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Diurnal Shifts in Nutritive Value of Alfalfa Harvested as Hay and Evaluated by Animal Intake and Digestion

J. C. Burns,* D. S. Fisher, and H. F. Mayland

ABSTRACT

Forages accumulate nonstructural carbohydrates during the day, with animals showing preference and improved daily responses from afternoon compared with morning cut hays. This study evaluated alfalfa (Medicago sativa L.) hay harvested at 0700, 1000, 1300, 1600, and 1900 h to determine how nutritive value changes during the day and to assess the impact of these changes on animal preference using cattle (Bos taurus L.), sheep (Ovis aries L.), and goat (Capra hircus L.) responses. Total nonstructural carbohydrates were altered by time of cut (cubic contrast, P < 0.01) ranging from 85 g kg⁻¹ at 0700 h to 83 g kg⁻¹ at 1000 h, then increasing to 97 g kg⁻¹ by 1600 h with little change at 1900 h (96 g kg⁻¹). Fiber fractions also varied diurnally, with a quadratic decrease from 418 g kg⁻¹ at 0700 h to 387 g kg⁻¹ by 1900 h in neutral detergent fiber. A combined analysis of three animal trials showed a linear increase in dry matter intake (DMI) with later hay harvest, a cubic response for dry matter digestion (DMD), and a linear increase in digestible DMI. Mean DMI increased from 27.5 g kg-1 body weight at 0700 h to a maximum of 30.8 g kg⁻¹ body weight at 1600 h, whereas DMD decreased from 658 g kg⁻¹ at 0700 to 647 g kg⁻¹ at 1300 h and peaked at 664 g kg⁻¹ at 1600 h. Digestible DMI increased from 18.1 g kg⁻¹ body weight at 0700 h to a maximum of 20.5 g kg⁻¹ body weight at 1600 h. No additional advantages in animal responses were noted by cutting after 1600 h.

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Abbreviations: ADF, acid detergent fiber; CP, crude protein; DMD, dry matter digestion; DMI, dry matter intake; IVTD, in vitro true dry matter disappearance; LOF, lack of fit; NDF, neutral detergent fiber; TNC, total nonstructural carbohydrate.

ANAGEMENT STRATEGIES that add quality to a product Management strategies care and any without altering either production or processing costs are valuable contributions to the industry. The accumulation of photosynthate in forage tissue during the day was reported in the literature (Bowden et al., 1968; Lechtenberg et al., 1971; Gordon, 1996) and has recently been applied to improve the quality of forage cut for hay. For example, forage cut in the afternoon for hay vs. the morning was consistently greater in total nonstructural carbohydrate (TNC) concentrations in tall fescue (Festuca arundinacea Schreb.) (95 vs. 79 g kg⁻¹; Fisher et al., 1999), orchardgrass (Dactylis glomerata L.) (125 vs. 115 g kg⁻¹; Griggs et al., 2005), switchgrass (Panicum virgatum L.) (81 vs. 72 g kg⁻¹; Fisher et al., 2005) and alfalfa (Medicago sativa L.) (54 vs. 42 g kg⁻¹; Fisher et al., 2002). The accumulation of TNC in these forages was also generally associated with a reduction in the concentrations of neutral detergent fiber (NDF) and its constituent fiber fractions.

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The significance of such diurnal shifts in nutritive value depends on the extent to which they can be detected by animals and produce an impact on daily animal response and hence forage quality. Preference trials have shown previously that steers, sheep, and goats could detect the difference between forage cut for hay in the afternoon vs. the morning for alfalfa (Fisher et al., 2002) and tall fescue (Fisher et al., 1999), but only by goats and sheep for switchgrass (Fisher et al., 2005). Such afternoon and morning variation in nutritive value and preference have also been reflected in daily dry matter intake (DMI) and dry matter digestion (DMD) of alfalfa fed to goats (Burns et al., 2005) and in long-term lactation experiments using lactating cows and heifers (Mayland et al., 2005). The management strategy of delaying cutting on a sunny day to accumulate maximum TNC concentrations reduces

day to accumulate maximum TNC concentrations reduces the hours available that day for drying purposes. This is especially important in humid regions where the time between rainfall events may be limited to 3 to 4 d. Because TNC concentrations increase during the day and ruminants prefer forages with greater TNC (Orr et al., 2001), an assessment is warranted to determine the time of day when the magnitude of the diurnal shift is at an optimum to be detected by ruminants and result in improved daily response. If concentrations of TNC increase rapidly and peak early rather than late in the afternoon, then hay could be cut earlier in the day and sufficient drying may occur to reduce plant respiratory losses and limit the risk of rain damage (Rotz and Muck, 1994; Moser, 1995; Rotz, 1995). Such data are not available in the literature. The objective of this study was to estimate the optimum time of day to cut alfalfa to maximize nutritive value and animal DMI and digestion. Hay treatments were evaluated by cattle, sheep, and goats in three experiments.

