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Dried Distillers Grains Plus Solubles with Corn Silage or Alfalfa Hay as the Primary Forage Source in Dairy Cow Diets[1]

D. H. Kleinschmit,[2] D. J. Schingoethe,[3] A. R. Hippen, and K. F. Kalscheur
Dairy Science Department, South Dakota State University, Brookings 57007-0647

ABSTRACT

Nine multiparous (250 ± 6 d in milk) and 3 primiparous (204 ± 6 d in milk) Holstein cows were utilized in a 3 × 3 Latin square design to evaluate the lactation performance of cows fed a diet containing dried distillers grains plus solubles (DDGS) with either corn silage or alfalfa hay as forage. Cows were fed total mixed diets containing corn silage (CS), 50% corn silage and 50% alfalfa hay (CSAH), or alfalfa hay (AH) as the forage source. All diets had a 50:50 forage-to-concentrate ratio, contained 15% DDGS, and were formulated to be equal in metabolizable protein. Dry matter intake increased when cows were fed CSAH (24.9 kg/d) compared with CS (21.9 kg/d) and AH (20.9 kg/d). Yields of milk (26.5, 28.4, 29.0 kg/d for CS, CSAH, and AH, respectively) increased linearly as proportions of alfalfa fed increased but 4% fat-corrected milk and energy-corrected milk were not affected by treatment. Feed efficiency (1.28, 1.23, and 1.45 kg of energy-corrected milk/kg of intake) improved when AH was fed compared with CS or CSAH. Milk fat concentration (3.67, 3.55, and 3.49%) decreased linearly when alfalfa replaced corn silage, but was observed only in primiparous cows, not multiparous cows. Milk protein concentration (3.32, 3.29, and 3.29%) was not affected by diet although yield (0.90, 0.96, and 0.98 kg/d) tended to increase linearly when alfalfa was added to the diet. This may have been due to an increase in essential amino acid (AA) availability and uptake by the mammary gland or to greater crude protein intake in cows fed AH. In addition, replacing corn silage with alfalfa increased the uptake of Lys by the mammary gland. Methionine was the first-limiting AA based on the transfer efficiency of AA in arterial plasma to milk protein. However, Lys was the first-limiting AA in CS and CSAH and Met was first limiting in AH for mammary gland extraction efficiency of AA from plasma. In conclusion, replacing corn silage with alfalfa hay in diets containing 15% DDGS increased milk yield and tended to increase milk protein yield linearly in cows during late lactation. Feeding alfalfa hay as the sole forage source improved feed efficiency compared with diets containing corn silage.

Key words: dried distillers grains plus solubles, forage, amino acids

INTRODUCTION

Dried distillers grains plus solubles (**DDGS**) is commonly used as a protein source for dairy cattle. The use of this coproduct has generally been limited to about 20% of the dietary DM because corn-based products tend to be low in Lys (Schingoethe, 1996; Nichols et al., 1998), which along with Met, is one of the first-limiting AA for milk protein synthesis (Schwab et al., 1992). Many of the past studies that evaluated feeding DDGS used diets in which one-half or more of the forage fraction contained corn silage. Overall, those studies found Lys to be the first-limiting AA (Nichols et al., 1998; Liu et al., 2000).

Many dairy producers, especially those in the western United States, feed diets that consist primarily of alfalfa in the forage portion. Most alfalfa hay fed to dairy cattle has a CP content at or exceeding 20% (DM basis) with approximately 80% being ruminally degradable (NRC, 2001). Therefore, feeding a source of RUP may complement alfalfa hay in a dairy ration quite well. Past studies determined that supplementing alfalfa-based diets to dairy cattle with a source of RUP increased the yields of milk and protein and the concentration of milk protein (Faldet and Satter, 1991). Compared with corn silage, the protein in alfalfa hay has a much greater concentration of Lys (NRC, 2001). As a result, replacing corn silage with alfalfa in diets containing corn-based protein might increase the milk protein content and yield via improved Lys availability. Holter et al. (1992) supplemented corn silage-based dairy cattle diets with corn gluten meal and milk protein content decreased compared with soybean meal, which was attributed to a Lys deficiency. In contrast,

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[2] Current address: Agri-King Inc., Box 208, Fulton, IL 61252.

[3] Corresponding author: david.schingoethe@sdstate.edu

Robinson et al. (1991) found that feeding corn gluten meal in an alfalfa-based diet had no effect compared with feeding soybean meal. Broderick et al. (1990) observed that cows fed a protein supplement containing DDGS and corn gluten meal in alfalfa-based rations had similar yields of milk and milk components as those fed expeller or solvent-extracted soybean meal.

We are not aware of research that has evaluated the lactation performance of dairy cattle fed only corn silage or alfalfa hay with DDGS. The objectives of this study were to evaluate the performance of lactating cows and the efficiency of AA utilization for milk protein synthesis when fed a diet containing primarily corn-based protein sources with the forage portion containing corn silage or alfalfa hay.

MATERIALS AND METHODS

Nine multiparous (250 ± 6 DIM) and 3 primiparous Holstein cows (204 ± 6 DIM) were used in four 3 × 3 Latin squares with 28-d periods. Cows were cared for in accordance with protocols approved by South Dakota State University Animal Care and Use. Cows were assigned to squares by parity and milk yield with 1 square for the primiparous cows and the 3 other squares being multiparous cows. Nine of the cows were pregnant at the beginning of the experiment with the average days of gestation being 62 ± 14 d. Weeks 1 and 2 of each period were used for adjustment to diets, and wk 3 and 4 were used for data collection.

Treatments consisted of TMR containing corn silage as the sole forage source (**CS**), corn silage and chopped alfalfa hay (50:50) as the forage source (**CSAH**), or chopped alfalfa hay as the sole forage source (**AH**). All diets contained 15% DDGS and consisted of 50% forage and 50% concentrate (Table 1). With the exception of 4% SBM in all diets, all true protein sources in the concentrate mixes originated from corn-based products. Urea was added to CS and CSAH to ensure that RDP concentration was not limiting for microbial synthesis (NRC, 2001). Diets were balanced to provide an MP balance of 81 g/d (NRC, 2001) for a 590-kg Holstein cow consuming 24 kg/d of DM and producing 38 kg/d of milk containing 3.70% fat and 3.10% protein. The administration of bST was discontinued 1 mo before the study, and average milk production decreased to 34 kg/d at the beginning of the experiment.

Alfalfa was chopped weekly with a New Direction 500 Mixer-Feeder wagon equipped with a hay chopper (New Direction Equipment, Sioux Falls, SD). Dry matter content of corn silage was determined weekly and the amount of corn silage (as fed) was adjusted to account for weekly differences in DM. Water was added to the CSAH and AH diets at the time of feeding to ensure that DM content was similar among all diets so to not affect intake or sorting. Cows were housed in a free-stall barn and individually fed once daily (0800 h) for ad libitum consumption allowing for 10% orts using a Calan Super Data Ranger and Calan Broadbent feeder doors (American Calan, Inc., Northwood, NH). Feed intakes were recorded daily.

