

Performance of Lactating Dairy Cows Fed Red Clover or Alfalfa Silage

P. C. HOFFMAN,^{1,2} D. K. COMBS, N. M. BREHM,¹
and D. A. WELCH¹

Department of Dairy Science,
University of Wisconsin, Madison 53706

ABSTRACT

The objective of this study was to compare utilization of red clover (*Trifolium pratense* L.) and alfalfa (*Medicago sativa* L.) by lactating dairy cows. Red clover and alfalfa were harvested and conserved as silage at two maturities in 2 consecutive yr. Each year, diets containing experimental forages and supplemental grain were fed to 16 multiparous Holstein cows in early lactation in a replicated 4 × 4 Latin square lactation trial. Lactation performance and nutrient intake responses caused by forage type (red clover vs. alfalfa), maturity (early vs. late), and equivalent acid detergent fiber (ADF) content (yr 1, late alfalfa vs. late red clover; yr 2, early alfalfa vs. late red clover) were compared. Milk yield was not different between cows fed red clover or alfalfa in yr 1, but milk yield was higher for cows fed red clover in yr 2. When cows were fed alfalfa and red clover with similar ADF content, the milk yield of cows fed red clover was lower than that of cows fed alfalfa in yr 1, and milk yields were similar between cows fed alfalfa and red clover in yr 2. Milk protein yield and percentage were lower for cows fed red clover in yr 1 and 2, respectively. Intake of ADF and neutral detergent fiber was lower for cows fed red clover in both years. When red clover contained the same ADF content as did alfalfa, cows fed red clover ate less ADF, neutral detergent fiber, and dry matter, resulting in lower milk yield potential.

(**Key words:** red clover, alfalfa, lactation)

Abbreviation key: **AE** = early alfalfa, **AL** = late alfalfa, **NIRS** = near infrared reflectance spectroscopy, **RE** = early red clover, **RL** = late red clover.

INTRODUCTION

Red clover is grown widely as forage for dairy cattle in regions with poorly drained soils that are not

suited for alfalfa production. Investigations regarding the utilization of red clover by lactating dairy cows are limited, and it is commonly assumed that red clover and alfalfa have similar nutritional characteristics (24).

Recent research, however, suggested that red clover has unique nutritional characteristics. In a recent study, Hoffman et al. (8) observed markedly lower ruminal degradation of NDF of red clover compared with that of alfalfa. Ruminal NDF degradation was approximately 50% lower for red clover than for alfalfa at any of the three maturities. Ruminal degradation of red clover NDF was lower than that of alfalfa NDF because red clover contained more undegradable NDF than did alfalfa. Differences in undegradable NDF fractions between red clover and alfalfa could not be explained by lignification because red clover fiber was less lignified than was alfalfa. Buxton (4), however, did not observe appreciable differences in in vitro NDF degradation between the stem tissues of alfalfa and red clover.

Red clover contains less soluble and more slowly degradable CP than does alfalfa (17). Albrecht and Muck (1) reported less extensive proteolysis in red clover silage than in alfalfa silage. Jones et al. (10) reported that the reduced proteolysis in red clover silage was due to polyphenol oxidases in red clover tissue that inhibit protein hydrolysis. Those data (1, 10, 17) suggest that protein fractions of red clover silage contain more undegradable CP than do protein fractions of alfalfa. Whether red clover protein fractions are efficacious for high yielding dairy cows has not been determined.

The purpose of this study was to reexamine previously observed differences in NDF degradation between red clover and alfalfa and to determine whether potential differences alter the utilization of red clover by lactating dairy cows.

MATERIALS AND METHODS

Forage Harvest

One-half of early spring growth herbage from two 5-ha fields containing alfalfa (*Medicago sativa* L.) or

Received April 18, 1996.

Accepted April 25, 1997.

¹Marshfield Agricultural Research Station.

²Reprint requests.

red clover (*Trifolium pratense* L.) was cut and swathed on May 30, 1992. Forages were wilted for approximately 36 h, harvested as low moisture silage, and conserved in 2.5- × 46-m plastic silage bags. Herbage remaining in each field was cut, harvested, and conserved by identical procedures 15 d later. Forage harvesting and conservation protocols were repeated the next year (1993) on the same two 5-ha fields. Because of inclement weather, harvest dates were later for the 2nd yr: June 14 and 29 for early and late cut forages, respectively.

Four experimental forages, early alfalfa (**AE**), late alfalfa (**AL**), early red clover (**RE**), and late red clover (**RL**), were harvested and conserved each year. The phenological stage of RE and RL at harvest was 3 and 5, respectively, for yr 1 and 6 and 7, respectively, for yr 2 as characterized by Ohlsson and Wedin (18). Phenological stages of AE and AL at harvest were 4 and 5, respectively, for yr 1 and 6 and 7, respectively, for yr 2 as characterized by Kalu and Fick (11).

All loads of experimental forage from each species, cutting date, and field were sampled prior to ensiling, and CP, ADF, and NDF were estimated using near infrared reflectance spectroscopy (**NIRS**) (NIRS Systems, Inc., Silver Spring, MD). The forage analysis by NIRS was used to formulate experimental diets.