MATERIALS AND METHODS Source of Hays

Second regrowth hay (40 cm in height) was harvested on 4 September at the early-bud stage from a third-year production stand of Germain WL 322HQ alfalfa near Kimberly, ID ($42^{\circ}32'22.42''N$, $114^{\circ}20'47.92''W$, elevation 1200 m). Subsequent cuts of the regrowth and associated solar radiation were taken during a sunny day at 0700 (nil), 1000 (670 W m^{-2}), 1300 (2720 W m^{-2}), 1600 (5060 W m^{-2}), and 1900 h (6430 W m^{-2}). The hay, with a mean dry matter yield of 3800 kg ha⁻¹, was field cured in the absence of rain and hay from each cut was baled separately on the same day. Each bale was tagged separately using color coding and each treatment stored separately under tarps. The hay was transported to Raleigh, NC, for subsequent animal evaluation. Once at Raleigh, the hay was grouped by treatment and stored on wood pallets in a metal building designed for the storage of experimental hay.

Alfalfa hay was harvested at the late vegetative stage in Raleigh and used for all standardization periods. Just before feeding, all hay was passed through a hydraulic Van Dale 5600 bale processor (J. Starr Industries, Fort Atkinson, WI) with stationary knives spaced 10 cm apart. This reduced the hay length (range 8–13 cm) for feeding with minimal leaf loss.

Intake and Digestion Procedure and Design

Dry matter intake and digestion trials were conducted with conventional protocols using steers, sheep, and goats (Burns et al., 1994). The animal care and handling procedures were approved by the North Carolina State University Institution Animal Care and Research Committee (Approval no. 03-047A). In the steer trial, 20 Angus steers were confined in the intake facility fitted with electronic gates (Calan gate system, American Calan, Northwood, NH) (Burns et al., 1997). Each steer was fitted with a key to permit access to only one manger, but animals could lounge together and had free access to trace-mineralized salt and water. After acclimation to the gates, each animal was fed the standard alfalfa hay for 14 d. The steers, ranging from 263 to 354 kg, were blocked by weight into four groups of five animals each, and randomly assigned to one of the five treatments within each group using a randomized complete block design with four animals (replicates) per treatment. Each of the four replicates was accommodated in the facility on a staggered schedule due to the limited number of digestion crates. Each period lasted 18 d and consisted of a 9-d intake phase followed by a 9-d digestion phase consisting of a 4-d adjustment to the crates followed by a 5-d total fecal collection period.

The intake and digestion trials with sheep and goats were conducted using conventional wooden crates. Five Katahdin wether sheep (initial weight range 34-44 kg) and five Boer × Spanish wether goats (initial weight range 34-42 kg) were each used in 5 \times 5 Latin square designs. The animals were placed into digestion crates located in an enclosed, well-ventilated building. Each animal was fitted with a conventional collection harness with canvas fecal collection bags unzipped and positioned to avoid collection of feces during the acclimation and intake phases. All animals were initially fed the standard alfalfa hav for a 14-d adjustment period. Animals were then randomly assigned to one of the experimental hays to begin the first period of the Latin square. Each period lasted 28 d and consisted of a 7-d adjustment to the experimental forage, followed by a 14-d intake period, then followed by a 7-d digestion period with daily total fecal collection occurring during the last 5 d. Total fecal collection was achieved by simply repositioning the canvas collection bags, inserting a plastic bag liner, and zipping them up. At the end of the third period, the animals were removed from the crates for a 7-d break and were fed the standard alfalfa hay until initiating Period 4.