Samples of corn silage, alfalfa hay, concentrate mixes, and TMR were collected weekly for analyses and stored at −20°C before being composited by period and dried at 55°C for 48 h in a Despatch oven (style V-23; Despatch Oven Co., Minneapolis, MN). An undried sample was evaluated for particle size as described by Kononoff et al. (2003) and as shown in Table 2. Composites were ground through a 4-mm screen of a Wiley mill (model 3; Arthur H. Thomas Co., Philadelphia, PA) and then reground through an ultracentrifuge mill (Brinkman Instruments Co., Westbury, NY) with a 1-mm screen. Composites were analyzed for NDF (Van Soest

Table 1. Ingredients of diets containing 100% corn silage (CS), 50% corn silage and 50% alfalfa hay (CSAH), or 100% alfalfa hay (AH) as forage fed to lactating dairy cows

	Diet		
Ingredient	CS	CSAH	AH
	(% of DM)		
Corn silage	50.07	25.01	0.00
Alfalfa hay	0.00	25.01	50.01
Ground corn	25.59	27.85	30.04
Dried distillers grains plus solubles	15.02	15.00	15.01
Corn gluten meal	1.64	0.81	0.00
Soybean meal, 44% CP	4.00	4.00	4.00
Urea	0.98	0.49	0.00
Salt	0.50	0.50	0.50
Dicalcium phosphate	0.15	0.15	0.15
Limestone	1.70	0.85	0.00
Magnesium oxide	0.06	0.04	0.00
Dairy micro premix[1]	0.23	0.23	0.23
Vitamin E premix (44,000 IU/kg)	0.06	0.06	0.06

[1] Contained 10% Mg; 2.6% Zn; 1.7 mg/kg of Mn; 4,640 mg/kg of Fe; 4,712 mg/kg of Cu; 396 mg/kg of I; 119 mg/kg of Co; 140 mg/kg of Se; 2,640,000 IU/kg of vitamin A; 528,000 IU/kg of vitamin D_3; and 10,560 IU/kg of vitamin E.

Table 2. Particle size distribution (% of particles on an as-fed basis) of diets containing 100% corn silage (CS), 50% corn silage and 50% alfalfa hay (CSAH), and 100% alfalfa hay (AH) as forage fed to lactating dairy cows

	Diet		
Particle size (mm)[1]	CS	CSAH	AH
>19	3.8	10.8	16.5
19 to 8.0	42.7	34.2	16.6
8.0 to 1.18	36.3	43.7	52.3
<1.18	17.2	11.3	14.5

[1] Kononoff et al., 2003.

et al., 1991) and ADF (Robertson and Van Soest, 1981) sequentially by using an Ankom fiber analyzer with the filter bag technique (Ankom Technology Corp., Fairport, NY); CP, ether extract, ash, Ca, P, Mg, K, and S were analyzed according to AOAC (2000). Samples of TMR were prepared for fatty acid analysis as butyl esters in an adapted method of that described by Sukhija and Palmquist (1988) for analysis using gas chromatography (model 6890, Hewlett-Packard, Palo Alto, CA). The adaptation used was as described by Loor et al. (2005) with samples analyzed using a flame-ionization detector. The injector port temperature was 230°C with a split ratio of 20:1. The column was 100 m and inside diameter was 0.25 mm (CP-Sil 88, Varian, Lake Forest, CA). Carrier gas was helium at a rate 2.0 mL/min. Initial oven temperature was 50°C held for 1 min and then increased to 145°C at a rate of 5°C per min and held for 30 min. Temperature was then increased at 10°C/min to 190°C and held for 30 min. Finally, the temperature was raised at 5°C/min to 210°C and held for 35 min. The total run per sample was 123.5 min. Samples were corrected to 100% DM basis by drying an aliquot of the composites at 105°C for 24 h.

Cows were milked at 0600, 1400, and 2100 h. Milk samples were collected at the end of wk 3 and 4 of each period from each milking on 2 consecutive days. Milk samples were mixed by gentle inversion and composited in volumes corresponding to the respective milk weights for each cow on the sampling day and analyzed by Heart of America DHIA Laboratory (Manhattan, KS) according to approved procedures of AOAC (2000). Milk true protein, fat, and lactose were determined by near infrared spectroscopy (Bentley 2000 Infrared Milk Analyzer, Bentley Instruments, Chaska, MN). Concentration of MUN was determined using chemical methodology based on a modified Berthelot reaction; AOAC, 2000 (ChemSpec 150 Analyzer, Bentley Instruments), and somatic cells were counted using a flow cytometer laser (Somacount 500, Bentley Instruments). Butyl esters of fatty acids were prepared as previously described.

Cows were weighed and scored for body condition on a scale of 1 to 5 (Wildman et al., 1982) approximately 3 h after feeding on 3 consecutive days at the end of each period. Blood was collected from the coccygeal region to represent arterial blood and the subcutaneous abdominal mammary vein simultaneously into heparin Vacutainer tubes (Becton Dickinson and Co., Franklin Lakes, NJ) at the end of each period. Plasma was obtained by centrifuging (4°C) at 500 × g and supernatant stored at −20°C until analyzed for AA via HPLC (model 1100, Agilent Technologies, Inc., Palo Alto, CA) with a PCX 5200 postcolumn derivatizer (Pickering Laboratories Inc., Mountain View, CA) as described by Mondina et al. (1972), Pickering (1989), and Grunau and Swiader (1992). Amino acid concentrations in the coccygeal artery, subcutaneous abdominal mammary vein, and milk (Jacobson et al., 1970), in addition to estimated mammary blood flow from an equation derived from antipyrine absorption in 21 experiments (mammary blood flow, mg/min = 1.0 + 0.42 × milk yield, kg/d; Kronfeld et al., 1968), were used to calculate apparent AA uptake, transfer efficiency, and extraction rates to evaluate the limiting essential AA (**EAA**) of each diet.

After discarding the first 150 mL to minimize saliva contamination, approximately 250 mL of ruminal fluid was collected by applying vacuum pressure to an esophageal tube fitted with a suction strainer approximately 3 h after feeding on 2 consecutive days at the end of each period. A 10-mL aliquot was mixed with 2 mL of 25% (wt/vol) metaphosphoric acid and frozen at −20°C until centrifuged at 30,000 × *g* and analyzed for concentrations of ammonia-N (Weatherburn, 1967) and VFA. Concentrations of VFA were measured using a gas chromatograph (model 6890; Hewlett-Packard, Avondale, PA) equipped with a 0.25-mm i.d. × 15-m column (Nukol, 17926 to 01C, Supelco, Inc., Bellefonte, PA). The split ratio in the injector port (250°C) was 100:1 with a flow of 1.3 mL/min of He. Column and detector temperature were maintained at 130 and 225°C, respectively.