Lactation Trials

Forages (AE, AL, RE, RL) from each year were evaluated in one of two lactation trials. The design of lactation trials was identical except different cows were used for each trial. Each lactation trial commenced in November of the year that the forages were harvested. Sixteen multiparous Holstein cows in early lactation were blocked by DIM and assigned to replicated 4 × 4 Latin squares. Mean DIM for cows upon assignment to squares 1, 2, 3, and 4 were 28, 42, 68, and 101 d for yr 1 and 37, 51, 89, and 104 d for yr 2, respectively. Experimental periods were 21 d. The first 14 d served as the adaptation period, and all data collection occurred during d 15 to 21. Cows were randomly assigned to treatment diets within squares.

Treatments consisted of AE, AL, RE, or RL supplemented with a grain mix. Two grain mixes were used in yr 1 (Table 1) because preliminary NIRS analyses of experimental forages indicated large differences in CP content between early and late harvested forages. Two grain mixes were utilized to create isonitrogenous treatment diets in yr 1. Pretrial NIRS data in yr 2 suggested no appreciable differences in the CP content of the experimental forages; one grain mix was utilized for all diets.

TABLE 1. Ingredient and nutrient composition of grain mixes.

| Item | yr 1 | | yr 2 |
|---|-----------|-------|-------|
| | Early | Late | |
| | (% of DM) | | |
| Ingredient | | | |
| Shelled corn | 79.68 | 60.05 | 61.24 |
| Expeller soybean oil meal | 8.41 | ... | ... |
| Soybean oil meal | ... | 27.71 | 34.65 |
| Meat and bone meal | 8.41 | 8.31 | ... |
| Mineral and vitamin premix ¹ | 2.80 | 3.23 | 3.31 |
| Trace-mineralized salt | 0.70 | 0.70 | 0.80 |
| Composition | | | |
| DM, % as fed | 89.4 | 89.4 | 86.4 |
| CP | 16.1 | 22.5 | 23.2 |
| ADF | 4.5 | 5.6 | 5.6 |
| NDF | 18.3 | 17.8 | 13.5 |
| Ca | 1.73 | 1.72 | 0.82 |
| P | 0.82 | 0.87 | 0.76 |

¹Contained 16% Ca, 12.5% P, 20.0% NaCl, 551,150 IU/kg of vitamin A, 216,050 IU/kg of vitamin D, and 661 IU/kg of vitamin E.

Diets were formulated to contain similar forage to grain ratios. Forage to grain ratios (DM basis) for yr 1 and 2 were 58:42 and 50:50, respectively. Different forage to grain ratios were used for yr 1 and 2 in an effort to standardize the relative concentrations of NDF in the diets between years. Treatment diets were deficient in the energy content required for dairy cows in early lactation. Energy densities were purposely low to test the milk yield potential of experimental forages.

Treatment diets were mixed and fed as TMR at 0800 h daily. Amount of TMR offered was recorded, and treatment diets were sampled daily during d 15 to 21 of each period. Weighing, recording, and sampling of orts followed the same procedures as did treatment diets. Forages were sampled three times per week throughout the experiments. Forage samples were split, and an undried subsample was frozen for later VFA and pH determination.

The remaining forage subsample was immediately analyzed for DM by oven-drying for 48 h at 55°C. Dry matter contents of the treatment diets and orts were determined by identical methods. Dried forage and samples of the treatment diets were ground through a Wiley mill (1-mm screen; Arthur H. Thomas, Philadelphia, PA) and saved for chemical analysis. All experimental forage and samples of the treatment diets were analyzed for CP and absolute DM (3). Acid detergent fiber and NDF were determined according to the procedures of Robertson and Van Soest (21), utilizing the modifications of Mertens (13).

Silage pH was determined, and extract for determination of organic acid was prepared according to the

procedures of Hoffman et al. (9). Final determination of organic acid was conducted by ion exclusion chromatography (6) using a Dionex QIC ion chromatograph (Dionex, Sunnyvale, CA). Calcium and P were determined by atomic absorption spectroscopy and colorimetric methods, respectively (Coleman Instruments, Inc., Maywood, IL).

In vitro NDF digestion kinetics and in vitro DM digestibility of the silages were evaluated according to the procedures of Goering and Van Soest (7). In vitro analyses were conducted on a composite of the weekly forage samples for each period. Samples were incubated for 0, 4, 8, 12, 24, 48, and 72 h. Ruminal fluid was collected from a dry cow fitted with a ruminal cannula and fed alfalfa grass silage.

Neutral detergent fiber kinetics were analyzed using the nonlinear regression procedures of SAS (22) and were fitted to the model of Mertens and Loften (14). Ruminal availability of NDF was estimated using the equation: slowly digested fraction (B)[$k_d / (k_d + k_p)$], where k_p = ruminal passage rate of 0.06/h, and k_d = fractional degradation rate.

