example, alfalfa forage preserved as silage or hay has been shown to elicit different responses on chewing behavior of dairy cows (Nelson and Satter, 1990; Grant and Weidner, 1992) that might result in changes of rumen environment. We decided to use tallow because it is a relatively inexpensive and commonly used fat source to increase energy density of diets without providing additional nutrients (Shaver, 1990).

MATERIALS AND METHODS

Animals

Eighteen Holstein cows averaging 134 ± 14 DIM and 623 ± 54 kg of BW were used in a replicated 6×6 Latin square design with 21-d periods; 14 d for diet adaptation and 7 d for data collection. One square consisted of primiparous cows, one of multiparous cows without rumen fistulas, and one of multiparous cows with rumen fistulas. Cows within a square were assigned randomly to dietary treatments. Treatment sequences were ordered to minimize residual or carryover effects of any treatment in the succeeding period. Animals were handled according to procedures approved by the Research Animal Resources Center of the UW-Madison (RARC # A-00898-3-10-98). Cows were housed individually in a tie-stall and stanchion barn and had free-choice access to water. All cows were injected every 14 d with bovine somatotropin (Posilac, Monsanto Company, St. Louis, MO) on the same day of the experimental period. Two cows were removed from the trial. One cow was removed at the beginning of the third period because she developed toxic mastitis; a replacement cow was used during the last three periods. The other cow was removed from the trial during periods five and six due to teat injury, and was not replaced.

Diets

Treatments were arranged as a 2×3 factorial with 0 or 2% supplemental tallow (DM basis) and three forage treatments: 1) 50% of diet DM as CS, 2) 37.5% CS and 12.5% AS, and 3) 25% CS and 25% AS. Concentrate comprised 50% of the diet DM. Ingredient composition of experimental diets is presented in Table 1. All corn silage diets with and without 2% supplemental tallow in the present study were formulated with the same ingredient composition as in Onetti et al. (2001). When adding alfalfa, the amount of soybean meal in the diet was reduced and corn was increased to make the diets isonitrogenous and isoenergetic. This also allowed formulation of diets similar in nonfibrous carbohydrates (NFC), because the decrease in NFC from replacing CS with AS was offset by an increase in NFC when replacing soybean meal with corn. To minimize treatment differences in NDF, a high-grain yielding corn hybrid (Pioneer 3563, Pioneer Hi-Bred International, Des Moines, IA) was used. Alfalfa and corn silage NDF contents averaged 38 and 40% (DM basis), respectively. Corn silage was harvested at a theoretical length of chop of 12.7 mm and was processed (1-mm roll clearance). The theoretical length of chop of AS was 10 mm. Urea was added to the 100% CS treatments to minimize the likelihood that $\text{NH}_3\text{-N}$ would be limiting for microbial growth (NRC, 2001). The tallow was incorporated into concentrates and then added to the TMR. Cows were fed the diets twice daily (0900 and 1800 h) as a TMR to allow ad libitum consumption and 10% feed refusal (as-fed basis). Diets were formulated to meet or exceed NRC (1989) nutrient allowances.

Table 2 shows the chemical composition of the experimental diets. Crude protein content was similar across diets averaging 17% of diet DM, except for the treatment containing tallow and 25% CS-25% AS, which was 2 percentage units of CP higher than the average. The reason for the increased CP content in this particular treatment is unknown, but it may be related to the higher proportion of soybean meal in this treatment when compared to the same forage treatment without fat, a mixing error, or a sampling error. Neutral detergent fiber content of the diets containing fat was, on average, approximately 2 percentage units lower than diets without supplemental fat (32.3 vs. 34.6% of the diet DM for diets with and without supplemental tallow, respectively). Reduced NDF in fat-supplemented diets is likely due to the lower inclusion of soybean hulls. Diets containing tallow had a fatty acid (FA) content of 0.7 percentage units lower than expected. Fat sticking to the walls of the Wiley mill while processing the samples could partially account for this lower fatty acid content. It is also likely that fat might

Table 1. Ingredient composition of experimental diets.