The means of DMI, milk yield, and milk composition during wk 3 and 4 of each period were used for statistical analysis. Amino acid profile from plasma and rumen fluid components during wk 4 were used for statistical analysis. Analysis of variance was conducted using the MIXED procedure (Littell et al., 1996) of SAS (SAS Institute, 1999). Cow served as the experimental unit. The model was Y = treatment + square + period + treatment × square + treatment × period + square × period + treatment × square × period, with cow(square) being designated as a random variable. Insignificant interactions were removed from the model for each variable tested. Linear and quadratic contrasts were designed to test for the effect of increasing the amount of alfalfa hay in the diet. Significance was declared at $P < 0.05$ and tendencies were noted at $P < 0.10$.

RESULTS AND DISCUSSION

Composition of Feeds

The concentrations of CP and RDP increased and RUP decreased as alfalfa replaced corn silage in the diet (Table 3). The diets were balanced to provide similar amounts of MP to the small intestine as estimated by NRC (2001); however, the CP in corn silage was greater and the CP in alfalfa was less than that measured before the study and DMI differed among diets (discussed later). Due to these factors, the estimated RUP

Table 3. Chemical composition of concentrate mixes, corn silage, alfalfa hay, and diets containing 100% corn silage (CS), 50% corn silage and 50% alfalfa hay (CSAH), and 100% alfalfa hay (AH) as forage fed to lactating dairy cows[1]

	Concentrate mix			Corn	Alfalfa	Diet		
Measurement	CS	CSAH	AH	silage	hay	CS	CSAH	AH
DM,[2] %	90.0	89.4	89.6	31.9	89.1	46.4	45.7	44.6
	(% of DM)							
CP	25.8	22.5	19.0	8.09	18.2	16.9	18.3	18.7
RDP,[3] % of CP	ND[4]	ND	ND	ND	ND	62.6	65.4	69.0
Balance,[5] g/d	ND	ND	ND	ND	ND	117	437	603
RUP,[3] % of CP	ND	ND	ND	ND	ND	37.4	34.6	31.0
Balance,[5] g/d	ND	ND	ND	ND	ND	143	360	−104
MP Balance,[5] g/d	ND	ND	ND	ND	ND	117	296	−85
Lys,[6] % of MP	ND	ND	ND	ND	ND	5.61	5.73	6.06
Met,[6] % of MP	ND	ND	ND	ND	ND	1.94	1.91	1.93
Lys:Met	ND	ND	ND	ND	ND	2.89	3.00	3.14
Ether extract	6.36	6.16	6.65	3.59	2.90	4.44	4.58	4.26
Fatty acids	ND	ND	ND	ND	ND	3.17	3.47	3.35
NDF	19.9	20.5	21.6	47.6	44.7	32.9	33.5	34.0
ADF	5.98	6.12	6.60	25.9	32.6	15.7	18.0	19.7
Ash	7.53	5.82	4.18	4.67	8.74	6.22	6.40	6.66
Ca	1.33	0.72	0.32	0.31	1.24	0.80	0.83	0.86
P	0.47	0.46	0.48	0.24	0.24	0.37	0.35	0.35
Mg	0.28	0.25	0.23	0.31	0.30	0.31	0.31	0.29
K	0.68	0.64	0.68	0.98	1.98	0.88	1.12	1.33
S	0.34	0.32	0.32	0.12	0.20	0.25	0.25	0.27
NE_L,[3] Mcal/kg	ND	ND	ND	ND	ND	1.56	1.56	1.58

[1] Chemical analyses conducted in duplicate by period, except for mineral analyses, which were composited and conducted by a commercial laboratory (Dairyland Laboratories, Inc., Arcadia, WI).

[2] DM content of diets included added water.

[3] Calculated based on NRC (2001).

[4] Not determined.

[5] Based on estimated requirements (NRC, 2001) for a 590-kg Holstein cow producing 34 kg of milk/d with 3.70% fat and 3.10% true protein, with milk yield being a known parameter at the beginning of the experiment and all others being estimated.

[6] Amino acid concentrations were based on book values of feed ingredients from NRC (2001).

and MP balances for cows fed CS and CSAH were in a positive balance, and cows fed AH had negative balances for RUP and MP.

All diets contained similar concentrations of ether extract and total fatty acids. The concentrations of C16:0, C16:1, C18:0, and C18:3 increased and C18:1 and C18:2 decreased as alfalfa was added to the diets (Table 4). In all diets, approximately 45% of the fatty acids consisted of C18:2, which was due primarily to the inclusion of DDGS (Schingoethe et al., 1999). The concentration of NDF did not differ among diets; however, ADF content increased with added alfalfa.

Table 4. Fatty acid composition (mg/100 mg of fatty acids) of diets containing 100% corn silage (CS), 50% corn silage and 50% alfalfa hay (CSAH), and 100% alfalfa hay (AH) as forage fed to lactating dairy cows

	Diet		
Fatty acid	CS	CSAH	AH
$C_{14:0}$	0.30	0.32	0.28
$C_{16:0}$	18.78	20.44	21.88
$C_{16:1}$	0.34	1.09	1.13
$C_{18:0}$	2.43	2.54	2.73
$C_{18:1}$	23.66	22.60	21.88
$C_{18:2}$	46.69	45.13	43.29
$C_{18:3}$	2.64	3.81	4.76
Other	5.19	4.17	4.05

Milk Yield and Composition

A quadratic relationship ($P < 0.01$) in DMI was observed such that cows fed CSAH (24.9 kg/d) consumed more feed compared with those fed CS (21.9 kg/d) and AH (20.9 kg/d; Table 5). A quadratic ($P < 0.01$) response was also observed in CP intake. In this study, there was a linear ($P < 0.05$) increase in milk yield and a tendency for yield of ECM to increase linearly ($P < 0.10$) when alfalfa was added to the diet. However, this effect was not observed in the yield of 4% FCM. In past studies (Broderick, 1985; Dhiman and Satter, 1997), feeding alfalfa-based diets did not affect milk production compared with diets based on corn silage; therefore, the linear effects observed in this experiment may have been due to an interaction of the DDGS with the forage source. Voss et al. (1988) fed a corn silage-based diet

Table 5. Dry matter and CP intake, milk yield and composition, and feed efficiency for lactating dairy cows fed diets containing 100% corn silage (CS), 50% corn silage and 50% alfalfa hay (CSAH), and 100% alfalfa hay (AH) as forage[1]

