

Influence of rumen protein degradability and supplementation frequency on performance and nitrogen use in ruminants consuming low-quality forage: Cow performance and efficiency of nitrogen use in wethers^{1,2}

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ABSTRACT: Two studies were conducted to determine the influence of CP degradability and supplementation frequency (SF) on ruminant performance and N efficiency. Treatments included an unsupplemented control (CON) and degradable intake protein (DIP; 82% of CP) or undegradable intake protein (UIP; 60% of CP) provided daily, every 3 d, or every 6 d. Seven wethers (36 ± 1 kg BW) were used in the digestibility study with DIP and UIP treatments formulated to meet CP requirements. Eighty-four Angus × Hereford cows (512 ± 42 kg BW) in the last third of gestation were used for the performance study. The DIP treatments were calculated to provide 100% of the DIP requirement and UIP treatments were provided on an isonitrogenous basis compared with DIP. Basal diets consisted of low-quality (5% CP) meadow hay. Forage DMI and N intake by lambs decreased ($P < 0.05$) linearly as SF decreased.

Additionally, DMI, OM intake, N retention, N digestibility, and digested N retained were greater ($P < 0.01$) for supplemented wethers than for controls with no difference due to crude protein degradability. Nitrogen balance, DMI, and OM intake decreased linearly ($P < 0.05$) as SF decreased. Plasma urea (PU; mM) was measured over a 6-d period and supplemented lambs had increased ($P < 0.01$) PU compared with CON. Plasma urea linearly decreased ($P < 0.01$) as SF decreased. Pre- and postcalving (within 14 d and 24 h of calving, respectively) cow weight and body condition score change were more positive ($P < 0.05$) for supplemented groups than for controls. Results suggest CP supplements consisting of 20 to 60% UIP can be effectively used by ruminants consuming low-quality forage without adversely affecting N efficiency and animal performance, even when provided as infrequently as once every 6 d.

Key Words: Degradability, Frequency, Nitrogen, Protein, Ruminants, Supplementation

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Introduction

Crude protein can be divided into degradable intake protein (DIP) and undegradable intake protein (UIP). Degradable intake protein is broken down within the rumen by ruminal microorganisms, and UIP is presented to the small intestine for enzymatic digestion and potential absorption (NRC, 1985b; NRC, 1996). The use of low-quality forage by ruminants is dependent on the cellulolytic activity of ruminal microorganisms. Many of

the nutrients required for growth of these microorganisms are derived from ruminal degradation of DIP (Allison, 1969; Russell and Hespell, 1981). The resulting microbial protein is the main source of protein flowing to the small intestine of ruminants consuming low-quality forage. Consequently, DIP is generally considered to be the most beneficial supplement to low-quality forages.

Ammonia is probably the most important source of N for growth of ruminal bacteria (Allison, 1969; Tillman and Sidhu, 1969). However, infrequent supplementation with DIP can supply ammonia in excess of the immediate demands of the rumen microbial population, especially on the day of supplementation. This excess ammonia is transported to the liver and converted to urea N that is excreted in the urine or recycled back to the rumen (Leng and Nolan, 1984). Undegradable intake protein can be absorbed from the small intestine as free amino acids and peptides. These amino acids and peptides are used directly by the animal or deaminated by the liver to urea N. Therefore, UIP may be better suited to less frequent supplementation because of its delayed degradation

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Table 1. Ingredient and nutrient content of meadow hay and supplements

Item	Lamb study			Cow study		
	Meadow hay	DIP supplement	UIP supplement	Meadow hay	DIP supplement	UIP supplement
Supplement composition						
Soybean meal, % DM	—	97.5	—	—	100.0	—
SoyPLUS, % DM ^a	—	—	67.7	—	—	69.4
Blood meal, % DM	—	—	29.8	—	—	30.6
Molasses, % DM	—	2.5	2.5	—	—	—
Nutrient composition						
CP, % DM	5.2	52.8	59.8	5.0	54.8	62.8
UIP, % CP ^b	19.0	17.6	59.9	19.0	17.6	59.9
OM, % DM	91.6	92.6	94.4	91.8	92.5	94.8
NDF, % DM	60.1	11.5	28.6	57.7	8.5	25.7
ADF, % DM	32.0	5.1	6.6	32.1	3.6	5.1

^aSoyPLUS is an expeller-processed soybean meal from West Central Soy, Ralston, IA.

^bUndegradable intake protein. Estimates are based on in situ degradabilities. Techniques were similar to those described by Mass et al. (1999) and Bohnert et al. (1998) for meadow hay and supplements, respectively.

compared with DIP. This could result in increased N recycling to the gut (due to lower ruminal ammonia levels) and decreased urinary N excretion. This study was designed to determine whether infrequent supplementation of low-quality forage with UIP would allow for acceptable performance and more efficient use of dietary N by ruminants compared with DIP.

Materials and Methods

Digestion Study

Seven wethers (36 ± 1 kg) were used in an incomplete 7×4 Latin square design (Cochran and Cox, 1957) to evaluate the efficiency of N use in lambs supplemented with a DIP or UIP supplement (18 and 60% UIP as a percentage of CP, respectively; Table 1). Estimates of UIP and DIP were determined based on in situ degradability using techniques similar to those described by Mass et al. (1999) and Bohnert et al. (1998) for meadow hay and supplements, respectively. Wethers were randomly allotted to treatments and housed in individual metabolism crates within an enclosed barn with continuous lighting. The experimental protocol was approved by the Institutional Animal Care and Use Committee at Oregon State University.