Cows were housed in a free-stall barn equipped with Calan gates (American Calan, Inc., Northwood, NH) and were milked twice daily at 0230 and 1430 h. Milk weights were recorded daily, and milk was sampled twice daily on d 16, 18, and 20 of each period. Milk fat and protein were determined on individual milk samples by automated techniques (Central Wisconsin DHIA, Colby, WI).

Statistics

Data were analyzed using the general linear models procedures of SAS (22). Preliminary statistical analysis for the combined data from yr 1 and 2 indicated that year effects were significant for nearly all variables measured. Because of significant year effects, data for each year were analyzed separately according to the following models.

For the lactation trial,

$$Y = \mu + S_i + C_j(S_i) + P_k + T_m + (S \times T)_{im} + E_{ijkm}$$

where

$$\begin{aligned} \mu &= \text{overall mean of the population,} \\ S_i &= \text{average effect of square } i, \\ C_j(S_i) &= \text{average effect of cow } j \text{ nested within square } i, \\ P_k &= \text{average effect of period } k, \\ T_m &= \text{average effect of treatment } m, \\ (S \times T)_{im} &= \text{average effect of square } i \text{ and treatment } m, \text{ and} \end{aligned}$$

E_{ijkm} = unexplained residual error, assumed to be normally and independently distributed.

For the in vitro analyses,

$$Y = \mu + P_i + T_j + E_{ij}$$

where

$$\begin{aligned} \mu &= \text{overall mean of the population,} \\ P_i &= \text{average effect of period } i, \\ T_j &= \text{average effect of treatment } j, \text{ and} \\ E_{ij} &= \text{unexplained residual error, assumed to be normally and independently distributed.} \end{aligned}$$

The average effect of square was tested using cow (square) as the error term. All other terms were tested using the residual mean square. Preplanned contrasts were made between forage (alfalfa vs. red clover), maturity (early vs. late), and equivalent ADF content (yr 1, AL vs. RL and yr 2, AE vs. RL).

RESULTS AND DISCUSSION

Forage Quality

The nutrient composition of silages is presented in Table 2. In yr 1, RE was higher in CP and lower in ADF and NDF than was AE. Observations were consistent with those of Smith (23), who observed that spring growth red clover matured slower than did alfalfa. As a consequence, when red clover and alfalfa are harvested on the same calendar day, as in this study, red clover will be at an earlier physiological maturity and will have a lower fiber content (23).

Concentrations of CP, ADF, and NDF in RL and AL in yr 1 were similar. In yr 2, red clover contained less NDF than did alfalfa (RE < AE; RL < AL), which was consistent with previously mentioned concepts. Crude protein contents of all four silages were similar in yr 2. The NDF content of RL was higher than that of RE, but the NDF content of AL was not higher than that of AE. The lack of a decline in CP in forages as maturity advanced, coupled with a lack of fiber accretion in alfalfa as maturity advanced, indicated that classic effects of forage maturity were not observed in yr 2. We think that these inconsistencies in yr 2 were the result of lodging in late harvested forages. When forage is lodged, the upper portions of the plant, which contain a greater percentage of leaves, must be harvested; these portions have a higher quality than stem tissue (2).

Fermentation characteristics of experimental forages are also presented in Table 2. Forages were

TABLE 2. Nutrient and fermentation characteristics of experimental silages.¹

| Item | Alfalfa | | Red clover | | SD |
|--------------|-------------------------------|------|------------|------|------|
| | Early | Late | Early | Late | |
| | ————— yr 1 ² ————— | | | | |
| Nutrient | | | | | |
| DM, % as fed | 27.2 | 47.8 | 30.8 | 49.3 | 10.4 |
| CP | 18.8 | 18.5 | 22.5 | 17.6 | 2.1 |
| ADF | 30.8 | 31.9 | 24.9 | 32.5 | 4.3 |
| NDF | 39.1 | 41.8 | 31.7 | 43.3 | 5.6 |
| Ca | 1.16 | 1.29 | 1.27 | 1.15 | 0.14 |
| P | 0.33 | 0.34 | 0.31 | 0.31 | 0.07 |
| Fermentation | | | | | |
| pH | 4.7 | 4.7 | 4.6 | 4.9 | 0.17 |
| Lactic acid | 9.56 | 7.44 | 14.25 | 5.43 | 4.10 |
| Acetic acid | 5.39 | 1.44 | 3.75 | 1.00 | 2.61 |
| Butyric acid | 0.11 | 0.01 | 0.01 | 0.01 | 0.09 |
| | ————— yr 2 ³ ————— | | | | |
| Nutrient | | | | | |
| DM, % as fed | 55.8 | 36.8 | 53.5 | 36.8 | 9.4 |
| CP | 14.4 | 15.7 | 14.9 | 16.4 | 1.6 |
| ADF | 37.7 | 41.2 | 33.1 | 35.5 | 3.6 |
| NDF | 49.8 | 48.4 | 41.7 | 44.7 | 5.7 |
| Ca | 0.85 | 1.02 | 0.94 | 0.98 | 0.10 |
| P | 0.25 | 0.31 | 0.25 | 0.26 | 0.03 |
| Fermentation | | | | | |
| pH | 4.7 | 4.44 | 4.7 | 4.3 | 0.22 |
| Lactic acid | 4.01 | 8.12 | 3.73 | 7.56 | 2.11 |
| Acetic acid | 1.16 | 2.53 | 1.23 | 2.22 | 0.68 |
| Butyric acid | ... | ... | ... | ... | ... |

¹All values are percentages of DM unless otherwise specified.