	Diet				*P*-value[2]		
Item	CS	CSAH	AH	SE	Trt	L	Q
DMI, kg/d	21.9	24.9	20.9	1.1	0.02	0.47	<0.01
CP intake, kg/d	3.70	4.55	3.91	0.20	0.01	0.38	<0.01
Milk yield, kg/d	26.5	28.4	29.0	2.8	0.13	0.05	0.53
4% FCM, kg/d	25.0	26.7	26.7	1.6	0.20	0.12	0.37
ECM,[3] kg/d	27.4	29.2	29.5	1.8	0.19	0.10	0.44
Fat, %	3.67	3.55	3.49	0.08	0.08	0.03	0.61
Fat yield, kg/d	0.96	1.02	1.01	0.06	0.31	0.25	0.30
Protein, %	3.36	3.33	3.33	0.05	0.40	0.28	0.40
Protein yield, kg/d	0.88	0.94	0.96	0.06	0.17	0.07	0.70
Lactose, %	4.71	4.72	4.74	0.08	0.87	0.62	0.88
Lactose yield, kg/d	1.27	1.36	1.38	0.09	0.21	0.10	0.52
MUN, mg/dL	15.1	16.5	16.9	0.4	0.01	<0.01	0.17
FE,[4] ECM/DMI	1.28	1.23	1.45	0.10	0.02	0.03	0.03
N eff,[5] protein yield/CP intake	0.245	0.213	0.252	0.017	<0.01	0.41	<0.01
SCC, $\log_{10}$/mL	5.34	5.33	5.22	0.16	0.63	0.39	0.67
BW change, kg/28 d	17.6	28.3	14.1	5.2	0.04	0.49	0.02
BCS	3.47	3.44	3.28	0.09	0.01	0.007	0.20
BCS change/28 d	0.12	0.22	–0.01	0.07	0.05	0.15	0.05

[1] Least squares means.

[2] Trt = treatment effect; L = linear contrast; Q = quadratic contrast.

[3] ECM = 0.3246 × milk yield (kg) + 12.86 × fat yield (kg) + 7.04 × protein yield (kg); Orth, 1992.

[4] Feed efficiency: ECM (kg/d)/DMI (kg/d).

[5] Nitrogen efficiency (Milk protein, kg/CP intake, kg).

that replaced soybean meal with 11% DDGS and 5.5% corn gluten meal to cows in early lactation. Cows fed the corn protein diet had lower milk yields compared with the control. However, a second experiment with a 50:50 mix of corn silage and alfalfa haylage and inclusion of corn protein had no effect on milk yield compared with the control.

Milk fat content decreased linearly ($P < 0.03$) with added alfalfa, but no differences in milk fat yield were observed among diets. There was a square × treatment interaction, such that the decrease in milk fat content occurred in the square containing primiparous cows but not in squares containing multiparous cows. Similarly, Dhiman and Satter (1997) found that primiparous cows fed solely alfalfa silage tended to produce milk with a lower fat concentration compared with those fed two-thirds or one-third alfalfa silage with corn silage, whereas no effect occurred in multiparous cows. The authors could not explain those results. In a meta-analysis that evaluated the effect of feeding DDGS to dairy cattle, it was found that cows fed DDGS in alfalfa-based diets produced milk with a similar fat content compared with those fed a corn silage-based diet (Kalscheur, 2005). However, none of the studies reviewed comparatively measured the impact of feeding DDGS with different forages to dairy cattle. This finding may also be attributed to a dilution effect because milk fat yield was not affected.

The estimated duodenal supplies of Lys (138, 157, and 136 g/d for CS, CSAH, and AH, respectively) and Met (48, 52, and 43 g/d for CS, CSAH, and AH, respectively), as estimated by NRC (2001), were greater in the CSAH compared with the CS and the AH diets. There was a tendency for protein yield to increase linearly ($P < 0.07$) with increased dietary alfalfa (Table 4). The CSAH and AH diets probably provided a more ideal ratio of Lys to Met (Schwab and Ordway, 2004), as discussed later. The linear tendency in milk protein yield corresponded to the linear increase in milk yield because milk protein content was not affected by diet.

As expected, treatment did not affect the lactose content or yield in milk; however, there was a tendency ($P < 0.10$) for a linear increase in lactose yield when greater concentrations of alfalfa were fed. Feeding alfalfa in the diets increased ($P < 0.01$) concentrations of MUN linearly, related to increasing dietary CP content in those diets. The target MUN values for dairy cows to maximize N efficiency and minimize N excretion range from approximately 8 to 12 mg/dL (Kohn et al., 2002). Based on the MUN values in this study, protein was not utilized efficiently by the cows.

Feed efficiency had a quadratic response ($P < 0.03$) as cows were fed increasing amounts of alfalfa. This was the result of less DMI by cows fed AH compared with CSAH and numerically greater ECM yield in cows fed AH compared with CS. Efficiency of N utilization

Table 6. Fatty acid composition (mg/100 mg of fatty acids) of milk from lactating dairy cows fed diets containing 100% corn silage (CS), 50% corn silage and 50% alfalfa hay (CSAH), and 100% alfalfa hay (AH) as forage[1]

	Diet				P-value[2]		
	CS	CSAH	AH	SE	Trt	L	Q
Fatty acid							
$C_{4:0}$ to $C_{14:1}$	28.9	29.5	28.2	0.50	0.02	0.09	0.01
$C_{16:0}$	27.4	26.7	25.9	0.41	<0.01	<0.01	0.94
$C_{18:0}$	10.6	10.8	11.5	0.35	<0.01	<0.01	0.22
$C_{18:1}$	22.5	22.3	22.9	0.42	0.31	0.39	0.21
$C_{18:1}$ isomers							
Trans-6	0.26	0.18	0.22	0.05	0.40	0.48	0.25
Trans-9	0.28	0.31	0.30	0.01	0.01	0.05	0.01
Trans-10	0.47	0.51	0.48	0.02	<0.01	0.16	<0.01
Trans-11	1.26	1.22	1.26	0.05	0.76	0.93	0.46
Cis-6	0.22	0.28	0.32	0.04	0.29	0.12	0.98
Cis-9	19.3	19.0	19.5	0.40	0.40	0.55	0.22
Cis-11	0.78	0.80	0.83	0.02	0.08	0.03	0.78
$C_{18:2}$	3.96	4.24	4.37	0.11	<0.01	<0.01	0.43
$C_{18:3}$	0.29	0.43	0.59	0.03	<0.01	<0.01	0.79
CLA[3] *cis*-9, *trans*-11	1.15	1.08	1.22	0.12	0.62	0.63	0.40
Other	5.10	4.95	5.36	0.14	0.10	0.19	0.09

[1] Least squares means.

[2] Trt = treatment effect; L = linear contrast; Q = quadratic contrast.

[3] Conjugated linoleic acid.

was lower in CSAH compared with CS and AH, which was the result of a greater intake of CP but similar protein yield as those observed in the other diets. Past research (Cressman et al., 1980) showed that increasing dietary CP from 12 to 15 to 18% decreased efficiency of dietary N utilization for milk protein synthesis. Overall, greater N intake increases N excretion (Rotz, 2004). Somatic cell count was similar for all cows.

The average BW of cows in this study was 702 ± 28 kg and the average BCS was 3.36 ± 0.09 (Table 4). Cows fed the CSAH diet gained more weight ($P < 0.02$) and more body condition ($P < 0.05$) within each period compared with those fed AH, with CS being numerically intermediate but similar to both treatments. Generally, BCS throughout the experiment changed little in cows fed AH. The cows in this experiment were in late lactation and were gaining weight throughout the experiment, thus indicating that energy and nutrient requirements were met for milk production even though DMI differed among treatments. The effects on BW and BCS were not surprising because cows fed CSAH consumed more DM compared with other diets.