Wethers had continuous access to fresh water and chopped (4 to 8 cm length) low-quality meadow hay (Table 1). Low-quality meadow hay was harvested from native flood meadows consisting of approximately 82% meadow foxtail (*Alopecurus pratensis* L.) with the majority of the remaining vegetation consisting of rushes (*Juncus* spp.), sedges (*Carex* spp.), and blue wild rye (*Elymus triticoides* Buckl.; Wenick, 2000). Hay was provided daily in two equal portions (0730 and 1730) at 120% of the average intake for the previous 5 d, with feed refusals from the previous day determined before the 0730 feeding. A trace mineral salt mix was available free choice (2.4% Ca, 2.3% P, 20.4% Na, 31.65% Cl, 0.2%

K, 0.4% mg, 0.1% S, 1,309 ppm Mn, 2,046 ppm Fe, 7 ppm Cu, 1,930 ppm Zn, 42 ppm Co, 120 ppm I, 16 ppm Se, 1,325 IU/kg vitamin E, and 552 and 50 kIU/kg vitamins A and D, respectively). In addition, an intramuscular injection of vitamins A, D, and E (200,000, 20,000, and 600 IU of vitamins A, D, and E, respectively; Vitamin E-AD 300; AgriLabs, St. Joseph, MO) was administered to each lamb at the onset of the trial to safeguard against deficiency. Treatments were arranged as a 2×3 factorial, two levels of ruminal protein degradability and three supplementation frequencies (**SF**), with a negative control (**CON**; no supplementation). Protein supplements were offered every day (**D**), every third day (**3D**), or every sixth day (**6D**) immediately prior to the 0730 feeding. The wethers on the DIP and UIP treatments received the same amount of total supplemental N over a 6-d period; therefore, the 3D and 6D treatments received threefold and sixfold the amount of supplement (N basis) on their respective supplementation d compared with D treatments. The amount of CP supplied by each supplement was approximately 0.19% of BW/day (averaged over a 6-d period) based on intake and protein requirements (NRC, 1985a). In order to avoid bias due to different weight changes due to treatment during each period, the quantity of supplement provided in each period was based on initial BW.

Experimental periods were 24 d with at least 3 d between periods (to remove wethers from metabolism crates). Dry matter intake was determined on d 17 to 22. In addition, samples of meadow hay, protein supplements, and orts were collected for d 17 to 22 and dried at 55°C for 48 h. On d 19 to 24, total fecal and urine output was collected. Urine was composited daily by wether (10% of total; weight basis) and stored at 4°C. Sufficient 6 N HCl (150 mL) was added daily to urinals to maintain urine pH < 3. A subsample of each daily fecal sample (7.5%; weight basis) was dried at 55°C for 96 h for calculation of fecal DM. On d 19 to 24, 20 mL

of blood was collected via jugular venipuncture 4 h after the 0730 feeding using a heparinized syringe. Blood samples were immediately transferred to vacutainers (Fisher Scientific, catalog no. 0268360), placed on ice for transport to the lab, and centrifuged ($5,000 \times g$, 4°C , 15 min), and plasma was harvested and stored (-20°C). Dried samples were ground through a Wiley mill (1-mm screen). Samples of ground meadow hay and protein supplements were composited by period and daily orts were composited by lamb (within period) on an equal weight basis (20% as-fed). Ground fecal samples were composited by lamb within period. Feed, orts, and fecal samples were analyzed for DM and OM (AOAC, 1990) and NDF (Robertson and Van Soest, 1981) and ADF (Goering and Van Soest, 1970) using procedures modified for use in an Ankom 200 Fiber Analyzer (Ankom Co., Fairport, NY). Feed, orts, fecal, and urine samples were analyzed for N using a Kjeltac Auto 1030 Analyzer (Tecator AB, Höganäs, Sweden). Plasma samples were assayed for urea N using the Sigma Diagnostics Procedure 535 (Sigma Chemical Co., St. Louis, MO) and a UV/VIS spectrophotometer (Spectronic 710 Spectrophotometer, Bausch & Lomb, Rochester, NY).

Performance Study

Eighty-four pregnant (approximately 200 d) Angus \times Hereford beef cows (512 ± 42 kg BW) were stratified by age, body condition score (BCS; 1 = emaciated, 9 = obese; Herd and Spratt, 1996), and expected calving date and assigned randomly within stratification to one of seven treatments (as described in the lamb digestion study above) in a 2×3 factorial arrangement (two levels of crude protein degradability and three SF) with a negative CON (no supplementation). They were then sorted by treatment and allotted randomly to 1 of 21 pens (four cows/pen; three pens/treatment). A trace mineralized salt mix was available free choice (7.3% Ca, 7.2% P, 27.8% Na, 23.1% Cl, 1.5% K, 1.7% Mg, 0.5% S, 2,307 ppm Mn, 3,034 ppm Fe, 1,340 ppm Cu, 3,202 ppm Zn, 32 ppm Co, 78 ppm I, 85 ppm Se, 79 IU/kg vitamin E, and 397 kIU/kg vitamin A). Cows were provided ad libitum access to low-quality meadow hay (Table 1).

The DIP treatments were formulated to provide 100% of the estimated DIP requirement assuming a microbial efficiency of 11% (NRC, 1996). An equal amount (N basis) of the UIP supplement (approximately 80% of DIP requirement) was provided, thereby ensuring that the DIP and UIP treatments were providing the same amount of total supplemental N over a 6-d period. Protein supplements were offered D, 3D, or 6D at 0800 to provide approximately 0.08% of BW/day of CP (averaged over a 6-d period) until calving. The experiment began on January 19, 2000, with experimental diets fed from start date to calving (78 ± 4 d).