Feeding and Sampling

All animals were fed at approximately 112% ad libitum intake in all trials. A recorded weight of hay was fed twice daily based on the previous day's intake. To adequately reflect the composition of the hay fed throughout the trial, a daily sample of each hay was obtained during each experimental period and composites were made for a 7-d period. Orts from each animal were weighed twice daily and composited every 7 d. The two 7-d samples were further composited for each experimental period. In the digestion phase of each trial, the feed and ort samples were composited for the 5-d collection period and analyzed separately from the samples taken during the intake phase. All forage samples were thoroughly mixed, subsampled, oven dried (55°C), ground in a Wiley mill (Thomas Scientific, Swedesboro, NJ) to pass a 1-mm screen, and stored in an air-tight container at room temperature until analyzed.

In the digestion trials, feces were collected and weighed for each consecutive 24-h period. Feces were thoroughly mixed daily, and approximately 5% of the fresh weight placed in a freezer (-14° C). At the end of the 5-d collection, the composite frozen samples were oven dried (55°C), weighed for dry matter determination, ground in a Wiley mill to pass a 1-mm screen, thoroughly mixed, subsampled, and stored at room temperature until analyzed. All intake and apparent digestion data are presented on an oven-dry-matter basis.

Laboratory Analyses

Composition and in vitro true dry matter disappearance (IVTD) of fed hays and orts and composition of fecal samples from each of the three experiments were determined using a near-infrared reflectance spectrophotometer (NIRS). All samples were first scanned through a Model 5000 NIRS (Foss North America, Eden Prairie, MN). Samples with different spectra (using H-distance > 0.6) were designated for laboratory analyses.

Alfalfa feed and orts samples (n = 281) were compared with a library containing previously analyzed alfalfa. Ten spectral outliers (H-distance > 3.0) were removed before selection against the library. Thirteen of the 281 samples were selected for laboratory analysis (fiber fractions, crude protein [CP], and IVTD) and an additional 10 samples were selected at random for inclusion in the calibration data set. Fecal samples (n = 70) were compared with a

Table 1. Calibration range for each forage, ort, and fecal constituent predicted by near-infrared reflectance spectroscopy, the standard error of calibration (SEC) and standard error of cross-validation (SECV), and associated coefficients of determination (R^2) for appropriate calibration data sets.

Sampla	Variablat		Range	Mean	Calibration		Validation	
Sample	Variable [†]	n		wear	SEC	R^2	SECV	R^2
				g kg-1 –			g kg ⁻¹	
Forage/orts	DM	179	908–954	937	1.5	0.980	1.9	0.967
Forage/orts	NDF	181	327–682	464	6.3	0.994	8.8	0.988
Forage/orts	ADF	183	248-540	351	7.1	0.989	8.4	0.985
Forage/orts	cellulose	180	186–423	268	5.8	0.988	6.4	0.985
Forage/orts	lignin	177	54–118	76	2.2	0.979	2.5	0.973
Forage/orts	CP	181	80–259	193	4.1	0.990	5.4	0.983
Forage/orts	IVTD	178	512-845	724	15.4	0.948	17.5	0.933
Forage	TNC	98	18–106	55	3.0	0.982	4.6	0.958
Forage	MS	95	3–23	13	1.3	0.903	1.6	0.855
Forage	starch	99	0.6–29	10	1.7	0.930	2.3	0.874
Fecal	DM	316	893–972	929	3.5	0.948	4.0	0.935
Fecal	NDF	316	439–743	643	9.4	0.983	12.4	0.971
Fecal	ADF	317	288–435	363	7.8	0.929	8.9	0.907
Fecal	cellulose	311	185–329	263	4.6	0.976	5.2	0.969
Fecal	lignin	308	55–155	93	4.0	0.974	4.6	0.965
Fecal	CP	312	66–174	108	2.9	0.985	3.2	0.982

[†]DM, dry matter; NDF, neutral detergent fiber; ADF, acid detergent fiber; CP, crude protein; IVTD, in vitro true dry matter digestion; TNC, total nonstructural carbohydrates; MS, monosaccharides. library containing only fecal samples. Four samples were selected for laboratory analysis (fiber fractions and CP) and inclusion in the calibration data set. Only the alfalfa feed samples from the intake and digestion phases (n = 145) were analyzed for TNC. After removal of two outliers, samples were compared with a library of previously analyzed alfalfa hay. Five samples were selected and an additional 15 samples were chosen for analysis of TNC and constituent monosaccharides and starch concentration. After chemical analysis and inclusion of newly selected samples in their respective libraries, all calibrations were redeveloped (Table 1).