²Early and late forages were harvested on May 30 and June 14, respectively.

³Early and late forages were harvested on June 14 and 29, respectively.

all well fermented, visually free of mold, and of normal appearance. The pH of silages in yr 1 and 2 were within normal ranges (12). Lactic and acetic acid contents of RE and AE in yr 1 were uncharacteristically high. High organic acid production was possible because forages were harvested at an early maturity when carbohydrate content (fermentation substrate) was high (5).

NDF Digestion

In vitro digestion characteristics of the silages are presented in Table 3. In yr 1 and 2, late harvested forages (RL and AL) contained less ($P < 0.05$) slowly digested NDF and more ($P < 0.05$) undigested NDF than did early harvested forage. Digestion rate of late harvested forages was higher ($P < 0.05$) in yr 1 but not in yr 2. There was no difference in lag time of NDF digestion between forages harvested early or late in either year.

We observed increased undigestible NDF and decreased slowly digestible NDF fractions as maturity advanced. Data from the present study were consistent with previous findings (5). In the present study, we observed faster NDF digestion rates between early and late harvested forages in yr 1 but not in yr 2.

Data from yr 1 were in contrast to those of Cherney et al. (5), who observed decreasing NDF digestion rates as maturity advanced. Cherney et al. (5), however, evaluated NDF digestion kinetics of grasses and not legumes. In a previous study, Hoffman et al. (8) also observed decreased NDF digestion rates in grasses as maturity advanced, but results for alfalfa, red clover, and trefoil were variable, and no clear relationship between NDF digestion rate and maturity of legumes was established. In yr 2, no difference in NDF digestion rates were observed, perhaps because of the narrow range in nutrient composition between experimental forages.

In the present study, we did not observe a decrease in potentially digestible NDF as maturity advanced. However, it is unlikely that maturity effects on potential digestible NDF existed in this study for the following reasons. First, maturity effects in yr 2 might have been altered by the harvesting of lodged forages. Second, in a previous study, Hoffman et al. (8) did not observe a decrease in potentially digestible NDF in red clover as maturity advanced. Finally, NDF contents of AE and AL in yr 1 and 2 were similar, and in vitro NDF digestion assays may not be sensitive enough to elicit differences.

The effect of forage type on NDF digestion characteristics is also presented in Table 3. In yr 1, red clover contained less ($P < 0.01$) undigested NDF and more ($P < 0.01$) slowly digested NDF than did alfalfa. Digestion rate of NDF was slower ($P < 0.05$) for red clover than for alfalfa. No differences were observed in NDF digestion lag time, and potentially digestible NDF was higher ($P < 0.01$) for red clover than for alfalfa. In yr 2, there were no differences in NDF digestion characteristics between red clover and alfalfa.

Contrasts were conducted within each year on red clover and alfalfa, which were most similar in ADF content. The ADF contrast was conducted to compare red clover and alfalfa when forage quality was similar. Acid detergent fiber was chosen as the measure of equivalent forage quality because NE_L content of forage is most often calculated from ADF (26). In yr 1, differences in in vitro NDF digestion between AL and RL were statistically contrasted. The AL and RL contained 31.9 and 32.5% ADF, respectively. Red clover (RL) contained more ($P < 0.01$) slowly digested NDF and less ($P < 0.01$) undigested NDF

than did AL. Digestion rate of RL was slower ($P < 0.05$) than that of AL. Potentially digestible NDF for RL and AL were similar. In yr 2, contrasts were made between AE and RL at 37.7 and 35.5% ADF, respectively. No differences were observed in NDF digestion characteristics between these two forages.

Data from this study did not support the pretrial hypothesis that red clover has inferior NDF digestion characteristics compared with alfalfa. The present study supported the findings of Buxton (4), who observed no appreciable difference in the in vitro digestibility of stem tissue of alfalfa or red clover. Reasons for low NDF digestibility in red clover in the previous study are unclear. The previous study evaluated NDF digestibility of nonensiled perennial legume and grass species using in situ methods. In vitro techniques were used on ensiled forages in the present study for ease of replication and under the premise that in vitro techniques should rank NDF digestibilities similar to in situ methods (26). We are cognizant of the idiosyncracies of each method (16, 26), yet are unable to

construct a valid argument regarding possible procedural errors.

In vitro DM digestibility is also presented in Table 3. In yr 1, in vitro DM digestibility was higher ($P < 0.01$) for red clover than for alfalfa, lower ($P < 0.05$) for late harvested forages, and higher ($P < 0.01$) for RL than for AL (equal ADF). In yr 2, no differences were observed in in vitro DM digestibility between forage type (red clover vs. alfalfa) or forages equal in ADF (AE vs. RL). Late harvested forages had lower ($P < 0.01$) in vitro DM digestibility than did early harvested forages.