Cow BW and BCS were measured every 14 d until calving and within 24 h of calving. All weights were obtained following an overnight shrink (16 h). Cow BCS was judged independently by three observers. The same

technicians measured BCS throughout the experiment. In addition, calf weights were obtained within 24 h of birth. Hay and supplement samples (approximately 200 g) were collected weekly, dried at 55°C for 48 h, ground through a Wiley mill (1-mm screen), and composited by period for analysis of ADF and NDF, N, and OM as described in the digestion study.

Statistical Analysis

Digestion Study

Data were analyzed as an incomplete 7×4 Latin square (Cochran and Cox, 1957) using the GLM procedure of SAS (1996). The model included period, wether, and treatment. Because the treatment structure consisted of a 2×3 factorial plus a negative CON, orthogonal contrasts were used to partition specific treatment effects. Contrast statements included 1) CON vs protein supplementation; 2) DIP vs UIP; 3) linear effect of SF; 4) quadratic effect of SF; 5) contrast 2 \times contrast 3; and 6) contrast 2 \times contrast 4. Response variables included 1) DM and OM intake; 2) total tract digestibility of DM, OM, NDF, and N; 3) N balance; and 4) digested N retained ([daily N retention, g/kg BW/daily N digested, g/kg BW] \times 100). Plasma concentration of urea N was analyzed using the REPEATED statement with the MIXED procedure of SAS (SAS Inst. Inc., Cary, NC). The model included lamb, period, treatment, day, and treatment \times day. In addition, lamb \times period \times treatment was used to specify variation between lambs (using the RANDOM statement). Lamb \times period \times treatment was used as the SUBJECT and autoregression used as the covariance structure. The same contrasts noted above were used to partition treatment sums of squares.

Performance Study

Cow performance data were analyzed as a randomized complete block design using the GLM procedure of SAS (SAS Inst. Inc.). The model included block and treatment. The same orthogonal contrasts described in the digestion study were used to partition specific treatment effects. Response variables included 1) cow weight change; 2) cow BCS change; and 3) calf birth weight.

Results and Discussion

Digestion Study

Intake of hay DM and OM by lambs was not affected by CP supplementation or degradability, whereas total DM, OM, and N intake increased ($P < 0.01$) with supplementation (Table 2). Also, hay DM and OM and total DM, OM, N, and NDF intake decreased linearly ($P < 0.05$) as SF decreased. Therefore, total nutrient intake decreased as SF decreased; nevertheless, total nutrient

Table 2. Effect of protein degradability and supplementation frequency on lamb intake, diet digestibility, and nitrogen balance

Item	Treatment ^a										P-value ^c			
	CON	DIPD	DIP3D	DIP6D	UIPD	UIP3D	UIP6D	SEM ^b	Con vs supp.	DIP vs UIP	L SF	Q SF	L SF vs CPD	Q SF vs CPD
Daily DM Intake, g/kg BW														
Hay	22.1	22.5	21.1	20.7	24.7	23.6	20.1	0.9	0.97	0.10	0.005	0.65	0.16	0.31
Supplement ^d	0.0	3.6	3.6	3.6	3.1	3.1	3.1							
Total	22.1	26.2	24.8	24.3	27.8	26.7	23.1	0.9	0.006	0.32	0.005	0.65	0.16	0.31
Daily OM Intake, g/kg BW														
Hay	20.2	20.6	19.3	19.0	22.6	21.6	18.4	0.8	0.95	0.10	0.004	0.69	0.16	0.32
Supplement ^e	0.0	3.4	3.4	3.4	2.9	2.9	2.9	0.0						
Total	20.2	24.0	22.7	22.3	25.6	24.5	21.3	0.8	0.005	0.29	0.004	0.69	0.16	0.32
Daily NDF Intake, g/kg BW	13.0	13.9	13.1	12.7	15.6	14.9	12.9	0.6	0.24	0.03	0.008	0.71	0.24	0.42
Daily N Intake, g/kg BW	0.185	0.486	0.487	0.477	0.494	0.487	0.455	0.010	<0.001	0.60	0.04	0.33	0.16	0.69
Total Tract Digestibility, %														
DM	50.4	58.7	57.8	56.6	57.3	59.1	59.4	0.9	<0.001	0.27	0.98	0.60	0.04	0.72
OM	54.0	62.4	61.9	60.4	60.9	63.2	63.2	0.9	<0.001	0.28	0.87	0.34	0.04	0.68
NDF	45.9	53.1	53.9	49.7	53.2	57.8	57.4	1.8	<0.001	0.02	0.83	0.13	0.05	0.97
N	23.7	66.2	64.5	65.7	62.2	63.8	65.4	1.7	<0.001	0.27	0.44	0.63	0.30	0.63
Daily N excretion, g/kg BW														
Fecal	0.140	0.167	0.172	0.164	0.189	0.174	0.158	0.009	0.007	0.44	0.07	0.65	0.13	0.66
Urinary	0.054	0.217	0.227	0.219	0.208	0.220	0.228	0.008	<0.001	0.68	0.18	0.48	0.26	0.64
Daily N balance, g/kg BW	-0.009	0.101	0.088	0.094	0.098	0.094	0.068	0.008	<0.001	0.25	0.04	0.94	0.18	0.15
Daily digested N retained, % ^f	-16.3	35.3	27.7	29.6	31.2	25.8	24.0	7.6	<0.001	0.54	0.41	0.63	0.92	0.82
Plasma urea N, mM	1.30	5.22	4.39	4.32	4.88	4.31	4.36	0.22	<0.001	0.50	0.008	0.10	0.41	0.85

^aCON = control; DIPD = degradable intake protein every day; DIP3D = DIP every 3rd d; DIP6D = DIP every 6th d; UIPD = undegradable intake protein every day; UIP3D = UIP every 3rd d; UIP6D = UIP every 6th d.