In vitro true dry matter disappearance was determined using ruminal inoculum collected from a cannulated mature Hereford steer fed a mixed alfalfa and orchardgrass hay. After batch incubation for 48 h with ruminal inoculum combined with artificial saliva (Burns et al., 1970) in fermentation vessels (Ankom Technology Corp., Fairport, NY), samples were extracted with neutral detergent solution for estimation of IVTD. Fiber fractions (NDF, acid detergent fiber [ADF], cellulose, and H₂SO₄ lignin) were estimated on the feed, ort, and fecal samples according to Van Soest and Robertson (1980) in a batch processor (Ankom Technology Corp., Fairport, NY). Crude protein was calculated on the same samples as 6.25 times the N concentration as determined with an autoanalyzer (AOAC International, 1990). Total nonstructural carbohydrates were analyzed according to Burns et al. (2006). The TNC were fractionated into monosaccharides and starch, with disaccharides and short-chain polysaccharides determined by difference.

Following laboratory analyses, the spectra from the nearinfrared reflectance spectroscopy for each sample and corresponding laboratory values were used to develop appropriate calibration equations. These equations were then applied to the remainder of the samples to estimate concentrations of each variable (Table 1). All data were reported on a dry-matter basis.

Statistical Analysis

The data from the steer trial were analyzed as a randomized complete block design. The model included terms for animal and treatment. All data from the sheep and goat trials were analyzed as a 5 \times 5 Latin square design. In all cases, the model included terms for animal, period, and treatment. In both the randomized complete block and the Latin squares, the error term was used to test fixed effects for significance according to the F test (Steel and Torrie, 1980). Means for all variables analyzed were examined for timeof-cut effects. The four degrees of freedom in the sum of squares for the five treatments were separated into single degree of freedom contrasts testing for linear, quadratic, and cubic diurnal effects, and lack of fit (LOF). If the LOF was significant, then the means for one or more cuts deviated significantly from linear, quadratic, and cubic fits. All significant contrasts are presented. A meta-analysis using the Proc Mixed procedure of SAS (SAS Institute, 2004) provided a way of comparing diurnal changes using the composition of the "as fed" hay from all three experiments. The individual trials and replications within each animal trial were considered random and the time of cut was a fixed effect. All forage and composition data were considered significant at $P \le 0.05$. Simple linear correlation was used to examine the relationship among estimates of nutritive value and measured DMI, DMD, and digestible DMI from the intake and digestion trials as well as other relationships of interest.

Because of the small difference expected among hay cut every 3 h and the variation normally present in intake and digestion trials, a decision was made a priori to consider animal intake, digestion, and digestible intake significant with statistical tests at $P \le 0.10$. For the summary discussion, a meta-analysis (as described above) for all the daily intake (as a proportion of animal weight) and digestion data from all three animal trials was conducted using the Proc Mixed procedure of SAS (SAS Institute, 2004).

RESULTS AND DISCUSSION Diurnal Shifts

The best estimates of the mean nutritive value of the alfalfa hay from the five diurnal cutting times (0700–1900 h) were obtained by combining estimates from the "as fed" hay samples collected at feeding of each animal replication in each of the three animal experiments (see below). This provided an estimate of the rela-

tive diurnal variation in the forage that was actually delivered to the feed bunks of the animals.

Because of the active accumulation of soluble carbohydrates with the onset of sunlight (Bowden et al., 1968; Lechtenberg et al., 1971; Gordon, 1996), the TNC concentrations were viewed as driving the observed diurnal cycle in nutritive value. This may take the form of simple dilution or more complex aspects of synthesis, metabolism, and transport. Total nonstructural carbohydrate concentrations were altered by time of cut, increasing (significant cubic contrast) from 85 g kg⁻¹ at 0700 h to 97 g kg⁻¹ by 1600 h, with little further change occurring by 1900 h (Table 2). In general, the constituent monosaccharides and starch followed the same diurnal cycle as noted for TNC, whereas di- and polysaccharides increased linearly.