Corn silage	0% Tallow			2% Tallow		
	50%	37.5%	25%	50%	37.5%	25%
(% of the DM)						
Alfalfa silage	0.0	12.5	25.0	0.0	12.5	25.0
Corn silage	50.0	37.5	25.0	50.0	37.5	25.0
Blood meal	1.0	1.0	1.0	1.0	1.0	1.0
Meat and bone meal	3.0	3.0	3.0	3.0	3.0	3.0
Distiller's grain, dry	5.0	5.0	5.0	5.25	5.25	5.25
Soybean meal, 48% CP	13.6	10.6	7.6	15.6	13.6	11.6
Cracked corn	12.0	16.0	20.0	10.5	13.5	16.5
Soybean hulls	13.25	13.0	12.4	10.5	10.2	9.5
Urea	0.40	0.0	0.0	0.40	0.0	0.0
Limestone	0.90	0.60	0.20	0.90	0.65	0.40
Magnesium oxide	0.05	0.0	0.0	0.05	0.0	0.0
Trace mineral salt ¹	0.50	0.50	0.50	0.50	0.50	0.45
Vitamin mix ²	0.30	0.30	0.30	0.30	0.30	0.30
Tallow ³	0.0	0.0	0.0	2.0	2.0	2.0

¹Contained 0.55% Mn, 0.55% Zn, 0.35% Fe, 0.14% Cu, 0.008% I, 0.006% Se, and 0.002% Co.

²Contained 3304 IU/g of DM of vitamin A, 1101 IU/g of DM of vitamin D, and 11 IU/g of DM of vitamin E.

³Fatty acid composition (g/100 g of fatty acids) of tallow (Packerland Packing Co., Inc., Green Bay, WI) was 3% C14:0, 25.1% C16:0, 2.7% C16:1, 19.7% C18:0, 42.1% C18:1, 3% C18:2, 0.3% C18:3, and 4.1% others.

have been lost due to sticking to the walls of the mixer during TMR preparation. The concentration of NE_L was 0.09 Mcal/kg DM higher for fat-supplemented diets, and was decreased by 0.01 to 0.02 Mcal/kg DM with each incremental level of AS in the diet.

Sampling and Laboratory Analysis

Dry matter content of the forages sources and concentrates were determined weekly using a 60°C forced-air oven; results were used to adjust as-fed ratios of forages and concentrate in the TMR. The amounts of TMR offered and refused were measured daily. Orts were collected on d 17 to 21 of each period and dried overnight in a 100°C forced-air oven for DMI determination. Corn silage, alfalfa silage, and concentrate samples were collected weekly, dried for 48 h in a 60°C forced-air oven, and ground to pass a 2-mm Wiley mill screen (Arthur H. Thomas, Philadelphia, PA). Organic matter was determined by oven drying at 550°C for 720 min. Samples were analyzed for NDF (Mertens, 1999), CP (AOAC, 1990), ether extract (AOAC, 1990), and fatty acid (Sukhija and Palmquist, 1988). The nonfibrous carbohydrate component was calculated as 100 - (NDF + ether extract + CP + ash).

Cows were milked twice daily, and milk production was recorded at each milking during the final 7 d of each period. Milk samples from the a.m. and p.m. milking collected on 5 consecutive days (d 17 to 21 of each period) were analyzed for fat, CP, and solids not fat (SNF) by infrared analysis (AgSource Milk Analysis Laboratory,

Menomonie, WI). Milk samples from d 18 and 19 were composited for fatty acid analysis (Griinari et al., 1998).

To ensure the availability of food ad libitum during rumen sample collection, cows were fed the total amount of feed once daily (0800 h). Rumen fluid was sampled from fistulated cows before feeding (0 h) and at 2, 4, 6, 8, 10, and 12 h after feeding on d 19 of each period. Samples were taken from five different locations in the rumen with a metal filter probe. Samples were taken from nonfistulated cows at 4 h after feeding by rumenocentesis (Nordlund and Garrett, 1994) with the objective of increasing n for this particular timepoint. Rumen pH was determined immediately after the samples were collected (Twin pH-meter model B-213, Spectrum Technologies Inc., Plainfield, IL). One milliliter of rumen fluid was acidified with 20 µl of 50% H₂SO₄ and frozen until analysis for VFA (Supelco Inc., Bellefonte, PA), and 1 ml of rumen fluid was mixed with 20 µl of 50% TCA and frozen until analysis for NH₃-N (Chaney, 1962).