^bn = 4.

^cCon vs supp. = control vs supplemented treatments; DIP vs UIP = DIP vs UIP treatments; L SF = linear effect of supplementation frequency; Q SF = quadratic effect of supplementation frequency; L SF vs CPD = interaction of the linear effect of supplementation frequency and ruminal protein degradability; Q SF vs CPD = interaction of the quadratic effect of supplementation frequency and ruminal protein degradability.

^dDIPD received 3.6 g/kg BW daily; DIP3D received 10.8 g/kg BW every 3rd d; DIP6D received 21.6 g/kg BW every 6th d; UIPD received 3.1 g/kg BW daily; UIP3D received 9.3 g/kg BW every 3rd d; UIP6D received 18.6 g/kg BW every 6th d.

^eDIPD received 3.4 g/kg BW daily; DIP3D received 10.2 g/kg BW every 3rd d; DIP6D received 20.4 g/kg BW every 6th d; UIPD received 2.9 g/kg BW daily; UIP3D received 8.7 g/kg BW every 3rd d; UIP6D received 17.4 g/kg BW every 6th d.

^fCalculated as (Daily N retention, g/kg BW/Daily N digested, g/kg BW) × 100.

intake was greater for supplemented lambs than for CON.

The lack of an increase in forage intake due to protein supplementation is in contrast to other studies in which protein supplementation increased intake of low-quality forage (DelCurto et al., 1990; Köster et al., 1996; Bandyk et al., 2001). A possible explanation for this discrepancy is differences in NDF intake. It has been suggested that dry matter intake is maximized when NDF intake is approximately $12.5 \text{ g}\cdot\text{kg BW}^{-1}\cdot\text{d}^{-1}$ (Mertens, 1985, 1994). In the current study, NDF intake by unsupplemented lambs was $13.0 \text{ g}\cdot\text{kg BW}^{-1}\cdot\text{d}^{-1}$ with a range of 12.7 to 15.6 for supplemented lambs (Table 2). This agrees with results reported by Ferrell et al. (1999) in which NDF intake by lambs consuming low-quality forage averaged $13.0 \text{ g}\cdot\text{kg BW}^{-1}\cdot\text{d}^{-1}$ and did not increase due to protein supplementation. In contrast, NDF intake in unsupplemented controls was approximately 6.4, 5.1, and 8.2 and increased with protein supplementation to a maximum of 14.3, 11.3, and $13.3 \text{ g}\cdot\text{kg BW}^{-1}\cdot\text{d}^{-1}$ in the studies of DelCurto et al. (1990), Köster et al. (1996), and Bandyk et al. (2001), respectively. This agrees with the suggestion of Ferrell et al. (1999) that an intake response to protein supplementation can be expected if intake of NDF from low-quality forage is low (below approximately $12.5 \text{ g}\cdot\text{kg BW}^{-1}\cdot\text{d}^{-1}$) but not likely if NDF intake is high (above approximately $12.5 \text{ g}\cdot\text{kg BW}^{-1}\cdot\text{d}^{-1}$). Therefore, based on NDF intake in the current study, protein supplementation was not expected to increase forage intake.

Beaty et al. (1994) supplemented steers consuming wheat straw daily or three times weekly. They observed steers supplemented three times a week consumed 17% less straw DM and 12% less total DM compared with daily supplemented steers. As SF decreased in the current study from D to 6D, DIP- and UIP-supplemented lambs had an 8 and 19% decrease in forage and 7 and 17% decrease in total DM intake, respectively. This decrease in forage and total intake can be partially explained by the depression in forage intake observed for the 2 d following supplementation with the DIP and UIP 6D lambs (data not shown). Daily forage intake increased from 18.5 to 21.6 (17%) and 18.4 to 21.9 g/kg BW (19%) from the day of to the fifth day following supplementation for DIP and UIP 6D lambs, respectively. However, total intake decreased from 40.2 to 21.6 (46%) and 36.8 to 21.9 g/kg BW (40%) from the day of to the fifth day following supplementation for DIP and UIP 6D lambs. In contrast, Huston et al. (1999a) reported no difference in hay and total DM intake for ewes consuming wheat straw and supplemented with cottonseed meal daily or once every 7 d. They also noted an increase in forage and total intake due to protein supplementation with no difference between ewes receiving supplement daily or every 7 d. Similarly, Krehbiel et al. (1998) reported increased total intake for ewes consuming bromegrass hay and supplemented with soybean meal every 24 or 72 h compared with unsupplemented controls. In addition, they reported no

difference in total intake between 24- and 72-h supplemented ewes.

Apparent total tract digestibility of DM, OM, NDF, and N was increased ($P < 0.01$) with CP supplementation, whereas UIP increased ($P < 0.05$) digestibility of NDF compared with DIP (Table 2). Also, a linear effect of SF \times crude protein degradability interaction was noted with DM and OM digestibility ($P < 0.05$). This was the result of a decrease in DM and OM digestibility as SF decreased with DIP compared with an increase in digestibility with UIP.