The overall diurnal increase in TNC is consistent with published literature for afternoon samples of alfalfa (Plhak, 1991) and perennial ryegrass (Lolium perenne L.) (Orr et al., 2001) and was associated with increased IVTD (r = 0.83; $P \le 0.01$), but decreased NDF (r =-0.78; $P \le 0.01$) and constituent fiber fractions of ADF $(r = -0.80; P \le 0.01)$, hemicellulose (r = -59; P = 0.02), cellulose (r = -0.80; $P \le 0.01$), and lignin (r = -0.81; P ≤ 0.01). The IVTD increased linearly from 757 g kg⁻¹ at 0700 h to 777 g kg⁻¹ by 1900 h. The decrease in NDF was quadratic, with little change noted until 1600 h (Table 2). Crude protein concentration showed a quadratic change, with a decline from 0700 to 1300 h and an increase through 1900 h; however, the concentrations at 1600 and 1900 h were similar. These data indicate that N concentrations in the forage increased as the day progressed and did not decline in response to carbohydrate accumulation.

Table 2. Mean in vitro true dry matter disappearance (IVTD), crude protein (CP), neutral detergent fiber (NDF) and constituent fiber fractions, and nonstructural carbohydrate concentrations of alfalfa hay fed to steers, sheep, and goats (oven-dry basis).

Time			Fiber fraction [†]			Nonstr	uctural	carbohy	/drates [‡]		
Time IVTD		СР	NDF	ADF	HEMI	CELL	Lignin	MS	D/PS	St	TNC
h						g l	<g-1< td=""><td></td><td></td><td></td><td>_</td></g-1<>				_
0700	757§	204	418	312	107	241	66	18	47	20	85
1000	757	202	419	310	110	240	66	17	48	18	83
1300	760	199	414	304	110	235	65	19	51	22	92
1600	772	204	397	293	104	228	63	22	52	24	97
1900	777	206	387	291	96	223	61	21	51	24	96
Significance (P)											
Time	< 0.01	0.11	<0.01	< 0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
Trend¶ (P ≤ 0.05)) L	Q	L, Q	L	L, Q	L	L	L, C	L	L, C	L, C
SE	4	3.0	7	4	4	4	0.10	0.4	1	0.8	2

[†]ADF, acid detergent fiber; HEMI, hemicellulose; CELL, cellulose.

[‡]MS, monosaccharide; D/PS, di- and polysaccharides; St, starch; TNC, total nonstructural carbohydrates. [§]Each value is the mean from four steers, five goats, and five sheep (n = 14).

¹L, linear; Q, quadratic; C, cubic.

The diurnal shifts in these nutritive value constituents were all statistically significant, but with little change after the 1600-h cutting time. Furthermore, the magnitudes of the diurnal changes in concentration were relatively small, and raise the question of the importance of the effect on forage quality. This aspect is addressed in this research through animal intake and digestion trials.

Animal Assessment

The nutritive value of alfalfa was significantly improved (i.e., increased IVTD and reduced NDF) as cuts were delayed from morning to afternoon. The biological significance of these diurnal shifts in alfalfa was examined using three species of ruminants with their intake, apparent digestion, and apparent digestible intake as the major criteria of evaluation.

Evaluation with Steers

Steer Responses Significant differences were found among the hay with steers as DMI was altered by time of cut (P = 0.01). Linear, quadratic, and cubic contrasts were not significant, however, from 0700 to 1900 h (Table 3). The significant LOF indicates, however, that the means differed among the five treatments, with the greatest DMI from the 1600-h hay cut. Dry matter digestion was not different among the cutting times. Multiplying DMI by the DMD coefficient showed digestible DMI to reflect DMI, as differences occurred among cuts (P = 0.04). Again, no linear, quadratic, or cubic trend was noted, but the significant LOF indicated that means differed, with digestible DMI greatest at 1600 h.

Crude protein digestion was not altered by time of cut nor was any diurnal trend evident (Table 3). Table 3. Mean daily dry matter intake (DMI), apparent digestion, and apparent digestible intakes of dry matter (DM), crude protein (CP), and neutral detergent fiber (NDF) of alfalfa hay fed to steers (oven-dry basis).

Time	DMI [†]	Apparent digestion			Apparent digestible intake					
Time	DIVIT	DM	CP	NDF	DM	CP	NDF			
		g kg ^{_1}								
0700	28.1‡	651	782	513	18.3	4.6	5.8			
1000	29.4	637	760	495	18.8	4.7	5.8			
1300	27.4	622	748	465	17.1	4.2	5.1			
1600	31.6	656	767	499	20.7	5.0	6.0			
1900	28.1	620	755	431	17.5	4.4	4.6			
	Significance (P)									
Time	0.01	0.41	0.12	0.28	0.04	0.02	0.13			
Trend§ (<i>P</i> ≤ 0.10)	LOF	NS	NS	L	C, LOF	LOF	LOF			
SE	0.09	21	10	35	0.10	0.02	0.05			

[†]Intake expressed on a body-weight basis.