On d 17 and 18 of each period, 25 × 35 cm Dacron polyester bags (52 ± 5 µm pore size, R102 Marvelaire White, N. Erlanger, Blumgardt and Co., Inc. New York, NY) were incubated in triplicate in the rumen for 12, 24, and 48 h to determine in situ DM and NDF disappearance. Bags containing fresh corn silage (24 ± 1.2 g DM) without drying or grinding were placed in a nylon laundry bag in the ventral sac of the rumen. After incubation, bags were washed in a commercial washing machine with cold water for three cycles of 15 min each (Cherney et al., 1990). The in situ bags were dried at

Table 2. Chemical composition of experimental diets.¹

Corn silage ²	0% Tallow			2% Tallow		
	50%	37.5%	25%	50%	37.5%	25%
DM, %	50.5	53.4	56.7	50.5	53.4	56.7
	(% DM)					
CP	16.6	16.6	17.1	17.4	17.4	19.0
NDF	35.1	34.8	34.0	32.8	32.3	31.8
NFC ³	40.6	40.7	40.5	40.3	40.7	39.0
EE ⁴	2.9	2.6	2.6	4.6	4.2	4.3
NE _L ⁵	1.67	1.66	1.65	1.76	1.75	1.73
Fatty acid	3.2	3.2	3.0	4.4	4.1	4.4
	(g/100 g of fatty acids) ⁶					
C14:0	0.3	0.5	0.5	1.0	1.3	1.3
C16:0	16.1	16.8	18.1	20.2	22.3	21.6
C16:1	0.5	0.7	1.0	1.5	1.5	1.6
C18:0	3.3	3.7	3.3	7.9	8.1	7.4
C18:1	23.3	21.2	19.2	26.3	23.7	22.0
C18:2	47.4	42.7	40.3	35.0	31.4	29.8
C18:3	5.3	8.8	11.4	5.5	7.8	10.9
Other	3.8	5.5	6.1	2.6	3.9	5.5

¹Diets contained on average 0.91% Ca, 0.47% P, and 0.25% Mg.

²Diets consisted of 50% forage and 50% concentrate (DM basis). Forage treatments were: 1) 50% of diet DM as corn silage, 2) 37.5% corn silage and 12.5% alfalfa silage, and 3) 25% corn silage and 25% alfalfa silage.

³NFC = Nonfibrous carbohydrates = 100 - (NDF + CP + EE + ash).

⁴EE = Ether extract.

⁵Calculated from NRC (1989).

⁶Values are relative percentages calculated as area under the curve of individual fatty acid divided by total area under the curve. Assumes total area under the curve is 100% fatty acid.

60°C in a forced-air oven for 48 h to determine DM disappearance. Residues from triplicate bags were then composited and analyzed for NDF.

Statistics

Data were analyzed as a 6 × 6 Latin square design with a 2 × 3 factorial arrangement of treatments using the mixed procedure of SAS (SAS User's Guide, 1998). Polynomial effects of forage treatments were tested. For DMI, milk yield and composition data, and rumen measurements at 4 h after feeding, the model included the effects of square, period, fat, linear effect of forage, quadratic effect of forage, fat × linear effect of forage, and fat × quadratic effect of forage. Interaction terms square × fat, square × linear effect of forage, square × quadratic effect of forage, square × fat × linear effect of forage, square × fat × quadratic effect of forage, period × fat, period × linear effect of forage, period × quadratic effect of forage, and period × fat × quadratic effect of forage were excluded from the model at $P > 0.25$. Cow within square was the specified term for the random statement. Period × treatment interactions were not significant ($P > 0.25$) for any of the above-mentioned variables.

Ruminal pH, NH₃-N, and VFA from fistulated cows, and in situ DM and NDF degradability were analyzed

by time as repeated measures. The model included period, time, fat, linear effect of forage, quadratic effect of forage, fat × linear effect of forage, fat × quadratic effect of forage, time × fat, time × linear effect of forage, and time × quadratic effect of forage. The terms specified for the random statement were cow and cow × period × fat × forage. The covariance structure used to best fit the model was selected based on the Akaike's Information Criterion of the Mixed models of SAS (SAS User's Guide, 1998).

Least square means are reported throughout. In all cases, significance was declared at $P < 0.05$ unless otherwise noted. No significant interactions of fat and linear or quadratic effects of forage were detected; results for main effects of fat supplementation and forage treatments will be discussed separately.

RESULTS AND DISCUSSION

Lactation Performance

Fat effects. Treatment effects on DMI, milk production, and milk composition are shown in Table 3. Dry matter intake was 0.8 kg/d lower ($P < 0.003$) for cows fed supplemental tallow than for cows fed diets without fat (23.3 and 24.1 kg of DM/d, respectively). Intake of NE_L was not affected by supplemental tallow (40.0 vs.

Table 3. Least square means for DMI and milk production and composition.