If we assume supplement DM digestibility in the current study was 80%, estimated apparent forage digestibility for each treatment was 50.4, 55.6, 54.3, 52.5, 54.5, 56.4, and 55.9% for CON, DIP D, DIP 3D, DIP 6D, UIP D, UIP 3D, and UIP 6D, respectively. These estimations suggest that forage digestibility was increased with protein supplementation, regardless of SF or crude protein degradability. Increased DM digestibility with protein supplementation of low-quality forage has been reported in previous experiments (DelCurto et al., 1990; Beaty et al., 1994; Bandyk et al., 2001). This is most likely a result of improved N availability for the ruminal microflora (Petersen, 1987). It is not readily apparent why, as SF decreased, DM and OM digestibility decreased with DIP and increased with UIP. This could be a consequence of the large amount of DIP supplement provided on the 3D ($10.8 \text{ g}\cdot\text{kg BW}^{-1}\cdot\text{supplementation event}^{-1}$) and 6D ($21.6 \text{ g}\cdot\text{kg BW}^{-1}\cdot\text{supplementation event}^{-1}$) treatments, which may have altered ruminal fermentation and decreased ruminal digestibility. In contrast, the UIP treatments provided approximately 50% of the DIP provided by the DIP treatments, potentially minimizing the negative effects on ruminal fermentation. This could also help explain the observed increase in NDF digestibility with UIP compared with DIP.

Apparent total tract N digestibility for supplemented lambs was approximately 170% greater than the CON. The low N digestibility observed with CON (23.7%) is probably a result of the high fiber and low CP of the forage used in the current study (Table 1). This should result in a significant proportion of the N in the feces being metabolic fecal N. Ferrell et al. (1999) calculated metabolic fecal N, assuming metabolic fecal N as 4.8 g/kg DMI, from lambs consuming low-quality forage (4.3% CP, 74% NDF). They estimated that 90 to 105% of observed fecal N loss was attributed to metabolic fecal N; therefore, they suggested that caution should be used when trying to interpret apparent N digestibilities when ruminants are consuming low-quality forage. If we use 4.8 g metabolic fecal N/kg DMI to calculate metabolic fecal N, we estimate that 69 to 76% of fecal N in the current study was from metabolic fecal N.

Daily fecal and urinary excretion of N was increased ($P < 0.01$) with CP supplementation (Table 2); however, no difference was noted due to crude protein degradability or SF. Daily N balance and digested N retained were

greater with CP supplementation. In addition, daily N balance decreased linearly ($P < 0.05$) as SF decreased.

Coleman and Wyatt (1982) supplemented steers consuming low-quality forage with cottonseed meal every day, every 2 d, or every 3 d and measured efficiency of N use. In contrast to our results, they reported no difference in daily fecal N excretion compared with the control. However, they did note an increase in daily urinary N excretion with protein supplementation but no difference because of SF. Similarly, Brown et al. (1996) supplemented lambs consuming low-quality forage with soybean meal every day, every 2 d, or every 3 d and reported no difference in urinary N excretion or N retention (as a percentage of intake) for supplemented lambs.

The linear decrease in N balance as SF decreased (7 and 31% decrease from D to 6D for DIP and UIP, respectively) indicates that N retention was decreasing. However, N balance remained greater for DIP 6D (0.094 g/kg BW) and UIP 6D (0.068 g/kg BW) supplemented lambs compared with CON (-0.009 g/kg BW). A possible explanation for the linear decrease in N balance is the corresponding linear decrease in N intake that occurred as SF decreased. This is supported by the fact that there was no difference in digested N retained by supplemented lambs, suggesting similar N efficiency between treatments. These results imply that ruminants consuming low-quality forage are capable of efficiently conserving N when supplemented with protein as infrequently as once every 6 d. In addition, our data indicate that crude protein degradability, in the range of 18 to 60% UIP, has little to no effect on efficiency of N use in lambs consuming low-quality forage.

Treatment \times time interactions ($P < 0.01$) were observed for plasma urea N. However, after considering the nature of the interactions, we concluded that discussing treatment means while providing the treatment \times time figure would aid in interpretation and discussion of the data (Figure 1). Lamb plasma urea N was greater ($P < 0.01$) in CP-supplemented lambs than in CON (Table 2). No difference was observed due to crude protein degradability; however, plasma urea N decreased ($P < 0.01$) as SF decreased.

Plasma urea concentration is positively correlated with N intake (Harmeyer and Martens, 1980). This is consistent with the lack of a difference in plasma urea N concentration between DIP- and UIP-supplemented lambs. In addition, the linear decrease in N intake as SF decreased can at least partially explain the decrease in plasma urea concentration as SF decreased. Figure 1 provides an illustration of plasma urea N over the 6-d supplementation period. It is of interest to note the peaks in urea N on the day following supplementation for the 3D and 6D treatments. Plasma urea N concentration demonstrated a bimodal pattern in the 3D-supplemented lambs (a moderate peak following each supplementation), whereas a large, single peak in urea N was observed on the day following supplementation with the 6D treatment. This agrees with the work of

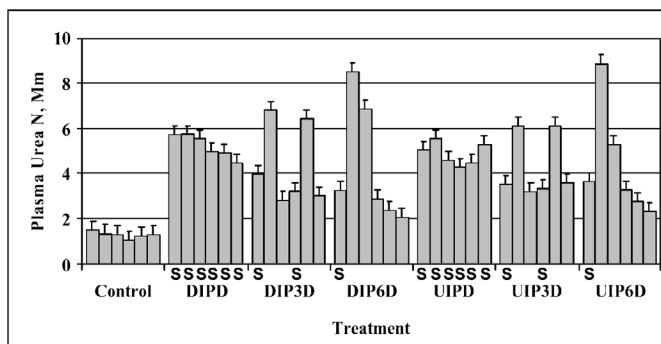


Figure 1. Effect of protein degradability and supplementation frequency on plasma urea N (mM) of lambs. Columns from left to right for each treatment represent d 1, 2, 3, 4, 5, and 6 of a 6-d supplementation period, respectively. Treatments were as follows: Control; DIPD = degradable intake protein every day; DIP3D = DIP every 3rd d; DIP6D = DIP every 6th d; UIPD = undegradable intake protein every day; UIP3D = UIP every 3rd d; UIP6D = UIP every 6th d. Each column with an S below represents a supplementation day. Treatment \times day interaction ($P < 0.0001$). SEM = 0.38.