[‡]Each value is the mean of four animals.

§C, Cubic; LOF, lack of fit; NS, not significant.

On the other hand, NDF digestion declined linearly from 0700 to 1900 h. Digestible intake of CP and NDF were altered by time of cut, but contrasts showed only a significant LOF. The significant LOF indicates that the response was not linear, quadratic, or cubic but at least one treatment mean differed from the others. The 1600-h mean was greater for both digestible CP and NDF intakes. Generally, there was no advantage in animal response after the 1600-h cut.

Hay Composition and In Vitro True Dry Matter Disap-

pearance The "as fed" alfalfa from this experiment showed TNC to be altered by time of cut. The response was cubic, with TNC increasing from 0700 to 1600 h but with concentration greatest at 1600 h (Table 4).

The IVTD concentrations increased linearly from 0700 to 1900 h (Table 4). Crude protein concentrations were not altered by time of cut nor did they show any diurnal trends. Concentrations of NDF, however, were altered by time of cut, showing a linear decrease from 0700 to 1900 h. This is consistent with the responses observed for TNC concentration and IVTD.

Evaluation with Sheep

Sheep Responses Sheep detected changes in the hay based on time of cut, since DMI was altered by time of cut and showed a linear diurnal increase (Table 5). Dry matter digestion varied with time of cut (cubic response from 0700–1900 h) with greatest DMD at 1600 h. Digestible DMI increased linearly with time of cut and was greatest at 1900 h (Table 5).

Crude protein digestion was not altered by time of cut nor was any diurnal trend evident but the digestion

Table 4. Mean in vitro true dry matter disappearance (IVTD), crude protein (CP), neutral detergent fiber (NDF), and total nonstructural carbohydrates (TNC) concentration of alfalfa hay fed to steers (oven-dry basis).

Time	IVTD	СР	NDF	TNC			
h		g l	<g<sup>-1</g<sup>				
0700	764 ⁺	209	407	87			
1000	765	210	402	84			
1300	761	202	403	92			
1600	777	208	385	102			
1900	780	209	373	100			
	Significance (P)						
Time	0.08	0.22	<0.01	<0.01			
Trend [‡] ($P \le 0.05$)	L	NS	L	L, C			
SE	6	4	8	3			

*Each value is the mean of four samples.

[‡]L, linear; NS, not significant; C, cubic.

of NDF declined (cubic trend), with a peak at 1600 h and reduced digestion at 1000 and 1900 h (Table 5).

Digestible intake of CP was altered by time of cut, showing a linear diurnal increase. Digestible NDF intake was not significantly altered by time of cut in an analysis of variance, but contrasts showed a cubic diurnal trend, with means declining from 0700 to 1000 h then increasing at 1300 h, with little difference among the later cut times (Table 5). As noted for steers, there was generally no improvement in animal response after the 1600-h cut.

Hay Composition and In Vitro True Dry Matter Disappearance The concentration of TNC was altered by time of cut, showing a linear diurnal increase peaking at 1600 h, with little difference in concentration between 1600 and 1900 h (Table 6). The diurnal shift in IVTD was similar to TNC. Crude protein concentrations showed a quadratic diurnal change, decreasing from 0700 to 1000 h, then increasing to 1900 h. Neutral detergent fiber concentrations were altered by time of cut and showed a linear diurnal trend (Table 6).

Evaluation with Goats

Goat Responses Goats generally did not discern differences among hays, as DMI, DMD, and digestible DMI were not altered by time of cut nor was any diurnal trend evident. Neither CP nor NDF digestion was altered by time of cut. This was also the case for digestible CP intake. Digestible NDF intake showed a significant LOF, indicating that while linear, quadratic, and cubic effects were not significant, at least one mean among the five was different (Table 7). The greatest digestible NDF intake occurred from hay cut at 1600 h.

Hay Composition and In Vitro True Dry Matter Disappearance The concentration of TNC was altered by time of

cut, showing a cubic diurnal shift (Table 8). Concentration declined from 0700 to 1000 h but increased to 1300 h, with no change thereafter. The associated concentrations of both IVTD and NDF were altered by time of cut, with the expected increase in IVTD and decrease in NDF, both showing linear diurnal trends. Major changes occurred between 1000 and 1600 h. Crude protein concentrations were not altered by time of cut.