The concentrations of fatty acids in milk are shown in Table 6. The concentrations of short and medium-chain fatty acids ($C_{4:0}$ to $C_{14:1}$), which are totaled together for brevity, were quadratically ($P < 0.01$) affected by treatment such that cows fed CSAH produced milk with a greater concentration of these fatty acids compared with AH and CS. Even though diets with alfalfa contained a greater concentration of $C_{16:0}$ compared with corn silage, the concentration of $C_{16:0}$ decreased linearly ($P < 0.01$) when alfalfa replaced corn silage. Because $C_{16:0}$ originates from both dietary and de novo synthesis, there was probably less de novo synthesis of this fatty acid when alfalfa was fed, possibly explaining the linear decrease in milk fat concentration. The concentration of $C_{18:0}$ increased linearly ($P < 0.01$) when alfalfa replaced corn silage, which was most likely the result of greater concentrations of this fatty acid being present in diets that contained alfalfa. There was no effect on the concentration of $C_{18:1}$ in milk fat; however, some of the individual isomers were affected, including the *cis*-11 isomer, which increased linearly as alfalfa replaced corn silage. There was also a quadratic ($P < 0.01$) effect on the *trans*-9 isomer. The *trans*-10 isomer of $C_{18:1}$ was greater from cows fed CSAH compared with those fed CS and AH as indicated by the quadratic response ($P < 0.01$); however, these small differences were probably not biologically significant. The concentrations of $C_{18:2}$ and $C_{18:3}$ in milk fat increased linearly when cows were fed alfalfa in place of corn silage. Increasing the concentration of alfalfa in the diet increased the concentration of dietary $C_{18:3}$, thus explaining the greater concentration of these fatty acids in milk. Concentrations of the *cis*-9, *trans*-11 CLA isomer were not affected by diet, and the *trans*-10, *cis*-12 isomer was not detected in the milk regardless of treatment.

Amino Acids

The concentrations of branched-chain AA (**BCAA**; Ile, Leu, and Val) and Phe in plasma obtained from the

Table 7. Amino acid concentrations (μmol/dL) in plasma from the coccygeal artery in lactating dairy cows fed diets containing 100% corn silage (CS), 50% corn silage and 50% alfalfa hay (CSAH), and 100% alfalfa hay (AH) as forage[1]

	Diet				*P*-value[2]		
AA	CS	CSAH	AH	SE	Trt	L	Q
Essential AA (EAA)							
Arg	11.1	10.4	11.5	0.55	0.32	0.64	0.16
His	5.49	5.49	5.66	0.25	0.80	0.57	0.75
Ile	11.2	13.3	14.8	0.91	<0.01	<0.01	0.60
Leu	21.6	24.7	25.5	1.62	<0.01	<0.01	0.14
Lys	7.14	7.29	7.71	0.54	0.49	0.26	0.74
Met	1.97	1.93	1.89	0.14	0.91	0.66	0.98
Phe	4.77	5.26	5.80	0.27	0.03	<0.01	0.92
Thre	8.70	8.26	8.90	0.47	0.29	0.64	0.14
Trp	3.12	3.78	3.92	0.37	0.28	0.14	0.56
Val	27.9	33.1	36.1	2.24	<0.01	<0.01	0.23
Total EAA	102.2	113.5	122.5	5.92	<0.01	<0.01	0.79
Nonessential AA (NEAA)							
Ala	22.3	21.5	22.1	1.07	0.85	0.85	0.60
Asp	1.70	1.24	1.13	0.18	0.07	0.03	0.41
Asn	2.44	2.53	2.47	0.19	0.94	0.89	0.74
Glu	9.35	7.21	6.55	0.84	<0.01	<0.01	0.04
Glun	23.7	24.5	25.7	1.13	0.28	0.12	0.88
Gly	22.5	18.6	18.7	1.77	0.04	0.03	0.14
Pro	10.4	10.4	9.75	0.46	0.48	0.32	0.51
Ser	8.70	7.78	8.20	0.42	0.03	0.05	0.02
Tyr	5.75	6.13	6.55	0.26	0.03	0.01	0.92
Total NEAA	106.5	99.9	101.6	4.59	0.34	0.30	0.29

[1] Least squares means.

[2] Trt = treatment effect; L = linear contrast; Q = quadratic contrast.

coccygeal artery (Table 7) increased linearly when alfalfa hay was added to the diet. The square containing the primiparous cows responded more to feeding alfalfa compared with multiparous cows, such that primiparous cows fed CSAH had greater concentrations of Leu ($P < 0.02$) and Val ($P < 0.001$) in the plasma compared with those fed CS. This finding was further enhanced by feeding AH compared with CSAH. Among the multiparous cows, this response was not as dramatic, especially in the square containing the cows with the lower milk production, in which no response was observed. The greater concentrations of these EAA were most likely due to both increased microbial synthesis by cows fed alfalfa and changes in the AA profile of the forages. Based on NRC (2001) feed values, concentrations of BCAA and Phe, on a DM basis, were greater in alfalfa hay than corn silage. Past in vitro studies revealed the growth of ruminal bacteria to be 60% more efficient when AA were provided (Argyle and Baldwin, 1989) and 400% more efficient when peptides were provided (Maeng et al., 1976) compared with urea. Past studies have found that when urea was fed as the only source of RDP for producing animals, plasma BCAA and Phe levels declined compared with those fed an RDP source that provided AA and peptides in addition to ammonia (Oltjen and Putnam, 1966; Freitag et al., 1968). Urea was a significant contributor to RDP in CS and may explain why cows fed this diet had lower concentrations of BCAA and Phe in plasma. However, because all diets contained 4% soybean meal and approximately 50% of the DDGS is degraded in the rumen (Firkins et al., 1984), peptides and AA should have been adequate for rumen microbial growth in all diets. Overall, the concentrations of total EAA increased ($P < 0.001$) linearly when alfalfa was fed due to the greater concentrations of BCAA and Phe. Based on the estimates of the NRC (2001), the increase in EAA by feeding alfalfa was not expected because the MP in AH was estimated to be in negative balance compared with the other diets. However, the NRC (2001) does not take the source of RDP into account when calculating microbial synthesis. A linear decrease in concentrations of Asp and Gly and a linear increase in Tyr with the addition of alfalfa were observed. There was also a quadratic response to Glu and Ser with increasing alfalfa. Overall, treatment did not affect concentrations of nonessential AA (**NEAA**) in the coccygeal artery.