Huston et al. (1999a) in which ewes consuming low-quality forage were supplemented with cottonseed meal every day, every 2 d, or every 7 d. They noted that serum urea N concentration increased by 57 and 170% the day after a supplementation event for ewes receiving supplement every 2 d and 7 d, respectively. Also, Krehbiel et al. (1998) reported that ewes supplemented with soybean meal every 3 d increased arterial urea N concentration by 100% the day following supplementation. In the current study, plasma urea concentration the day after a supplementation event increased by 72 and 74% for DIP 3D and UIP 3D and by 160 and 144% for DIP 6D and UIP 6D, respectively.

In their reviews of urea metabolism in ruminants, Harmeyer and Martens (1980) and Kennedy and Milligan (1980) indicated that dietary changes, such as restricted feeding and/or consumption of low-protein diets, can alter the permeability of the gastrointestinal tract to urea and change regulation of renal urea excretion. Furthermore, they suggest that the proportion of urea N produced by the liver that enters the gastrointestinal tract can vary from 10 to 95% and is negatively correlated with N intake. The quantity of urea that is excreted by the kidneys is most likely influenced by three factors: 1) changes in plasma urea concentration and the corresponding changes in filtered urea loads, 2) changes in glomerular filtration rates, and 3) changes in tubular resorption of urea (Harmeyer and Martens, 1980).

Krehbiel et al. (1998) reported that ewes consuming low-quality forage and supplemented with soybean meal every 3 d had increased urea N removal by the portal drained viscera on days between supplementation events compared with the day of supplementation.

Net removal of urea N by the portal-drained viscera was 12% of N intake on the day of supplementation compared with 74% on the 2nd d following supplementation. This suggests that the portal-drained viscera of infrequently supplemented ruminants consuming low-quality forage can increase its ability to remove urea N from the blood between supplementation events and, thereby, help sustain elevated ruminal ammonia concentrations. Also, the study of Krehbiel et al. (1998) supports the concept that the permeability of the gastrointestinal tract to urea can be altered by dietary modification, specifically supplementation frequency. It is also possible that changes in renal regulation may have improved the ability of lambs to conserve N by decreasing urinary excretion of N, thereby maintaining elevated plasma urea levels and helping to sustain N balance between supplementation events. Schmidt-Nielson et al. (1957) reported that 40% of urea N filtered by the glomeruli in camels consuming a maintenance diet was excreted in the urine compared with 1 to 2% when a N-deficient diet was fed. A similar phenomenon was observed with sheep (Schmidt-Nielson et al., 1957; Schmidt-Nielson and Osaki, 1958). Schmidt-Nielson and Osaki (1958) noted that the proportion of urea N excreted by the kidney decreased from 42% of that filtered by the glomeruli in ewes consuming a 7.5% digestible CP diet to 14% for those ewes consuming a 3% digestible CP diet. This adaptation began within 24 h and seemed to stabilize within 4 d. The hay used in the current study contained approximately 5% CP and was below the maintenance requirement of the lambs (NRC, 1985a). Therefore, 3D- and 6D-supplemented lambs would have received N-deficient diets in the 2 or 5 d between supplementation events, respectively. Consequently, it is plausible that renal regulation could have played a part in the ability of infrequently supplemented lambs to maintain similar N efficiencies compared with daily supplemented individuals.

Performance Study

Precalving (within 14 d of calving) and postcalving (within 24 h of calving) weight and BCS change were more positive ($P < 0.01$) with CP supplementation (Table 3). All weight and BCS changes were positive except for postcalving weight change on the CON treatment. The CON cows lost 39 kg compared with gains of 8, 17, 3, 4, 6, and 9 kg for the DIPD, DIP3D, DIP6D, UIPD, UIP3D, and UIP6D cows, respectively. In addition, an interaction concerning the linear effect of SF \times crude protein degradability was observed for precalving weight change. Crude protein supplementation, crude protein degradability, and SF had no effect ($P > 0.05$) on calf birth date or calf birth weight.

The ability of ruminants consuming low-quality forage and supplemented infrequently with protein to maintain acceptable performance compared with daily supplemented animals has been shown (Melton and Riggs, 1964; Hunt et al., 1989; Huston et al., 1999b).

Table 3. Effect of protein degradability and supplementation frequency on cow performance and calf birth weight

Item	Treatment ^a						P-value ^c					
	CON	DIPD	DIP3D	DIP6D	UIPD	UIP3D	UIP6D	SEM ^b	CON vs supp.	DIP vs UIP	L SF vs Q SF	L SF vs Q SF vs CPD
Supplement DMI, g/d ^d	0.0	740	740	740	627	627	627					
Initial weight, kg	521	514	514	511	519	516	490					
Initial body condition score	5.06	5.00	4.98	4.96	4.91	4.91	4.90					
Weight change, kg												
Precalving ^e	2	57	50	38	42	47	41	4	<0.001	0.17	0.04	0.30
Postcalving ^f	-39	8	17	3	4	6	9	6	<0.001	0.57	0.99	0.32
Body condition score change												
Precalving ^e	0.21	0.79	0.77	0.73	0.69	0.72	0.73	0.08	<0.001	0.44	0.90	0.89
Postcalving ^f	0.12	0.65	0.56	0.50	0.63	0.59	0.65	0.09	<0.001	0.48	0.50	0.68
Calf birth date, Gregorian d	83	78	74	73	78	71	85	4	0.16	0.40	0.79	0.13
Calf birth weight, kg	39	38	40	37	41	38	39	1	0.60	0.26	0.14	0.65

^aCON = control; DIPD = degradable intake protein every day; DIP3D = DIP every 3rd d; DIP6D = DIP every 6th d; UIPD = undegradable intake protein every day; UIP3D = UIP every 3rd d; UIP6D = UIP every 6th d.