Lactation Data

Ingredient and nutrient compositions of experimental diets are presented in Table 4. Diets were formulated at fixed forage to concentrate ratios of 58:42 in yr 1 and 50:50 in yr 2. The diet with AE contained the lowest concentration of CP (18.33% DM), which was adequate to support a 590-kg cow yielding 53 kg of 4% FCM (15). Experimental diets were not isocaloric and were purposely formulated at low energy densi-

TABLE 3. In vitro digestion characteristics of experimental silages.¹

| Item | Alfalfa | | Red clover | | SE | P ² | | |
|----------------------------------|---------|------|------------|------|------|-----------------|----------|-----|
| | Early | Late | Early | Late | | Forage | Maturity | ADF |
| yr 1 | | | | | | | | |
| NDF Digestion | | | | | | | | |
| B, ³ % of NDF | 55.7 | 47.2 | 65.3 | 59.2 | 2.50 | ** | * | ** |
| C, ⁴ % of NDF | 44.3 | 52.8 | 34.7 | 40.8 | 2.50 | ** | * | ** |
| k _d , ⁵ /h | 0.08 | 0.14 | 0.08 | 0.10 | 0.01 | * | * | * |
| Lag, ⁶ h | 2.0 | 2.1 | 3.8 | 2.7 | 0.71 | NS ⁷ | NS | NS |
| PD NDF, ⁸ % | 31.7 | 32.8 | 36.0 | 36.7 | 2.60 | * | NS | NS |
| IVDMD ⁹ | 78.2 | 77.7 | 87.6 | 83.6 | 1.00 | ** | * | ** |
| yr 2 | | | | | | | | |
| NDF Digestion | | | | | | | | |
| B, % of NDF | 56.5 | 47.3 | 54.1 | 53.5 | 2.46 | NS | * | NS |
| C, % of NDF | 43.5 | 54.7 | 45.9 | 46.5 | 2.46 | NS | * | NS |
| k _d , /h | 0.06 | 0.08 | 0.08 | 0.07 | 0.01 | NS | NS | NS |
| Lag, h | 0.4 | 1.6 | 0.7 | 1.9 | 0.80 | NS | NS | NS |
| PD NDF, % | 28.2 | 25.9 | 29.9 | 28.9 | 1.75 | NS | NS | NS |
| IVDMD | 77.6 | 74.8 | 82.8 | 75.5 | 1.40 | NS | ** | NS |

¹All values are percentages of DM unless otherwise specified.

²ADF contrast: yr 1, late alfalfa versus late red clover; yr 2, early alfalfa versus late red clover.

³Slowly digested NDF fraction.

⁴Undigested NDF fraction.

⁵Fractional degradation rate.

⁶NDF digestion lag time.

⁷ $P > 0.05$.

⁸PD = Potentially digestible. Calculated as $B[k_d/(k_d + k_p)]$ where B and k_d are as defined previously and k_p = ruminal passage rate of 0.06/h.

⁹In vitro DM digestibility (48 h).

* $P < 0.05$.

** $P < 0.01$.

ties to allow expression of milk yield potential of experimental forages.

In yr 2, CP content of experimental diets was lower than expected. Pretrial CP content of experimental forages was overestimated by NIRS, resulting in diets that were lower in CP than were originally planned. The diet with RL was lowest in CP at 16.46%. In yr 2, diets containing AE, AL, RE, and RL were estimated (15) to contain between 1.58 and 1.66 Mcal/kg of NE_L. Although the CP content of diets in yr 2 was lower than planned, energy concentration was still the limiting nutrient (15) in all diets.

The effect of experimental forages on lactation performance and DMI is presented in Table 5. There were no significant interactions of square and treatment.

In yr 1, cows fed late harvested forages yielded less milk, milk fat, and milk protein and had lower DMI than did cows fed early harvested forages. These data were consistent with known (19) effects of forage maturity on cow performance. Milk fat percentage, milk protein percentage, and intake of CP, ADF, and NDF of lactating cows were not affected by late harvesting.

In yr 2, late harvest did not influence milk yield, milk fat percentage, milk fat yield, or milk protein yield. Milk protein percentage and intake of DM, CP, and ADF were reduced ($P < 0.01$) by late harvest.