Integration of Experiments

Rather striking is the degree to which animals can detect subtle compositional differences in offered feed. In the steer trial, the range differential in TNC that occurred between 0700 and 1600 h in the "as fed" hay averaged 18.8 g kg⁻¹ and the animals detected this difference, as reflected in DMI and DMD responses. Likewise, the range for sheep averaged 14.6 g kg⁻¹ and they detected this difference, as reflected in DMI and DMD responses. Goats, on the other hand, appeared less discerning in this experiment, as the TNC differential averaged 14.1 g kg⁻¹ but no significant change in goat DMI was noted. This lack of response was unexpected, as goats detected differences in alfalfa of only 10 g kg⁻¹ TNC in previous studies comparing preference and intake from afternoon vs. morning harvests (Fisher et al., 2002; Burns et al., 2005). Perhaps the magnitude of the TNC difference was not sufficient to be detected by the set of goats used in this trial or perhaps the goats differed from sheep in their selective consumption of the "as fed" hay.

Examination of the IVTD and NDF concentrations of the orts minus the concentrations of the "as fed" forage sample (difference values) may provide some insight since preference for leaf tissue, even in hay, would result in reduced IVTD and increased NDF of the orts vs. the "as fed" forage. The respective difference values (data for orts not shown) for IVTD and NDF were -37 and 48 g $\rm kg^{-1}$ for steers, -68 and $88~g~\rm kg^{-1}$ for goats, and -83 and 117 g kg⁻¹ for sheep. In a meta-analysis of difference values for IVTD and NDF among ruminant species, the species main effect was significant (P = 0.02) for both variables. Furthermore, difference values for steers were least compared with sheep (P < 0.01 for both IVTD and NDF) or goats (P = 0.05 for IVTD and P = 0.09 for NDF), whereas difference values were similar for sheep and goats (P =0.26 for IVTD and P = 0.17 for NDF). Although difference values between goats and sheep were similar, goats apparently did not perceive the differences as the sheep did. We speculate that when diurnal changes in TNC are small, as noted in this study, the degree of selectivity (or lack thereof as noted for goats) becomes more biologically critical than if diurnal changes are larger.

Combining the dry matter response data (i.e., DMI, DMD, and digestible DMI) for all three experiments in a meta-analysis showed an overall linear increase (P = 0.04)

Table 5. Mean daily dry matter intake (DMI), apparent digestion, and apparent digestible intakes of dry matter (DM), crude protein (CP), and neutral detergent fiber (NDF) of alfalfa hay fed to sheep (oven-dry basis).

Time	DMI	Digestion			Dige	Digestible intake [†]			
Time	DIVII	DM	СР	NDF	DM	CP	NDF		
h				—g kg ^{_1}					
0700	28.3‡	659	796	553	18.7	4.6	6.5		
1000	27.2	645	793	521	17.6	4.3	6.0		
1300	29.8	655	790	552	19.5	4.7	6.7		
1600	32.4	670	799	555	21.7	5.4	6.9		
1900	34.2	654	790	519	22.4	5.7	6.6		
	Significance(P)								
Time	0.02	0.29	0.78	0.17	0.02	0.01	0.25		
Trend§ ($P \le 0.10$)	L	С	NS	С	L	L	С		
SE	0.31	11	11	18	0.21	0.05	0.07		

[†]Intake expressed on a body-weight basis.

*Each value is the mean of five animals.

§L, linear; C, cubic; NS, not significant.

Table 6. Mean in vitro true dry matter disappearance (IVTD), crude protein (CP), neutral detergent fiber (NDF), and total nonstructural carbohydrates (TNC) concentration of alfalfa hay fed to sheep (oven-dry basis).

IVTD	СР	NDF	TNC
	g l	kg ⁻¹	
750 ⁺	200	429	82
755	194	431	85
757	194	423	93
772	202	402	96
778	206	389	95
	Signific	ance (P)	
0.03	0.09	<0.01	<0.01
L	L, Q	L	L
7	4	8	3
	750 [†] 755 757 772 778 0.03 L	g l 750 [†] 200 755 194 757 194 772 202 778 206 Signific 0.03 0.09 L L, Q	g kg ⁻¹ 750 [†] 200 429 755 194 431 757 194 423 772 202 402 778 206 389 Significance (<i>P</i>) 0.03 0.09 <0.01 L L, Q L

[†]Each value is the mean of five samples.