Corn silage ²	0% Tallow			2% Tallow			SE	Significant effects ($P < .1$) ¹		
	50%	37.5%	25%	50%	37.5%	25%		F	L	Q
DMI (kg/d)	23.1	24.6	24.7	22.3	23.7	23.9	0.4	0.003	0.001	0.02
NE _L ³ (Mcal/d)	38.7	40.9	40.7	39.3	41.5	41.5	0.7	NS	0.001	0.02
Milk (kg/d)	35.2	36.6	36.2	37.5	37.0	37.7	1.2	0.02	NS	NS
Fat (%)	3.11	3.17	3.32	2.82	2.97	3.06	0.13	0.001	0.002	NS
Fat (kg/d)	1.06	1.15	1.19	1.05	1.09	1.13	0.04	0.06	0.001	NS
Protein (%)	3.34	3.29	3.30	3.29	3.28	3.25	0.05	NS	NS	NS
Protein (kg/d)	1.15	1.19	1.19	1.23	1.21	1.22	0.04	0.03	NS	NS
SNF (%)	8.93	8.87	8.95	8.93	8.88	8.83	0.08	NS	NS	NS

¹F = Main effect of fat; L = linear effect of forage, and Q = quadratic effect of forage; NS = nonsignificant.

²Diets consisted of 50% forage and 50% concentrate (DM basis). Forage treatments were: 1) 50% of diet DM as corn silage, 2) 37.5% corn silage and 12.5% alfalfa silage, and 3) 25% corn silage and 25% alfalfa silage.

³NE_LI = NE_L intake.

40.8 Mcal NE_L/d for 0 and 2% added tallow, respectively). We observed a similar, but greater negative response for DMI in a previous study (Onetti et al., 2001), where the addition of 2% tallow to diets with CS as the sole forage source decreased DMI by 2 kg/d. No effects on DMI were observed by Smith et al. (1993) when supplementing 2.5% tallow to corn silage-based diets, or diets in which alfalfa hay replaced part of the CS. A recent summary of the literature by Allen (2000) indicated that the addition of unprocessed animal fats to dairy diets results in a negative linear decrease in DMI.

Tallow supplementation increased milk production by 1.4 kg/d ($P < 0.02$). Although not statistically different ($P < 0.13$), the slight numerical increase in NE_L intake may partially account for this increase in milk production. Smith et al. (1993) observed a 3.1% increase in milk production when tallow was included in diets. A summary of the literature showed that tallow supplementation to alfalfa/corn silage-based diets at up to 4 to 5% of the DM increased milk production by 1.7 kg/d (Shaver, 1990).

Milk fat percentage decreased ($P < 0.001$), and there was a trend for a decrease in milk fat yield ($P < 0.06$) when cows were supplemented with 2% tallow (Table 3). We observed a similar response when 2% supplemental tallow was added to diets with CS as the sole forage source in a previous study (Onetti et al., 2001). Milk fat depression (MFD) is associated with changes in the pattern of rumen biohydrogenation that leads to accumulation of *trans* FA in the rumen, and inhibition of milk fat synthesis in the mammary gland (Wonsil, 1994; Kalscheur et al., 1997). The presence of unsaturated FA as substrate, and an altered rumen environment appear to be necessary conditions for MFD (Griinari et al., 1998). In the present study, however, the rumen environment was not dramatically altered. Fur-

thermore, although 50% of the FA in tallow are unsaturated, they are mainly *cis*-9 C18:1, and these particular FA are thought to be hydrogenated directly to C18:0; therefore, they would not be a source of *trans* FA in the rumen (Harfoot and Hazlewood, 1988). Recently, it has been shown (Mosley et al., 2001) that various positional isomers of *trans* C18:1 are formed from hydrogenation of oleic acid by mixed rumen microorganisms in vitro, indicating *cis*-9 C18:1 as a possible source of *trans* C18:1 FA in the rumen. No beneficial effects of replacing part of the CS with AS in the diets were observed for milk fat percentage in the present study. This was in contrast to Smith et al. (1993), who observed that supplemental tallow at 2.5% of the diet DM tended to decrease milk fat percentage with corn silage-based diets, but increased milk fat percentage when part of the CS was replaced with alfalfa hay. The reasons for the difference between our results and those of Smith et al. (1993) are not entirely clear, but several factors might be involved. In their study, dietary NDF content increased substantially when alfalfa hay replaced portion of the CS in the diets. Grant and Weidner (1992) indicated that fat may be more likely to have negative effects at low dietary NDF concentrations. The form in which alfalfa was fed could also be a factor accounting for the differences in results between our study and those of Smith et al. (1993). We used alfalfa silage instead of alfalfa hay. Replacing alfalfa silage with long alfalfa hay has been shown to increase total chewing time, mainly due to increased rumination (Nelson and Satter, 1990; Grant and Weidner, 1992). Rumination activity plays an essential role in stabilizing ruminal pH (Allen, 1997). Kalscheur et al. (1997) demonstrated the direct effect of pH on complete biohydrogenation of fatty acids in the rumen. These authors concluded that low ruminal pH may play a role in the increased production of *trans* FA in the rumen and decreased milk fat produc-