TABLE 4. Ingredient and nutrient composition of experimental diets.

| Item | Alfalfa | | Red clover | |
|-------------------------|----------|-------|------------|-------|
| | Early | Late | Early | Late |
| | — yr 1 — | | | |
| Ingredient ¹ | | | | |
| Forage | 58.0 | 58.0 | 58.0 | 58.0 |
| Grain mix | 42.0 | 42.0 | 42.0 | 42.0 |
| Nutrient ² | | | | |
| CP | 18.33 | 19.19 | 19.84 | 20.07 |
| ADF | 22.15 | 24.76 | 19.46 | 23.24 |
| NDF | 32.60 | 35.18 | 30.29 | 35.41 |
| Ca | 1.39 | 1.33 | 1.48 | 1.43 |
| P | 0.55 | 0.57 | 0.67 | 0.59 |
| | — yr 2 — | | | |
| Ingredient | | | | |
| Forage | 50.0 | 50.0 | 50.0 | 50.0 |
| Grain mix | 50.0 | 50.0 | 50.0 | 50.0 |
| Nutrient | | | | |
| CP | 16.76 | 17.19 | 17.67 | 16.46 |
| ADF | 26.27 | 26.70 | 22.51 | 24.29 |
| NDF | 35.73 | 35.37 | 29.09 | 33.51 |
| Ca | 0.79 | 0.92 | 0.78 | 0.88 |
| P | 0.49 | 0.58 | 0.53 | 0.55 |

¹As formulated.

²As consumed.

In yr 2, forage quality between early and late harvested forages was similar (Table 2). Milk yield data supported nutrient composition data because no differences in lactation performance were observed in yr 2. Dry matter intake data from yr 2, however, did not support lactation or nutrient composition data. In yr 2, DMI of cows fed late harvested forages was lower ($P < 0.01$) than that of cows fed early harvested forages, suggesting a reduced intake potential of late harvested forages. Cows fed late harvested forages might have supported milk yield through increased fat mobilization, which would explain the inconsistency between the milk yield during yr 2 and DMI data. We cannot validate this argument because no BW measurements were taken.

In yr 1, milk yield of cows fed red clover was similar to milk yield of cows fed alfalfa. In yr 2, cows fed red clover had higher ($P < 0.05$) milk yields than did cows fed alfalfa. Data suggest that cows have similar or slightly improved milk yield potential when fed red clover than when fed alfalfa when both forages are harvested on the same day. Our data did not support equal milk yield potential of cows fed red clover and alfalfa when forages are harvested at similar ADF contents. In yr 1, ADF contents of RL and AL (yr 1, ADF contrast) were similar, and in vitro DM digestibility of RL was 5.9 percentage units higher ($P < 0.01$) than that of AL. Cows fed RL, however, yielded 1.9 kg/d less ($P < 0.05$) milk than did cows fed AL. In yr 2, ADF content and in vitro DM digestibility of RL and AE (yr 2, ADF contrast) were similar, and cows had similar milk yields. Data suggest that when ADF content of red clover and alfalfa are similar, the milk yield of cows fed red clover will be similar or slightly reduced compared with the milk yield of cows fed alfalfa.

Milk fat percentage was not different for cows fed red clover or alfalfa in either year. Milk fat yield was higher ($P < 0.05$) in yr 1 for cows fed alfalfa. Milk fat yield in yr 1 was not different for cows fed RL compared with cows fed AL (ADF contrast). In yr 1, 4% FCM data were reflective of data for milk fat yield. In yr 2, milk fat percentage, milk fat yield, and yield of 4% FCM were not different between red clover and alfalfa or between RL and AE.

The effect of red clover or alfalfa silage on milk protein percentage and yield of lactating cows is also presented in Table 5. In yr 1, milk protein percentage was not different between cows fed red clover or alfalfa or between RL and AL. Milk protein yield was lower ($P < 0.05$) for cows fed red clover. Milk protein yield was also lower ($P < 0.05$) in yr 1 for cows fed RL than for cows fed AL (ADF contrast). In yr 2, milk protein percentage was lower ($P < 0.01$) for cows fed

red clover, but milk protein yield was unchanged. In yr 2, milk protein yield was unchanged because the lower milk protein percentage of cows fed red clover was offset by a higher milk yield.

Milk protein depression in cows fed red clover may be related to altered ruminal protein or carbohydrate metabolism. Polyphenol oxidases that are responsible for enzymatic browning are present in red clover but not in alfalfa (10). Red clover contains several classes of oxidases that have the potential to oxidize phenols and diphenols (10). Oxidized phenols can covalently bond with other compounds, such as amino acids, which could alter their utilization (20). Cows fed red clover might have altered amino acid flow to the small intestine, which could have altered milk protein synthesis. Secondly, Waghorn (25) observed markedly lower ruminal propionate concentrations in Jersey cows fed red clover than in Jersey cows fed alfalfa. Decreased propionate supply to the liver of cows fed red clover could result in increased use of

amino acids for gluconeogenesis, thereby reducing the supply of amino acids for milk protein synthesis (27).