The AA concentrations in plasma from the subcutaneous abdominal vein are shown in Table 8. Concentrations of the BCAA and Trp increased linearly when alfalfa was fed. There was also a tendency ($P < 0.07$) for the Met concentration to decrease linearly when feeding alfalfa. There was probably a greater need of this AA for milk protein synthesis, which was validated

Table 8. Amino acid concentrations (μmol/dL) in plasma from the subcutaneous abdominal vein in lactating dairy cows fed diets containing 100% corn silage (CS), 50% corn silage and 50% alfalfa hay (CSAH), and 100% alfalfa hay (AH) as forage[1]

	Diet				*P*-value[2]		
AA	CS	CSAH	AH	SE	Trt	L	Q
Essential AA (EAA)							
Arg	7.75	7.51	7.55	0.61	0.95	0.81	0.84
His	4.29	4.39	4.28	0.18	0.84	0.99	0.56
Ile	7.42	8.98	9.78	0.82	<0.01	<0.01	0.51
Leu	14.3	17.6	17.3	1.46	0.03	0.03	0.10
Lys	3.14	3.22	3.06	0.47	0.92	0.86	0.72
Met	0.98	0.93	0.68	0.11	0.15	0.07	0.47
Phe	2.91	3.25	2.97	0.32	0.21	0.78	0.08
Thre	6.33	5.69	5.99	0.46	0.43	0.50	0.27
Trp	2.00	2.70	2.92	0.17	<0.01	<0.01	0.15
Val	21.8	27.4	29.0	1.94	<0.01	<0.01	0.14
Total EAA	71.0	81.7	83.7	5.07	<0.01	0.01	0.23
Nonessential AA (NEAA)							
Ala	18.5	19.2	18.5	1.05	0.73	0.98	0.43
Asp	1.02	0.95	0.94	0.09	0.69	0.44	0.74
Asn	1.76	1.56	1.64	0.15	0.57	0.54	0.39
Glu	4.53	4.65	8.59	0.49	<0.01	<0.01	<0.01
Gln	17.5	18.8	18.4	0.97	0.50	0.45	0.38
Gly	20.7	17.7	18.1	1.86	0.12	0.11	0.19
Pro	8.77	8.98	7.69	0.54	0.12	0.11	0.18
Ser	5.74	5.43	5.05	0.51	0.44	0.20	0.95
Tyr	4.09	4.35	4.28	0.34	0.69	0.56	0.54
Total NEAA	83.7	81.6	82.3	4.17	0.87	0.74	0.68

[1] Least squares means.

[2] Trt = treatment effect; L = linear contrast; Q = quadratic contrast.

by the tendency for increased yields of milk protein when alfalfa was fed. As observed in the coccygeal arterial plasma, concentrations of total EAA increased linearly ($P < 0.01$) when alfalfa was added to the diet. Among the NEAA, the concentration of Glu responded quadratically ($P < 0.01$) when alfalfa was fed, but differences in total NEAA were not observed.

Uptake of total EAA increased linearly ($P < 0.01$) when cows were fed alfalfa (Table 9), specifically, BCAA, Phe, and Lys. The increasing uptake of these EAA most likely occurred because of the greater supply available in the arterial blood for use by the mammary gland. Lysine only had a numerical increase in arterial plasma with increasing dietary alfalfa. However, this minor increase improved Lys uptake by the mammary gland. Guinard and Rulquin (1994b) duodenally infused increasing amounts of Lys (0, 9, 27, and 63 g/d) into dairy cows producing approximately 22 kg/d of milk and fed a Lys-deficient diet. The maximum uptake of Lys by the mammary gland was 7.01 μmol/dL with 27 g/d of Lys. Guinard et al. (1994) observed a linear increase in AA uptake by the mammary gland when the available AA in the blood was increased via casein infusion. Guinard and Rulquin (1994a) showed that the increased uptake of AA by increasing casein infusion was observed in all EAA but was inconsistent in the NEAA. Most likely, many of the EAA were metabolized, yielding NEAA. The linear increase in milk yield and the tendency for increased yield in milk protein for cows fed alfalfa hay was probably due to more EAA being available to the mammary gland. Feeding alfalfa hay tended to decrease net uptake of Asp and Gly and increase uptake of Tyr linearly. Furthermore, there was a quadratic ($P < 0.02$) response in the uptake of Ala and Glu by the mammary gland. The uptake of NEAA was not affected by diet.

The uptake to output ratios in the BCAA, Lys, and Phe increased ($P < 0.05$) linearly when alfalfa was fed (Table 10). Feeding AH increased the uptake to output ratios of Leu, Lys, and Val compared with that in CS and CSAH. Amino acids were designated as group I (limited uptake) and group II (excessive uptake) by Mepham (1982). The group I AA includes His, Met, Phe, and Trp with the remaining EAA, Arg, Ile, Leu, Lys, Thr, and Val being group II. In this experiment, Met (CSAH and AH) or Thr (CS) were the first-limiting AA for extraction by the mammary gland. Lysine was not one of the first-limiting AA in uptake even though it is often considered one of the first limiting for milk protein synthesis (NRC, 2001). Lysine uptake often exceeds milk output because it undergoes extensive oxidation in the mammary gland, which accompanies an increasing AA supply (Guinard and Rulquin, 1994b). Even though Tyr is not normally considered an EAA, it may

Table 9. Arterial-venous differences (μmol/dL) in AA in plasma from lactating dairy cows fed diets containing 100% corn silage (CS), 50% corn silage and 50% alfalfa hay (CSAH), and 100% alfalfa hay (AH) as forage[1]

	Diet				*P*-value[2]		
AA	CS	CSAH	AH	SE	Trt	L	Q
Essential AA (EAA)							
Arg	3.38	2.90	3.93	0.56	0.42	0.49	0.27
His	1.18	1.10	1.37	0.17	0.21	0.22	0.19
Ile	3.82	4.37	5.03	0.49	0.09	0.03	0.89
Leu	6.97	7.06	8.50	0.85	0.05	0.03	0.22
Lys	3.94	4.07	4.61	0.41	0.02	0.01	0.30
Met	1.09	1.00	1.30	0.14	0.28	0.27	0.25
Phe	1.97	2.01	2.82	0.27	0.06	0.04	0.24
Thre	2.41	2.57	2.96	0.33	0.41	0.20	0.74
Trp	1.03	1.08	0.79	0.29	0.75	0.56	0.62
Val	5.65	5.63	7.58	0.90	0.03	0.02	0.14
Total EAA	31.1	31.8	38.7	3.45	0.02	<0.01	0.17
Nonessential AA (NEAA)							
Ala	3.58	2.37	3.41	0.65	0.05	0.68	0.02
Asp	0.67	0.29	0.19	0.18	0.14	0.06	0.50
Asn	0.67	0.96	0.82	0.20	0.46	0.54	0.28
Glu	4.87	2.56	–2.02	0.58	<0.01	<0.01	<0.01
Gln	6.11	5.67	7.22	1.02	0.27	0.27	0.24
Gly	1.83	0.89	0.54	0.55	0.20	0.08	0.63
Pro	1.64	1.44	2.07	0.38	0.48	0.43	0.36
Ser	2.68	2.35	2.81	0.50	0.70	0.82	0.42
Tyr	1.65	1.78	2.27	0.31	0.19	0.09	0.54
Total NEAA	23.0	18.3	19.3	3.55	0.47	0.37	0.41

[1] Least squares means.