^bn = 3.

^cCon vs supp. = control vs supplemented treatments; DIP vs UIP = DIP vs UIP treatments; L SF = linear effect of supplementation frequency; Q SF = quadratic effect of supplementation frequency; L SF vs CPD = interaction of the linear effect of supplementation frequency and ruminal protein degradability; Q SF vs CPD = interaction of the quadratic effect of supplementation frequency and ruminal protein degradability.

^dDIPD received 740 g daily; DIP3D received 2,220 g every 3rd d; DIP6D received 4,440 g every 6th d; UIPD received 627 g daily; UIP3D received 1,881 g every 3rd d; UIP6D received 3,762 g every 6th d.

^eWithin 14 d of calving.

^fWithin 24 h after calving.

In a 4-yr study, Melton and Riggs (1964) supplemented cows daily, twice weekly, or three times weekly. They found that cows supplemented twice and three times weekly gained 101 and 95% of the weight of daily supplemented controls, respectively. Also, Hunt et al. (1989) supplemented beef steers consuming low-quality grass hay with cottonseed meal every 12, 24, or 48 h. They reported that steer ADG increased for all supplemented groups compared with an unsupplemented control and was greatest with steers receiving supplement once every 48 h. In a study by Huston et al. (1999b), cottonseed meal was supplemented to beef cows consuming low-quality native range in western Texas either daily, three times weekly, or once each week. Cow weight and BCS change were greater for supplemented cows than for the unsupplemented controls, with no difference because of SF. They also noted less variation in supplement intake for three times weekly (33%) and once weekly (31%) compared with daily supplemented cows. They attributed this to less competition for supplement in the infrequently supplemented groups. Therefore, infrequent supplementation should improve supplement distribution among group-fed cows.

The interaction concerning the linear effect of SF \times crude protein degradability was associated with a decrease in precalving weight change with DIP as SF decreased compared with essentially no change due to SF with UIP. This may relate to the same interaction observed for apparent total tract DM and OM digestion discussed in the digestion study. If we assume similar responses for forage and total intake between the digestion and performance studies, the quantity of digested DM and OM should have decreased as SF decreased for DIP cows. Consequently, cow weight and BCS change may have decreased due to the lower quantity of digested nutrients available as SF decreased with DIP. However, this is contradicted by postcalving weight change and pre- and postcalving BCS change in which no difference due to SF or crude protein degradability was noted. There was a tendency ($P < 0.14$) for calf birth weight to decrease as SF decreased, suggesting that fetal weight and the associated membranes and fluids could account for part of the observed response. In addition, there may have been differences in gut fill not accounted for by withholding feed and water for 16 h.

Implications

Infrequent supplementation (as infrequently as once every 6 d) of rumen degradable and undegradable intake protein to ruminants consuming low-quality forage ($< 6\%$ crude protein) results in nitrogen efficiency and animal performance that is similar to that of daily supplemented individuals. Ruminants may have the ability to conserve nitrogen over extended periods (possibly through changes in the permeability of the gastrointestinal tract to urea N and/or renal regulation of urea excretion), thereby sustaining nitrogen efficiency

between periods of supplementation. Infrequent supplementation of protein with ruminal degradability ranging from 40 to 80% is a management alternative that can help lower costs associated with supplementation without being detrimental to animal performance.