The effect of forages on DM and nutrient intake of lactating cows is presented in Table 5. Dry matter intake was not different for cows fed red clover or alfalfa in either year. Dry matter intake of cows fed red clover was lower (yr 1, $P < 0.05$; yr 2, $P < 0.01$) than that of cows fed alfalfa in yr 1 (RL vs. AL) and in yr 2 (RE vs. RL) when red clover and alfalfa contained similar ADF. The reduced DMI of cows fed red clover when red clover and alfalfa contained similar ADF was related to reduced fiber (ADF and NDF) intake. Intake of ADF and NDF was lower ($P < 0.01$) in yr 1 and 2 for cows fed red clover. Fiber intake was related to DMI and lactation performance. When red clover was compared with alfalfa on an equivalent ADF basis, the intake of ADF, NDF, and DM and the milk yield of cows were lower with the exception of the milk yield of cows fed RL in yr 2. Mechanisms that influence fiber intake of cows fed red clover warrant further investigation. Crude protein intake

TABLE 5. The effect of experimental forages on lactation performance and intakes of DM, CP, ADF, and NDF.

| Item | Alfalfa | | Red clover | | SE | P^1 | | |
|--------------------------|---------|------|------------|------|------|-----------------|----------|-----|
| | Early | Late | Early | Late | | Forage | Maturity | ADF |
| yr 1 | | | | | | | | |
| Lactation performance | | | | | | | | |
| Milk yield, kg/d | 33.5 | 31.0 | 33.6 | 29.1 | 0.5 | NS ² | ** | * |
| Milk fat, % | 3.81 | 3.62 | 3.58 | 3.81 | 0.07 | NS | NS | NS |
| Milk protein, % | 3.19 | 3.20 | 3.16 | 3.15 | 0.02 | NS | NS | NS |
| Milk fat yield, kg/d | 1.27 | 1.12 | 1.19 | 1.09 | 0.02 | * | ** | NS |
| Milk protein yield, kg/d | 1.06 | 0.98 | 1.05 | 0.91 | 0.02 | * | ** | ** |
| 4% FCM, kg/d | 32.4 | 29.1 | 31.3 | 28.0 | 0.5 | ** | * | NS |
| Intake, kg/d | | | | | | | | |
| DM | 21.7 | 21.6 | 21.9 | 18.8 | 0.7 | NS | * | * |
| CP | 4.0 | 4.1 | 4.4 | 3.8 | 0.2 | NS | NS | NS |
| ADF | 4.8 | 5.4 | 4.3 | 4.4 | 0.2 | ** | NS | NS |
| NDF | 7.1 | 7.6 | 6.7 | 6.7 | 0.2 | ** | NS | ** |
| yr 2 | | | | | | | | |
| Lactation performance | | | | | | | | |
| Milk yield, kg/d | 30.3 | 30.0 | 30.7 | 31.1 | 0.3 | * | NS | NS |
| Milk fat, % | 3.92 | 3.94 | 3.94 | 3.82 | 0.04 | NS | NS | NS |
| Milk protein, % | 3.39 | 3.31 | 3.28 | 3.23 | 0.02 | ** | ** | ** |
| Milk fat yield, kg/d | 1.17 | 1.16 | 1.19 | 1.18 | 0.02 | NS | NS | NS |
| Milk protein yield, kg/d | 1.02 | 0.98 | 0.99 | 0.99 | 0.01 | NS | NS | NS |
| 4% FCM, kg/d | 29.6 | 29.4 | 30.1 | 30.2 | 0.4 | NS | NS | NS |
| Intake, kg/d | | | | | | | | |
| DM | 20.5 | 18.8 | 20.9 | 18.5 | 0.3 | NS | ** | ** |
| CP | 3.4 | 3.2 | 3.7 | 3.0 | 0.1 | NS | ** | ** |
| ADF | 5.4 | 5.0 | 4.8 | 4.5 | 0.1 | ** | ** | ** |
| NDF | 7.3 | 6.8 | 6.1 | 6.2 | 0.2 | ** | NS | ** |

¹ADF contrast: yr 1, late alfalfa versus late red clover; yr 2, early alfalfa versus late red clover.

² $P > 0.05$.

* $P < 0.05$.

** $P < 0.01$.

was not affected by forage type in either year. In yr 2, CP intake of RL was lower ($P < 0.01$) than that of AE (ADF contrast) and was a reflection of DMI of the diets.

CONCLUSIONS

Data suggest that red clover supports similar or slightly improved milk yield compared with alfalfa when both are harvested for silage on the same day. When ADF content of red clover and alfalfa are similar, DMI will be lower, and milk yield may be reduced, for cows fed red clover.

Cows fed red clover also had reduced milk protein synthesis. Data demonstrate that the utilization of red clover in lactating dairy cows is different than that of alfalfa. Further research is required to establish mechanisms of nutritional deficiencies of red clover.