tion. Further, Griinari et al. (1998) showed that both unsaturated fat and low ruminal pH are required to form *trans*-10 C18:1, the *trans* isomer associated with MFD. No data regarding forage particle length were provided in the study by Smith et al. (1993). Forage particle size has also been proposed to modify the effect of fats on rumen fermentation through effects on rates of lipolysis and biohydrogenation (Gerson et al., 1988; Jenkins et al., 1998).

No effect of tallow supplementation on milk protein percentage and SNF content was observed (Table 3). Milk protein yield was increased with supplemental fat ($P < 0.03$). Wu and Huber (1994) summarized 83 treatment comparisons of fat-supplemented diets versus their controls, and, although milk protein percentage was usually depressed, milk protein yield either remained constant or increased.

Forage effects. A quadratic increase ($P < 0.02$) in DM and NE_L intakes was observed as the proportion of AS in the diets increased (Table 3). Feeding 60% of the DM as either AS or CS had no effect on DMI (Broderick, 1985). Smith et al. (1993) and Dhiman and Satter (1997) observed no effect of varying dietary proportion of alfalfa and corn silage on DMI.

No significant effect of forage on milk production was observed despite the increased DMI as the proportion of AS was increased in the diet. Increased DMI may have been the result of a higher NDF digestibility of AS compared with CS. However, a higher DMI may have reduced the rumen retention time (Oba and Allen, 1999), thus decreasing the difference in energy available for milk production between forage treatments. Potentially digestible NDF from AS was more digestible than that of CS (Broderick, 1985). Smith et al. (1993) and Dhiman and Satter (1997) observed no difference in milk production when a portion of the CS was replaced with alfalfa in the diet.

As the proportion of AS in the diets increased, there was a linear increase in milk fat concentration ($P < 0.002$) and milk fat yield ($P < 0.001$). Smith et al. (1993) reported that cows fed CS diets produced less milk fat than those fed alfalfa hay diets. Inclusion of AS in the diets resulted in a more favorable rumen environment in the present study, as indicated by a significant linear increase ($P < 0.01$) in rumen pH at 4 h after feeding (data not shown), and a higher ($P < 0.001$) acetate to propionate ratio (**A:P**) (Table 5). Low ruminal pH may interfere with ruminal biohydrogenation and cause an accumulation of *trans* C18 FA in the rumen. Kalscheur et al. (1997) showed that feeding low fiber-high concentrate diets decreased rumen biohydrogenation of total C18 FA, increased the content of *trans* FA in milk, and decreased milk fat percentage when compared to the same diets with added buffer. As reported by others

(Smith et al., 1993; Dhiman and Satter, 1997), no effect of AS:CS ratio in the diets on milk protein and SNF concentration was observed.

Milk Fatty Acid Composition

Fat effects. Table 4 shows the FA composition of milk fat. As expected, the proportion of short and medium chain FA (C4 to C14) in milk fat was decreased with supplemental fat ($P < 0.001$). Changes in FA profile of milk fat during MFD are consistent (Gaynor et al., 1995) and are characterized by a decreased concentration of short- and medium-chain FA and an increased concentration of long-chain FA. However, long-chain FA response to tallow supplementation in this study was variable. The proportion of C16:0 in milk fat was decreased by only one percentage unit by feeding supplemental tallow ($P < 0.01$), because substantial amounts of C16:0 fatty acids are provided by tallow. No effect of dietary fat was observed for C18:0. The concentration of C18:1 in milk fat increased when tallow was fed ($P < 0.001$). There was a trend for an increase in *cis*-9, *trans*-11 conjugated linoleic acid (**CLA**) when fat was added to the diets ($P < 0.06$). However, the response was minimal and in agreement with data from Chouinard et al. (2000), and might be explained by the low content of linoleic and linolenic acids in tallow. No effect was observed for the *trans*-10, *cis*-12 CLA isomer. Infusion of CLA to dairy cows inhibited milk fat synthesis and this effect was specific for the *trans*-10, *cis*-12 CLA isomer (Chouinard et al., 1999; Baumgard et al., 2000). Linoleic acid concentration in milk fat was not affected by supplemental tallow, and there was a significant but numerically small decrease in C18:3 in milk fat when tallow was fed ($P < 0.001$).

