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# Daily and alternate day supplementation of urea or soybean meal to ruminants consuming low-quality cool-season forage: I—Effects on efficiency of nitrogen use and nutrient digestion<sup>☆</sup>, <sup>☆☆</sup>, <sup>☆☆☆</sup>, <sup>★</sup>

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## ABSTRACT

Five Rambouillet × Polypay wethers ( $52 \pm 2$  kg BW; Experiment 1) and 5 Angus × Hereford steers ( $464 \pm 26$  kg BW; Experiment 2) were used in two incomplete  $5 \times 4$  Latin squares with four 18-d periods to determine the influence of supplemental N source and supplementation frequency (SF) on efficiency of N use, nutrient intake, and nutrient digestion in ruminants consuming hard fescue straw (4.7% CP). Wethers and steers were provided straw at 120% of the previous 5 d average intake in two equal portions at 0730 h and 1900 h. Treatments (TRT) included an unsupplemented control (CON) and a urea (29% CP) or soybean meal (SBM; 26% CP) supplement provided daily (D) or every-other-day (2D) at 0700 h. In Experiment 1, supplemental CP was provided at 0.10% of BW daily and 0.20% of BW every-other-day for D and 2D supplemented wethers, respectively. Feces and urine were collected on d 13–18 for calculation of N balance and blood samples were obtained 4 h post-supplementation on d 13–18 for analysis of plasma urea-N (PUN). In Experiment 2, D TRT were supplemented CP at 0.04% of BW/day while 2D TRT received 0.08% of BW every-other-day. Feces were collected on d 13–18 for estimation of nutrient digestibility. Dry matter intake, OM intake, N intake, N retention, DM, OM, and N digestibility, and digested N retained were greater ( $P < 0.01$ ) for supplemented wethers compared with CON with no differences ( $P > 0.05$ ) because of N source or SF. There were no differences in fecal or urinary N excretion because of supplementation, SF, or N source ( $P > 0.10$ ). However, PUN was increased ( $P < 0.01$ ) in supplemented lambs compared with CON, whereas urea TRT had greater ( $P < 0.01$ ) PUN compared with SBM. Plasma urea-N was also increased ( $P = 0.05$ ) for D compared with 2D TRT. Straw and total DM and OM intake were greater ( $P \leq 0.02$ ) for supplemented steers compared with CON; however, DM and OM digestibility was not influenced ( $P \geq 0.25$ ) because of supplementation or SF. These results suggest that supplements containing urea or SBM as the supplemental N source

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can be used by lambs and steers consuming cool-season, low-quality forage without adversely affecting N efficiency, nutrient intake, or nutrient digestibility, even when provided every-other-day.

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## 1. Introduction

Supplementation of natural protein to ruminants consuming low-quality forage (<6% CP) improved forage intake (Bandyk et al., 2001; DelCurto et al., 1990; Köster et al., 1996), nutrient digestibility (Bohnert et al., 2002a, 2002b; DelCurto et al., 1990), animal performance (Bohnert et al., 2002b; Clanton and Zimmerman, 1970), and reproductive efficiency (Sasser et al., 1988; Wiley et al., 1991) compared with non-supplemented controls. Due to its lower cost per unit of N compared with most sources of natural protein, urea (non-protein N; NPN) is a popular source of supplemental N. As with natural protein, providing supplemental urea to ruminants consuming low-quality forage increased forage intake (Egan, 1965; Romero et al., 1976; Tudor and Morris, 1971), nutrient digestibility (Ammerman et al., 1972; Briggs et al., 1947; Raleigh and Wallace, 1963), and animal performance (Currier et al., 2004a; Raleigh and Wallace, 1963; Redman et al., 1980) when compared with no supplemental N. Nevertheless, compared with sources of natural protein, NPN is often considered inferior as a supplemental N source for ruminants consuming low-quality forage (Clanton, 1978; Kropp et al., 1977); primarily because of its lack of amino acid N and metabolizable protein. However, when provided to mature ruminants consuming low-quality forage at a level that does not exceed the estimated requirement to maximize ruminal microbial protein synthesis (NRC, 1996), NPN and natural protein appear to be comparable as sources of supplemental N (Cooke and Arthington, 2008; Farmer et al., 2004).