REFERENCES

- 1 Albrecht, K. A., and R. E. Muck. 1991. Proteolysis in ensiled forage legumes that vary in tannin concentration. *Crop Sci.* 31: 464.
- 2 Aman, P. 1985. Chemical composition and in vitro degradability of major chemical constituents in botanical fractions of red clover harvested at different stages of maturity. *J. Sci. Food Agric.* 36:775.
- 3 Association of Official Analytical Chemists. 1990. *Official Methods of Analysis*. Vol. I. 15th ed. AOAC, Arlington, VA.
- 4 Buxton, D. R. 1989. In vitro digestion kinetics of temperate perennial forage legume and grass stems. *Crop Sci.* 29:213.
- 5 Cherney, D.J.R., J. H. Cherney, and R. F. Lucey. 1993. In vitro digestion kinetics and quality of perennial grasses as influenced by forage maturity. *J. Dairy Sci.* 76:790.
- 6 Gjerde, D. T., and J. S. Fritz. 1987. *Ion Chromatography*. 2nd ed. CIP-Kurztitelaufnahme der Deutschen Bibliothek., New York, NY.
- 7 Goering, H. K., and P. J. Van Soest. 1970. *Forage Fiber Analyses (Apparatus, Reagents, Procedures, and Some Applications)*. Agric. Handbook No. 379. ARS-USDA, Washington, DC.
- 8 Hoffman, P. C., S. J. Sievert, R. D. Shaver, D. A. Welch, and D. K. Combs. 1993. In situ dry matter, protein, and fiber degradation of perennial forages. *J. Dairy Sci.* 76:2632.
- 9 Hoffman, P. C., D. A. Welch, and N. M. Brehm. 1995. Potential of enzyme mixtures to improve silage quality and lactation performance of dairy cattle. *J. Prod. Agric.* 8:552.
- 10 Jones, B. A., R. E. Muck, R. D. Hatfield, and D. R. Mertens. 1994. Nutrient detergent fiber nitrogen and proteolysis in alfalfa silages treated with red clover herbage and extracts. *J. Dairy Sci.* 77(Suppl. 1):275.(Abstr.)
- 11 Kalu, B. A., and G. W. Fick. 1981. Quantifying morphological development of alfalfa for studies of herbage quality. *Crop Sci.* 21:267.
- 12 McCullough, M. E. 1978. Silage—some general considerations. Page 3 in *Fermentation of Silage—A Review*. Natl. Feed Ingredients Assoc., West Des Moines, IA.
- 13 Mertens, D. R. 1992. Critical conditions in determining detergent fibers. Page C-1 in *Proc. Natl. Forage Testing Assoc. Forage Anal. Workshop*, Denver, CO. Natl. Forage Testing Assoc., Omaha, NE.
- 14 Mertens, D. R., and J. R. Lofton. 1980. The effects of starch on forage fiber digestion in vitro. *J. Dairy Sci.* 63:1437.
- 15 National Research Council. 1989. *Nutrient Requirements for Dairy Cattle*. 6th rev. ed. Natl. Acad. Sci., Washington, DC.
- 16 Nocek, J. E. 1988. In situ and other methods to estimate ruminal protein and energy digestibility: a review. *J. Dairy Sci.* 71:2051.
- 17 Nocek, J. E., and A. L. Grant. 1987. Characterization of in situ nitrogen and fiber digestion and bacterial nitrogen contamination of hay crop forages preserved at different dry matter percentages. *J. Anim. Sci.* 64:552.
- 18 Ohlsson, C., and W. F. Wedin. 1989. Phenological staging schemes for predicting red clover quality. *Crop Sci.* 29:416.
- 19 Paterson, J. A., R. L. Belyea, J. P. Bowman, M. S. Kerley, and J. E. Williams. 1994. The impact of forage quality and supplementation regime on ruminant animal intake and performance. Page 59 in *Forage Quality, Evaluation, and Utilization*. G. C. Fahey, Jr., ed. Am. Soc. Agron., Crop Sci. Soc. Am., and Soil Sci. Soc., Madison, WI.
- 20 Reed, J. D. 1995. Nutritional toxicology of tannins and related polyphenols in forage legumes. *J. Anim. Sci.* 73:1516.
- 21 Robertson, J. B., and P. J. Van Soest. 1981. The detergent system of analysis and its application to human foods. Page 123 in *The Analysis of Dietary Fiber in Foods*. W.P.T. James and O. Theander, ed. Marcel Dekker, New York, NY.
- 22 SAS® User's Guide: Statistics, Version 5 Edition. 1985. SAS Inst., Inc., Cary, NC.
- 23 Smith, D. 1964. Chemical composition of herbage with advance in maturity of alfalfa, medium red clover, ladino clover and birdsfoot trefoil. *Wisconsin Agric. Exp. Stn. Res. Rep.* 16. Univ. Wisconsin, Madison.
- 24 Undersander, D., R. R. Smith, K. Kelling, J. Doll, G. Worf, J. Wedberg, J. Peters, P. Hoffman, and R. Shaver. 1990. *Red clover: establishment, management and utilization*. Publ. A3492. Univ. Wisconsin-Extension, Madison.
- 25 Waghorn, G. C. 1986. Changes in rumen digestion of cows during a restricted feeding period when offered fresh red clover, lucerne, or lucerne hay. *N.Z. J. Agric. Res.* 29:233.
- 26 Weiss, W. P. 1994. Estimation of digestibility of forages by laboratory methods. Page 644 in *Forage Quality, Evaluation, and Utilization*. G. C. Fahey, Jr., ed. Am. Soc. Agron., Crop Sci. Soc. Am., and Soil Sci. Soc., Madison, WI.
- 27 Wu, Z., F. T. Sleiman, C. B. Theurer, F. Santos, J. M. Simas, M. Francolin, and J. T. Huber. 1994. Effect of isocaloric infusion of glucose in the rumen or propionate in the duodenum. *J. Dairy Sci.* 77:1556.