Fat supplementation significantly increased the content of all the *trans* C18:1 isomers in milk fat except for *trans*-11 and *trans*-16, which were not affected by tallow addition. Concentration of *cis*-9 and *cis*-11 increased, *cis*-12 decreased, and *cis*-13 and *cis*-15 C18:1 isomers were not changed when tallow was fed. Total *trans* FA content (*trans* C18:1 isomers plus *trans*-10, *cis*-12 CLA) was 30% higher ($P < 0.001$) in milk fat from tallow-supplemented cows when compared with milk fat from cows that did not receive tallow. Kalscheur et al. (1997) observed an increase of 100% in *trans* C18:1 FA in milk fat of cows fed a diet consisting of 25% of DM as forage without buffer relative to the same diet with buffer (5.8 vs. 2.9 g *trans*-C18:1/100 g milk fat), or diets with 60% DM as forage (average of 3 g *trans*-C18:1/100 g milk fat). Increased concentration of *trans* C18:1 FA, especially *trans*-10 C18:1 and *trans*-10, *cis*-12 CLA, its immediate precursor in the rumen, have been associated with MFD (Griinari et al., 1998;

Table 4. Least square means for milk fatty acid composition of milk fat.

Corn silage ²	0% Tallow			2% Tallow			SE	Significance ($P < .1$) ¹		
	50%	37.5%	25%	50%	37.5%	25%		F	L	Q
Profile	(g/100 g of fatty acids) ³									
C4 to C14	25.3	24.6	24.7	21.4	21.1	21.4	0.6	0.001	NS	NS
C16:0	28.9	29.1	29.1	28.1	27.9	27.8	0.6	0.01	NS	NS
C18:0	8.1	8.7	8.7	8.2	9.1	9.2	0.4	NS	0.01	NS
C18:1	23.2	23.3	23.0	26.3	26.1	25.5	0.6	0.001	NS	NS
C18:1 isomers										
<i>trans</i> -6/8	0.37	0.39	0.37	0.56	0.54	0.50	0.03	0.001	NS	NS
<i>trans</i> -9	0.51	0.56	0.49	0.72	0.65	0.71	0.06	0.001	NS	NS
<i>trans</i> -10	1.3	1.0	0.9	2.2	1.8	1.4	0.2	0.001	0.001	NS
<i>trans</i> -11	1.1	1.1	1.0	0.94	1.1	1.0	0.01	NS	NS	NS
<i>trans</i> -12	0.41	0.41	0.42	0.42	0.50	0.52	0.03	0.01	0.04	NS
<i>trans</i> -16	0.06	0.05	0.05	0.03	0.04	0.05	0.01	NS	NS	NS
<i>cis</i> -9	17.8	18.2	17.8	20.0	19.8	19.4	0.5	0.001	NS	NS
<i>cis</i> -11	0.87	0.92	0.93	0.94	0.94	0.95	0.03	0.04	NS	NS
<i>cis</i> -12	0.41	0.44	0.52	0.22	0.33	0.44	0.03	0.001	0.001	NS
CLA ⁵ c9t11	0.60	0.57	0.58	0.61	0.63	0.62	0.03	0.06	NS	NS
CLA t10c12	0.01	0.01	0.02	0.01	0.02	0.02	<0.01	NS	NS	NS
C18:2	4.5	4.5	4.5	4.2	4.2	4.5	0.3	NS	NS	NS
C18:3	0.33	0.40	0.50	0.28	0.39	0.45	0.02	0.01	0.001	NS
Other ⁴	8.8	8.2	8.4	10.3	10.1	9.5	0.7	0.01	NS	NS
Total <i>trans</i> ⁶	3.7	3.4	3.3	4.9	4.6	4.3	0.2	0.001	0.01	NS

¹F = Main effect of fat; L = linear effect of forage; Q = quadratic effect of forage; NS = nonsignificant.

²Diets consisted of 50% forage and 50% concentrate (DM basis). Forage treatments were: 1) 50% of diet DM as corn silage, 2) 37.5% corn silage and 12.5% alfalfa silage, and 3) 25% corn silage and 25% alfalfa silage.

³Values are relative percentages calculated as area under the curve of individual fatty acid divided by total area under the curve. Assumes total area under the curve is 100% FA.