[‡]L, linear; Q, quadratic; C, cubic.

fed to goats (oven-dry basis).									
Time	DMI [†]		paren gestio		Apparent digestible intake [†]				
		DM	CP	NDF	DM	CP	NDF		
h				g kg-	1				
0700	26.2 [‡]	664	795	559	17.4	4.4	5.8		
1000	26.3	662	799	556	17.4	4.4	6.0		
1300	26.0	660	798	526	17.2	4.3	5.5		
1600	28.6	666	796	544	19.1	4.7	6.2		
1900	26.9	674	801	557	18.1	4.5	5.7		
	Significance (P)								
Time	0.54	0.37	0.95	0.53	0.39	0.79	0.40		
Trend§ (P < 0.10)	NS	NS	NS	NS	NS	NS	LOF		
SE	0.26	8	7	19	0.18	0.05	0.06		

Table 7. Mean daily dry matter intake (DMI), apparent diges-

tion, and apparent digestible intake of dry matter (DM), crude

protein (CP), and neutral detergent fiber (NDF) of alfalfa hay

[†]Intake expressed on a body-weight basis.

[‡]Each value is the mean of five animals.

 $^{\$}\text{NS},$ not significant; LOF, lack of fit.

Table 8. Mean in vitro true dry matter disappearance (IVTD), crude protein (CP), neutral detergent fiber (NDF), and total nonstructural carbohydrates (TNC) concentration of alfalfa hay fed to goats (oven-dry basis).

Time	IVTD	CP	NDF	TNC
h		g k	(g ⁻¹	
0700	759 ⁺	204	419	87
1000	753	201	424	81
1300	761	202	416	91
1600	766	202	404	94
1900	774	203	398	95
	Signific	ance (P)		
Time	0.05	0.97	0.04	<0.01
Trend [‡] ($P \le 0.05$)	L	NS	L	L, C
SE	5	4	7	3

⁺ Each value is the mean of five samples.

[‡]L, linear; NS, not significant; C, cubic.

in DMI with time of cut (i.e., 0700, 1000, 1300, 1600, and 1900 h), giving respective means of 27.5, 27.5, 27.7, 30.8, and 29.9 g kg⁻¹ body weight, respectively. Means for DMD followed a cubic response (P = 0.01), with respective values of 658, 648, 647, 664, and 651 g kg⁻¹. The digestible DMI showed a linear increase (P = 0.04), as noted for DMI, having respective values of 18.1, 17.9, 18.0, 20.5, and 19.5 g kg⁻¹ body weight.

Dry matter intake, as opposed to DMD, was the major animal response altered by time of cut in these experiments. This is indicated by the similar response reported for both DMI and digestible DMI. This is further supported by the relationship (*r*) among DMI, DMD, and digestible DMI and the nutritive value estimate of the "as fed" hay (Table 9). Dry matter intake was not correlated with DMD but showed significant correlations with digestible DMI and forage IVTD, TNC, and NDF. On the other hand, DMD was not correlated with any of these, whereas digestible DMI showed similar correlations to those noted for DMI.

CONCLUSIONS

The data indicate that ruminants can detect subtle shifts in plant composition and that no animal response advantage was noted by delaying the afternoon cut beyond 1600 h. Furthermore, on good drying days, cutting at 1600 vs. 1900 h may permit sufficient drying of tissue to reduce overnight respiration, which is associated with TNC losses and reduced nutritive value.

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Table 9. Correlation (r) among dry matter intake (DMI), apparent dry matter digestion (DMD), and apparent digestible dry matter intake (DDMI), and hay nutritive value.[†]

Parameter	DMD	DDMI	IVTD	TNC	CP	NDF
DMI	0.06	0.97	0.57	0.48	0.21	-0.46
	(0.85)‡	(<0.01)	(0.03)	(0.07)	(0.46)	(0.08)
DMD		0.32	-0.10	-0.05	-0.25	0.33
	_	(0.25)	(0.71)	(0.86)	(0.37)	(0.23)
DDMI			0.51	0.45	0.13	-0.35
		_	(0.05)	(0.10)	(0.65)	(0.20)

[†]IVTD, in vitro true dry matter disappearance; TNC, total nonstructural carbohydrates; CP, crude protein; NDF, neutral detergent fiber.

[‡]Probability of *r* and based on five treatments evaluated by steers, sheep, and goats (n = 15).

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