Infrequent (every-other-day to once every 7 d) supplementation of CP to ruminants consuming low-quality forage can decrease the costs associated with supplementen-

tation while maintaining acceptable performance, nutrient intake, and nutrient utilization when compared with daily supplementation (Bohnert et al., 2002a, 2002b; Currier et al., 2004a, 2004b; Huston et al., 1999a, 1999b). However, infrequent supplementation of urea to ruminants is not a common management practice because of the potential for NH<sub>3</sub> toxicity and decreased supplement intake (Chalupa, 1968; Helmer and Bartley, 1971). Consequently, there is limited information available concerning infrequent supplementation of NPN to ruminants consuming low-quality forage. Therefore, we conducted two experiments to compare daily and every-other-day supplementation of urea or soybean meal (SBM) on forage intake, nutrient digestibility, and N efficiency in ruminants consuming low-quality forage.

## 2. Materials and methods

The experimental protocol was approved by the Institutional Animal Care and Use Committee at Oregon State University.

### 2.1. Experiment 1: N balance study

Five wethers ( $52 \pm 2$  kg) were used in an incomplete  $5 \times 4$  Latin square design (Cochran and Cox, 1957) to evaluate the efficiency of N use in lambs offered supplements in which urea or SBM provided the primary source of supplemental N (Table 1). Wethers were allotted randomly to treatments and housed in individual metabolism crates within an enclosed barn with continuous lighting.

Wethers had continuous access to fresh water and chopped (4- to 8-cm length) hard fescue (*Festuca*

**Table 1**  
Ingredient and nutrient content of hard fescue straw and supplements.

Item	Lamb study (Experiment 1)			Steer study (Experiment 2)		
	Hard fescue straw	Urea supplement	Soybean meal supplement	Hard fescue straw	Urea supplement	Soybean meal supplement
Supplement composition (%)						
Urea	–	5.3	–	–	5.3	–
Soybean meal	–	–	31.0	–	–	31.0
Soybean hulls	–	91.0	65.3	–	91.0	65.3
Dried molasses	–	3.7	3.7	–	3.7	3.7
Nutrient composition						
CP, % DM	4.7	29.2	26.4	4.7	29.0	26.4
DIP <sup>a</sup> , % CP	76.0	83.0	76.4	76.0	83.0	76.4
OM, % DM	93.3	94.7	93.8	93.2	94.8	94.1
NDF, % DM	76.8	58.6	45.8	78.4	59.0	45.6
ADF, % DM	40.0	40.2	30.0	40.6	40.3	29.1

<sup>a</sup> Degradable intake protein. Estimates are based on dacron bag degradabilities. Techniques were similar to those described by Mass et al. (1999) and Bohnert et al. (1998) for straw and supplements, respectively.

*trachyphylla*) straw (Table 1). Treatments consisted of an unsupplemented control and a urea or SBM supplement provided daily (D) or every-other-day (2D; CON=control, UD=urea supplement every day, U2D=urea supplement every-other-day, SBMD=SBM supplement every day, and SBM2D=SBM supplement every-other-day). Supplements were offered D or 2D at 0700 h. The urea and SBM treatments received the same amount of total supplemental N over a 2-d period; therefore, the 2D treatments received double the quantity of supplemental N on their respective supplementation day compared with D treatments. The amount of CP supplied by each supplement was approximately 0.10% of BW/d (3.5 g supplement DM/kg BW; averaged over a 2-d period). To minimize bias because of potential BW changes resulting from treatment regimes during each period, the quantity of supplement provided in each period was based on initial BW at the beginning of the experiment. Forage was provided daily at 120% of the average intake for the previous 5 d in two equal portions (0730 h and 1900 h), with feed refusals from the previous day determined before the 0700 h feeding. Also, 35 g of a trace mineral salt mix (2.4% Ca, 2.3% P, 20.4% Na, 31.65% Cl, 0.2% K, 0.4% Mg, 0.1% S, 1309 ppm Mn, 2046 ppm Fe, 7 ppm Cu, 1930 ppm Zn, 42 ppm Co, 120 ppm I, 16 ppm Se, 1325 IU/kg Vitamin E, and 552 and 50 kIU/kg Vitamins A and D, respectively) was provided daily to each lamb at 0700 h. 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.

Experimental periods were 18 d with at least 3 d between periods to allow for removal of wethers from metabolism crates. Dry matter intake was determined on d 11–16. Samples of hard fescue straw and N supplements (approximately 150 g/d) were collected on d 11–16 while orts were collected and subsampled (20% of total daily refusals; as-fed basis) on d 12–17. Samples of feed and orts were dried at 55 °C for 48 h. On d 13–18, total fecal and urine output was collected. Sufficient 6 N HCl (100 mL) was added daily to urinals to maintain urine pH < 3 to minimize bacterial growth and N loss. This was verified with pH paper during the urine collection period. Urine was composited daily by wether (50% of total daily output; weight basis) and stored at 4 °C. A sub-sample of each total daily fecal sample (7.5%; wet weight basis) was dried at 55 °C for 96 h for calculation of fecal DM and then composited by lamb within period. On d 13–18, 12 mL of blood was collected via jugular venipuncture 4 h after the 0730 h straw 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, centrifuged (5000g for 15 min; 4 °C), and plasma harvested and stored at –20 °C.