⁴Calculated as $100 - (C4 \text{ to } C14 + C16:0 + C18:0 + C18:1 + \text{CLA } c9t11 + \text{CLA } t10c12 + C18:2 + C18:3)$.

⁵CLA = Conjugated linoleic acid.

⁶Total *trans* = C18:1 isomers plus *trans*-10, *cis*-12 CLA.

Baumgard et al., 2000; Onetti et al., 2001). Although we did not observe altered rumen conditions for tallow-supplemented cows in this study, our results suggest that incomplete biohydrogenation occurred as indicated by an increased concentration of total *trans* FA in milk fat.

Forage effects. Increasing the AS:CS ratio caused a linear increase in C18:0 ($P < 0.01$) and C18:3 ($P < 0.001$) content of milk fat (Table 4). *Trans*-10 C18:1 and total *trans* FA linearly decreased ($P < 0.001$ and $P < 0.01$, respectively) as AS in the diets was increased; and this corresponded to a higher milk fat percentage and milk fat yield. Including alfalfa in the diets might have resulted in a more stable rumen environment, leading to a more complete rumen biohydrogenation of unsaturated FA. Smith et al. (1993) observed no effect of varying the AS:CS ratio on milk fat fatty acid composition; however, these researchers did not investigate the isomeric profile of long-chain FA.

Rumen Fermentation

Fat effects. Rumen fermentation characteristics that are presented in Table 5 and discussed below corre-

spond to data analyzed by time as repeated measures from fistulated animals ($n = 6$). These results are similar to those obtained at 4 h postfeeding ($n = 18$), unless otherwise stated. No treatment \times time interactions were detected for any of the variables measured. No effect of supplemental fat on rumen pH was observed (Table 5). Rumen pH values at 4 h after feeding were 0.1 pH units lower ($P < 0.05$) for cows that did not receive tallow in the diets. This is likely due to the higher DMI of cows that were not supplemented with fat. Contrary to previous results (Ikwuegbu and Sutton, 1982; Broudicou et al., 1994; Onetti et al., 2001), $\text{NH}_3\text{-N}$ concentration in the rumen tended to increase ($P < 0.06$) when fat was included in the diets. At 4 h postfeeding, the difference was significant ($P < 0.01$). This difference was partially accounted for by the effect of the 25% CS-25% AS treatment with supplemental tallow and was probably a reflection of the higher content of CP (19% CP, DM basis) of this treatment when compared to the rest of the dietary treatments.

Total VFA concentration in the rumen tended to decrease ($P < 0.09$) when tallow was fed (Table 5). These data are consistent with the higher pH observed for tallow-supplemented cows. Feeding tallow had very lit-

Table 5. Least square means for ruminal pH, concentration of total VFA and ammonia, and molar proportion of VFA.

Corn silage ²	0% Tallow			2% Tallow			SE	Significant effects ($P < $) ¹		
	50%	37.5%	25%	50%	37.5%	25%		F	L	Q
pH	6.2	6.1	6.2	6.2	6.2	6.2	0.1	NS	NS	NS
NH ₃ -N (mg/dl)	13.3	12.0	13.2	14.0	13.1	16.7	0.2	0.06	NS	NS
VFA (mM)	96.6	96.9	99.4	92.8	96.4	95.8	2.3	0.07	0.09	NS
	(mol/100 mol)									
Acetate	51.5	52.8	55.4	49.6	51.9	54.4	1.1	0.05	0.001	NS
Propionate	27.1	28.4	25.2	28.6	27.8	26.2	1.1	NS	0.01	NS
Butyrate	15.4	13.4	13.7	15.2	14.3	13.4	0.6	NS	0.001	NS
Isobutyrate	0.8	0.8	0.9	0.9	0.9	1.1	<0.1	0.003	0.001	0.05
Isovalerate	1.9	1.7	1.8	2.2	1.9	2.0	0.14	0.04	NS	NS
Valerate	3.4	3.0	3.0	3.4	3.2	3.1	0.28	NS	NS	NS
A:P ³	1.92	1.90	2.22	1.78	1.88	2.11	0.11	NS	0.001	0.09

¹F = main effect of fat; L = linear effect of forage, and Q = quadratic effect of forage; NS = nonsignificant.

²Diets consisted of 50% forage and 50% concentrate (DM basis). Forage treatments were: 1) 50% of diet DM as corn silage, 2) 37.5% corn silage and 12.5% alfalfa silage, and 3) 25% corn silage and 25% alfalfa silage.