[2] Trt = treatment effect; L = linear contrast; Q = quadratic contrast.

be limiting for milk protein synthesis if it is not provided through dietary sources and inadequate Phe is supplied in the diet because Tyr is synthesized from Phe via Phe hydroxylase (Cant et al., 1993). In this experiment, there tended to be a linear increase ($P < 0.08$) in the uptake to output ratio of Tyr as alfalfa replaced corn silage. This was likely related to the increasing concentrations of Phe in diets containing alfalfa.

Transfer (Table 11) and extraction (Table 12) efficiencies are methods for evaluating the limitations of the available EAA in the arterial blood. Transfer efficiency evaluates the percentage of an individual EAA

Table 10. Uptake to output ratios[1] of essential AA (EAA) of lactating dairy cows fed diets containing 100% corn silage (CS), 50% corn silage and 50% alfalfa hay (CSAH), and 100% alfalfa hay (AH) as forage[2]

	Diet				*P*-value[3]		
EAA	CS	CSAH	AH	SE	Trt	L	Q
Arg	3.45 (10)[4]	2.89 (9)	3.84 (10)	0.59	0.51	0.64	0.29
His	1.36 (4)	1.27 (2)	1.58 (3)	0.20	0.20	0.23	0.17
Ile	1.71 (6)	1.95 (7)	2.24 (6)	0.23	0.08	0.03	0.89
Leu	1.90 (7)	1.93 (6)	2.31 (8)	0.25	0.05	0.03	0.24
Lys	1.45 (5)	1.48 (5)	1.69 (4)	0.16	0.05	0.03	0.28
Met	1.26 (2)	1.16 (1)	1.47 (1)	0.15	0.33	0.32	0.27
Phe	1.34 (3)	1.36 (4)	1.89 (5)	0.18	0.07	0.05	0.25
Thr	1.24 (1)	1.34 (3)	1.52 (2)	0.17	0.42	0.20	0.81
Trp	3.07 (9)	3.36 (10)	2.27 (7)	0.91	0.68	0.54	0.53
Val	2.16 (8)	2.14 (8)	2.90 (9)	0.37	0.04	0.03	0.15
Tyr[5]	1.17 [1]	1.26 [2]	1.61 [4]	0.22	0.18	0.08	0.55

[1] Uptake to output ratio = [arteriovenous difference of AA (g/L) × mammary blood flow (L/d) (Kronfeld et al., 1968)]/AA output in milk (g/d; Jacobson et al., 1970).

[2] Least squares means.

[3] Trt = treatment effect; L = linear contrast; Q = quadratic contrast.

[4] Numbers in parentheses indicate the apparent order of limiting AA.

[5] Numbers in brackets are ranking of tyrosine if it were considered an essential AA.

Table 11. Transfer efficiencies[1] (%) of essential AA (EAA) from blood to milk of lactating dairy cows fed diets containing 100% corn silage (CS), 50% corn silage and 50% alfalfa hay (CSAH), and 100% alfalfa hay (AH) as forage[2]

	Diet				*P*-value[3]		
EAA	CS	CSAH	AH	SE	Trt	L	Q
Arg	9.63 (10)[4]	10.2 (9)	9.02 (9)	0.57	0.13	0.28	0.09
His	16.7 (7)	16.1 (6)	16.2 (5)	0.86	0.76	0.56	0.66
Ile	22.6 (4)	18.3 (5)	15.7 (6)	1.77	<0.01	<0.01	0.50
Leu	19.5 (6)	16.0 (7)	14.9 (7)	1.57	0.02	<0.01	0.37
Lys	44.8 (2)	41.3 (2)	39.1 (2)	3.75	0.22	0.09	0.80
Met	47.6 (1)	47.2 (1)	49.8 (1)	3.93	0.88	0.70	0.74
Phe	33.5 (3)	29.0 (3)	26.7 (3)	2.21	0.04	0.01	0.59
Thr	22.5 (5)	24.0 (4)	23.0 (4)	1.42	0.50	0.72	0.27
Trp	12.3 (8)	11.0 (8)	9.55 (8)	0.88	0.09	0.03	0.95
Val	11.0 (9)	8.61 (10)	7.47 (10)	0.88	<0.01	<0.01	0.78
Tyr[5]	24.6 [4]	23.8 [5]	23.8 [4]	1.40	0.82	0.58	0.35

[1] Transfer efficiency = AA output in milk (g/d; Jacobson et al., 1970) × 100/[arterial AA concentration (g/L) × mammary blood flow (L/d; Kronfeld et al., 1968).

[2] Least squares means.

[3] Trt = treatment effect; L = linear contrast; Q = quadratic contrast.

[4] Numbers in parentheses indicate the apparent order of limiting AA.

[5] Numbers in brackets are ranking of tyrosine if it were considered an essential AA.

from the arterial plasma that is used to synthesize milk protein, whereas extraction efficiency evaluates the percentage of plasma EAA extracted by the mammary gland to be used for all needs such as protein synthesis and catabolism. Extraction efficiency may be the ideal method for evaluating AA requirements because it takes the entire EAA needs of the mammary gland into account instead of just the milk AA requirements, especially because many EAA such as Lys and BCAA are extracted in excess of the requirement for AA in milk protein (Mepham, 1982).

Feeding alfalfa linearly decreased ($P < 0.03$) the transfer efficiencies of the BCAA, Phe, and Trp (Table 11). There was also a tendency ($P < 0.09$) for a linear decrease in Lys transfer efficiency when alfalfa hay was fed and a tendency ($P < 0.09$) for a quadratic response in Arg with increasing alfalfa hay. Cows fed alfalfa hay had numerically greater concentrations of Arg in their arterial supply but similar amounts of Arg were estimated to be used for milk protein synthesis, thereby decreasing the transfer efficiency of Arg in AH. Methionine was the first-limiting EAA for milk protein synthe-

Table 12. Extraction efficiencies[1] (%) of essential AA (EAA) from blood to mammary gland of lactating dairy cows fed diets containing 100% corn silage (CS), 50% corn silage and 50% alfalfa hay (CSAH), and 100% alfalfa hay (AH) as forage[2]

	Diet				*P*-value[3]		
EAA	CS	CSAH	AH	SE	Trt	L	Q
Arg	30.2 (7)[4]	28.0 (7)	34.4 (4)	3.83	0.48	0.44	0.35
His	21.8 (9)	20.0 (9)	23.6 (8)	2.47	0.22	0.37	0.13
Ile	34.2 (5)	34.2 (4)	33.4 (5)	3.38	0.97	0.83	0.89
Leu	34.2 (5)	29.7 (6)	32.4 (7)	2.91	0.17	0.46	0.08
Lys	61.1 (1)	59.4 (1)	62.6 (2)	3.52	0.13	0.39	0.08
Met	46.7 (2)	51.4 (2)	64.4 (1)	6.60	0.16	0.07	0.60
Phe	40.3 (3)	38.7 (3)	48.5 (3)	5.10	0.06	0.07	0.12
Thr	27.9 (8)	30.5 (5)	33.3 (6)	3.45	0.45	0.21	0.98
Trp	36.3 (4)	24.7 (8)	22.9 (9)	4.36	0.07	0.03	0.35
Val	21.0 (10)	17.0 (10)	20.4 (10)	2.33	0.19	0.80	0.07
Tyr[5]	28.4 [8]	31.1 [5]	35.9 [4]	4.75	0.36	0.16	0.80

[1] Extraction efficiency = [arteriovenous difference of AA (g/L) × 100/concentration (g/L) of AA in plasma of coccygeal artery.