Literature Cited

- Allison, M. J. 1969. Biosynthesis of amino acids by ruminal microorganisms. *J. Anim. Sci.* 29:797–807.
- AOAC. 1990. Official Methods of Analysis. 15th ed. Association of Official Analytical Chemists, Arlington, VA.
- Bandyk, C. A., R. C. Cochran, T. A. Wickersham, E. C. Titgemeyer, C. G. Farmer, and J. J. Higgins. 2001. Effect of ruminal vs postruminal administration of degradable protein on utilization of low-quality forage by beef steers. *J. Anim. Sci.* 79:225–231.
- Beaty, J. L., R. C. Cochran, B. A. Lintzenich, E. S. Vanzant, J. L. Morrill, R. T. Brandt, Jr., and D. E. Johnson. 1994. Effect of frequency of supplementation and protein concentration in supplements on performance and digestion characteristics of beef cattle consuming low-quality forages. *J. Anim. Sci.* 72:2475–2486.
- Bohnert, D. W., B. T. Larson, M. L. Bauer, A. F. Branco, K. R. McLeod, D. L. Harmon, and G. E. Mitchell, Jr. 1998. Nutritional evaluation of poultry by-product meal as a protein source for ruminants: Effects on performance and nutrient flow and disappearance in steers. *J. Anim. Sci.* 76:2474–2484.
- Brown, D. R., F. C. Hinds, and R. M. Collins. 1996. Effect of supplementation frequency on diet digestion and nitrogen metabolism of growing lambs fed low-quality forage. *Prof. Anim. Sci.* 12:24–27.
- Cochran, W. G., and G. M. Cox. 1957. *Experimental Designs*. 2nd ed. John Wiley & Sons, New York.
- Coleman, S. W., and R. D. Wyatt. 1982. Cottonseed meal or small grains forages as protein supplements fed at different intervals. *J. Anim. Sci.* 55:11–17.
- DelCurto, T., R. C. Cochran, D. L. Harmon, A. A. Beharka, K. A. Jacques, G. Towne, and E. S. Vanzant. 1990. Supplementation of dormant tallgrass-prairie forage: I. Influence of varying supplemental protein and/or energy levels on forage utilization characteristics of beef steers in confinement. *J. Anim. Sci.* 68:515–531.
- Ferrell, C. L., K. K. Kreikemeier, and H. C. Freetly. 1999. The effect of supplemental energy, nitrogen, and protein on feed intake, digestibility, and nitrogen flux across the gut and liver in sheep fed low-quality forage. *J. Anim. Sci.* 77:3353–3364.
- 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.
- Harmeyer, J., and H. Martens. 1980. Aspects of urea metabolism in ruminants with reference to the goat. *J. Dairy Sci.* 63:1707–1728.
- Herd, D. B., and L. R. Sprott. 1996. Body condition, nutrition and reproduction of beef cows. *Texas Agric. Ext. Serv. Bull.* 1526, College Station.
- Hunt, C. W., J. F. Parkinson, R. A. Roeder, and D. G. Falk. 1989. The delivery of cottonseed meal at three different time intervals to steers fed low-quality grass hay: Effects on digestion and performance. *J. Anim. Sci.* 67:1360–1366.
- Huston, J. E., B. S. Engdahl, and K. W. Bales. 1999a. Supplemental feeding interval for adult ewes. *Sheep Goat Res. J.* 15:87–93.
- Huston, J. E., H. Lippke, T. D. A. Forbes, J. W. Holloway, and R. V. Machen. 1999b. Effects of supplemental feeding interval on adult cows in western Texas. *J. Anim. Sci.* 77:3057–3067.
- Kennedy, P. M., and L. P. Milligan. 1980. The degradation and utilization of endogenous urea in the gastrointestinal tract of ruminants: A review. *Can. J. Anim. Sci.* 60:205–221.
- Köster, H. H., R. C. Cochran, E. C. Titgemeyer, E. S. Vanzant, I. Abdelgadir, and G. St-Jean. 1996. Effect of increasing degradable intake protein on intake and digestion of low-quality, tallgrass-prairie forage by beef cows. *J. Anim. Sci.* 74:2473–2481.

- Krehbiel, C. R., C. L. Ferrell, and H. C. Freetly. 1998. Effects of frequency of supplementation on dry matter intake and net portal and hepatic flux of nutrients in mature ewes that consume low-quality forage. *J. Anim. Sci.* 76:2464–2473.
- Leng, R. A., and J. V. Nolan. 1984. Nitrogen metabolism in the rumen. *J. Dairy Sci.* 67:1072–1089.
- Mass, R. A., G. P. Lardy, R. J. Grant, and T. J. Klopfenstein. 1999. In situ neutral detergent insoluble nitrogen as a method for measuring forage protein degradability. *J. Anim. Sci.* 77:1565–1571.
- Melton, A. A., and J. K. Riggs. 1964. Frequency of feeding protein supplement to range cattle. *Texas Agric. Exp. Stn. Bull.* B-1025, College Station.
- Mertens, D. R. 1985. Factors influencing feed intake in lactating cows: From theory to application using neutral detergent fiber. In: *Proc. Georgia Nutr. Conf., Univ. of Georgia, Athens*. pp 1–18.
- Mertens, D. R. 1994. Regulation of forage intake. In: G. C. Fahey, Jr. (ed.) *Forage Quality, Evaluation, and Utilization*. pp 450–493. Am. Soc. of Agronomy, Inc., Crop Sci. Soc. of Am., Inc., and Soil Sci. Soc. of Am., Inc., Madison, WI.
- NRC. 1985a. *Nutrient Requirements of Sheep*. 6th ed. National Academy Press, Washington, DC.
- NRC. 1985b. *Ruminant Nitrogen Usage*. National Academy Press, Washington, DC.
- NRC. 1996. *Nutrient Requirements of Beef Cattle*. 7th ed. National Academy Press, Washington, DC.
- Petersen, M. K. 1987. Nitrogen supplementation of grazing livestock. In: *Proc. Grazing Livest. Nutr. Conf., Jackson, WY*. pp 115–121.
- Robertson, J. B., and P. J. Van Soest. 1981. The detergent system of analyses and its application to human foods. In: W. P. T. James and O. Theander (ed.) *The Analysis of Dietary Fiber*. pp 123–158. Marcell Dekker, New York.
- Russell, J. B., and R. B. Hespell. 1981. Microbial rumen fermentation. *J. Dairy Sci.* 64:1153–1169.
- Schmidt-Nielson, B., and H. Osaki. 1958. Renal response to changes in nitrogen metabolism in sheep. *Am. J. Physiol.* 193:657–661.
- Schmidt-Nielson, B., K. Schmidt-Nielson, T. R. Houpt, and S. A. Jarnum. 1957. Urea excretion in the camel. *Am. J. Physiol.* 188:477–484.
- Tillman, A. D., and K. S. Sidhu. 1969. Nitrogen metabolism in ruminants: Rate of ruminal ammonia production and nitrogen utilization by ruminants – A review. *J. Anim. Sci.* 28:689–697.
- Wenick, J. J. 2000. The effect of grazing interval on forage quality and production of meadow foxtail. M.S. thesis. Oregon State Univ., Corvallis.