³A:P = Acetate:propionate ratio.

tle effect on molar proportions of rumen VFA. The molar proportion of acetate decreased ($P < 0.05$), and propionate and A:P were not affected when tallow was included in the diets. Tallow supplementation to diets with CS as the sole forage source (Onetti et al., 2001) resulted in increased ruminal propionate and decreased A:P. Molar proportions of butyrate and valerate were similar for all diets, and that of isovalerate ($P < 0.04$) and isobutyrate ($P < 0.003$) were increased when tallow was fed. These changes probably were of minimal biological significance. In this study, MFD was observed without significant changes in rumen pH and molar proportions of VFA. This indicates that responses in milk fat percentage are independent of the effects of fat on rumen fermentation, and the ability to detect a fat \times forage interaction is probably not dependent on obtaining an altered rumen environment. A summary of feeding trials in which tallow was supplemented up to 5% of the diet DM with CS and alfalfa-based diets showed minimal effects on rumen fermentation (Shaver, 1990).

No effect of supplemental tallow on DM and NDF disappearance was observed (data not shown). Forty-eight-hour DM and NDF disappearance averaged 68.5 and 35.3%, respectively. Supplementing tallow and choice white grease at 2 or 4% of the diet DM when corn silage comprised the sole forage source did not affect rumen DM and NDF disappearance (Onetti et al., 2001). Smith et al. (1993) observed a trend for lower NDF digestibility when supplemental fats were fed with CS diets compared with alfalfa hay diets. Ben Salem et al. (1993) observed that 7% rapeseed oil depressed digestibility of fiber more when added to a CS-based diet than a grass hay-based diet.

Forage effects. Rumen pH and NH₃-N concentrations were not affected by forage treatments when data were analyzed by time as repeated measures (Table 5). At 4 h after feeding, there was a linear increase ($P < 0.01$) in pH as the proportion of CS in the diet was decreased (5.7, 5.8, and 5.9, and 5.8, 5.9, and 6.0 for 50, 37.5, and 25% CS in the diet, with 0 and 2% tallow, respectively). Increased pH might reflect the higher buffering capacity of alfalfa forage. Dhiman and Satter (1997) observed no changes in rumen pH when diets with different proportions of AS and CS were fed. A quadratic increase in NH₃-N concentration ($P < 0.01$) was observed at 4 h postfeeding in response to increasing AS in the diets. This is likely due to the high content of NPN in alfalfa silage and the extensive ruminal degradation of the protein (Broderick, 1992).

There was a trend ($P < 0.06$) for a linear increase in total VFA concentration as the proportion of AS increased in the diets. These data do not coincide with the increase in rumen pH observed 4 h postfeeding for cows fed a higher proportion of alfalfa in the diets. Molar percentage acetate increased ($P < 0.001$), that of propionate decreased ($P < 0.02$), and A:P increased ($P < 0.001$) when the dietary AS:CS ratio was increased. These results are consistent with those of Satter and Dhiman (1993), in which A:P decreased significantly with each increase of CS in the diets. Broderick (1985) reported that feeding 60% of the DM as CS reduced acetate and A:P and increased propionate in rumen fluid compared with feeding AS at 60% of the diet DM.

No significant effects of forage treatments were observed for in situ DM and NDF disappearance (data not shown). Smith et al. (1993) observed a trend for lower NDF digestibility when CS diets were compared

to diets in which alfalfa hay replaced part of the CS. Potentially digestible NDF total tract digestibility was highest for alfalfa hay, intermediate for AS, and lowest for CS (92.8, 82.8, and 71.8%, respectively) when these were fed as the sole forage at 60% of the diet DM (Broderick et al., 1985).

CONCLUSIONS

Tallow supplementation at 2% of diet DM to cows consuming CS and alfalfa silage-based diets resulted in lower DMI and milk fat production. Replacing CS from high-grain yielding hybrids with AS did not alleviate the negative effects of tallow on DMI and milk fat production. Increasing the proportion of AS increased DMI, milk fat percentage, and yield regardless of fat content of the diet. Our results indicate that the beneficial effects of replacing CS with AS in the diet are not dependent on the level of dietary fat. Despite the minimal effects of tallow on rumen fermentation in this study, the proportion of *trans* FA in milk fat increased by 30% when tallow was fed. Increasing the AS:CS ratio in the diets resulted in an increase in ruminal pH and A:P. *Trans* FA in milk fat decreased when the proportion of AS was increased in the diets. These results support the role of *trans* FA in MFD. However, the causes for the formation of these FA in the rumen in this particular study are not clear.

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