[2] Least squares means.

[3] Trt = treatment effect; L = linear contrast; Q = quadratic contrast.

[4] Numbers in parentheses indicate the apparent order of limiting AA.

[5] Numbers in brackets are ranking of tyrosine if it were considered an essential AA.

Table 13. Ruminal VFA and ammonia from lactating dairy cows fed diets containing 100% corn silage (CS), 50% corn silage and 50% alfalfa hay (CSAH), and 100% alfalfa hay (AH) as forage[1]

	Diet				*P*-value[2]		
Measurement	CS	CSAH	AH	SE	Trt	L	Q
VFA, %							
Acetate (A)	63.5	64.3	65.0	0.74	0.05	0.02	0.87
Propionate (P)	21.2	21.1	20.6	0.79	0.78	0.52	0.79
Isobutyrate	1.00	0.93	0.95	0.05	0.39	0.36	0.31
Butyrate	11.3	10.9	10.7	0.3	0.22	0.09	0.72
Isovalerate	1.74	1.37	1.24	0.07	<0.01	<0.01	0.22
Valerate	1.34	1.40	1.62	0.04	<0.01	<.01	0.08
Total, m*M*/L	52.1	54.4	45.6	7.2	0.52	0.42	0.42
A:P	3.11	3.13	3.18	0.11	0.29	0.14	0.70
Ammonia, mg/dL	5.59	5.31	4.46	1.02	0.64	0.37	0.78

[1] Least squares means.

[2] Trt = treatment effect; L = linear contrast; Q = quadratic contrast.

sis for all diets even though CS contained mostly corn-based feedstuffs. There tended to be a linear increase ($P < 0.07$) in the efficiency of extracting (Table 12) Phe and Met when alfalfa was added to the diet. Furthermore, the extraction efficiency of Trp decreased ($P < 0.03$) linearly by feeding alfalfa. There was also a tendency for a quadratic response for the extraction efficiency of the BCAA when increasing alfalfa, such that cows fed CSAH had a lower extraction efficiency of these AA compared with CS or AH. This may also relate to the fact that cows fed CSAH had a poorer utilization of N as previously discussed. Overall, Lys was the first-limiting AA being extracted from the blood in CS and CSAH, and Met was first limiting in AH. In a diet containing 20% DDGS, Nichols et al. (1998) found Met to be first limiting based on transfer efficiency but Lys was first limiting based on extraction efficiency. In contrast, Liu et al. (2000) fed a similar diet and found Lys to be first limiting using both procedures. Regardless of using extraction efficiency or transfer efficiency for determining the first-limiting AA, Lys, Met, and Phe were the first 3 limiting AA, which has been shown in other studies feeding DDGS (Nichols et al., 1998; Liu et al., 2000).

As calculated using NRC (2001), estimated intestinally available Lys (5.61, 5.73, and 6.06% of MP for CS, CSAH, and AH, respectively) and the ratio of Lys to Met (2.89, 3.01, 3.14 to 1 for CS, CSAH, and AH, respectively) increased as alfalfa was added to the diet. The estimated intestinally available Met (1.93, 1.92, 1.93% of MP for CS, CSAH, and AH, respectively) did not differ among diets. Based on dose response methods, the NRC (2001) summarized that a dairy cow requires 7.2% Lys and 2.4% Met in the MP to maximize milk protein yield and content. Similarly, Rulquin et al. (1993) summarized that milk protein content and yield was maximized when Lys and Met in the MP were 7.3 and 2.5%, respectively. This high percentage of Lys may be difficult to achieve in a commercial dairy diet; therefore, a more practical goal is to supply 6.6 and 2.2% Lys and Met, respectively, to the small intestine (Schwab and Ordway, 2004). Schwab and Ordway (2004) recommended that the ideal ratio of Lys to Met is 3:1, which reflects the proportion in which these AA are incorporated into milk protein (Jacobson et al., 1970). When the ratio is below 3:1, Met is supplied in excess of need; above 3:1, Lys is supplied in excess of need. Based on the extraction efficiencies in this experiment, this ratio accurately predicted which EAA would be first limiting for milk protein synthesis.

Ruminal Components

The proportion of ruminal acetate increased ($P < 0.04$) linearly without affecting the proportion of propionate when alfalfa was added into the diets (Table 13). Dhiman and Satter (1997) observed a tendency for an increased proportion of acetate when cows were fed forage containing solely alfalfa silage compared with corn silage. Similarly, Broderick (1985) fed either 60% alfalfa hay or 60% corn silage and found that alfalfa hay increased ruminal proportions of acetate. There was also a tendency ($P < 0.09$) for the concentrations of butyrate to decrease linearly with increasing concentrations of alfalfa. The ruminal concentrations of isovalerate decreased ($P < 0.01$) and valerate increased ($P < 0.01$) linearly as alfalfa was added to the diets. Isovalerate may have been converted to Leu in microbial protein, thus possibly indicating greater microbial growth and corresponding with the greater Leu concentrations in the plasma of cows fed CSAH and AH compared with CS. Total ruminal VFA concentrations and the ratio of acetate to propionate were similar among the diets. The concentrations of ruminal ammonia were similar among treatments even though dietary CP increased with increasing concentrations of alfalfa in the diet. Urea, a highly degradable CP source, was used in diets containing corn silage (Table 1), which may have kept dietary RDP about the same for all diets and thus kept ruminal ammonia concentrations relatively similar.

CONCLUSIONS

Feeding alfalfa hay instead of corn silage as the forage source in diets containing 15% DDGS to cows in late lactation linearly increased milk yield without significantly affecting protein content or yield. Feeding alfalfa hay as the only forage source improved feed efficiency due to an increase in milk yield but no effect on feed intake compared with those fed only corn silage. Based on the extraction of AA from the plasma, Lys was the first-limiting EAA when cows were fed solely corn silage as forage or both corn silage and alfalfa hay; Met was first limiting when cows were fed alfalfa hay. Overall, feeding alfalfa hay increased uptake of EAA from the blood, which may have resulted in the increased milk yield.

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