Dried samples were ground through a Wiley mill (1-mm screen). Samples of ground hard fescue straw and N supplements were composited by period and daily orts composited by lamb (within period). Feed, orts, and fecal samples were analyzed for DM, OM (AOAC, 1990), 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 Leco CN-2000 (Leco Corporation, St. Joseph, MI). Plasma samples were assayed for urea-N (PUNN) using the Sigma Diagnostics Procedure 535 (Sigma Chemical Co., St. Louis, MO) and a UV/vis spectrophotometer (Spectronic 710 Spectrophotometer, Bausch & Lomb, Inc., Rochester, NY).

## 2.2. Experiment 2: digestion study

Five Angus × Hereford steers (464 ± 26 kg) with ruminal cannulas were allotted randomly to 1 of 5 treatments in an incomplete 5 × 4 Latin square (Cochran and Cox, 1957) and housed in individual pens (4 × 8 m) within an enclosed barn with continuous lighting. Treatments were the same as described in Experiment 1. Urea treatments were formulated to provide 100% of the estimated degradable intake protein (DIP) requirement assuming a microbial efficiency of 11% (NRC, 1996; Model 1) while SBM treatments were provided on an isonitrogenous basis with urea treatments. Nitrogen supplements were placed directly into the rumen via the ruminal cannula at 0700 h for supplemented treatments. The D TRT were supplemented CP at 0.04% of BW/day (1.3 g DM/kg BW) while the 2D TRT were supplemented at 0.08% of BW (2.6 g DM/kg BW) every-other-day. As in Experiment 1, the quantity of supplement provided in each period was based on initial BW at the beginning of the experiment to minimize potential bias due to BW changes that could result from the different treatment regimens during each period. Steers had continuous access to fresh water and chopped (4- to 8-cm length) hard fescue straw. Nutrient content of the hard fescue straw and N supplements is listed in Table 1. Estimates of DIP were determined based on in situ degradability using techniques similar to those described by Bohnert et al. (1998) and Mass et al. (1999) for supplements and hard fescue straw, respectively. Straw was provided daily at 120% of the previous 5 d average intake in two equal portions (0730 h and 1900 h), with feed refusals from the previous day determined before feeding. 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, 2307 ppm Mn, 3034 ppm Fe, 1340 ppm Cu, 3202 ppm Zn, 32 ppm Co, 78 ppm I, 90 ppm Se, 79 IU/kg vitamin E, and 397 kIU/kg vitamin A). In addition, an intramuscular injection of vitamins A, D, and E (500,000, 50,000, and 1500 IU of Vitamins A, D, and E, respectively; Vitamin E-AD 300; AgriLabs; St. Joseph, MO) was administered to each steer at the onset of the trial to safeguard against deficiency.

Experimental periods were 18 d, with 9 d of diet adaptation and 9 d of sampling. Intake was measured beginning d 11 and concluded d 16 (subsamples of straw and supplements, approximately 150 g/d as-fed, were collected at this time) while orts were measured and subsampled (5% of total daily refusal; as-fed basis) on d 12–17.

On d 11 and 12, treatment effects on ruminal particulate fill were determined by manually removing reticulorumen contents 4 h after feeding. This allowed sampling

when D and 2D supplements were provided and when only D supplements were provided. Total ruminal contents were weighed, mixed by hand, and sub-sampled in triplicate (approximately 400 g/triplicate). The remaining ruminal contents were replaced immediately into the animal. Ruminal samples were weighed, dried in a forced-air oven (55 °C; 96 h), reweighed in order to calculate DM, ground to pass a 1-mm screen in a Wiley mill, and composited within period and day by steer. A more complete description of these procedures is provided in a companion paper (Cappelozza et al., 2013).

Feces were collected on d 13–18. Steers were fitted with harnesses and fecal bags on d 13 at 0730 h. Fecal bags were weighed and emptied twice daily at 0730 h and 1630 h. Feces collected at 1630 h were stored individually by steer in a sealed 190-L polyethylene bag for mixing with the 0730 h collection the following morning (24-h fecal collection). Feces were manually mixed and a 2.5% sub-sample (wet weight) obtained, dried for 96 h at 55 °C, re-weighed for DM, and composited by steer within period. Dried samples of straw, orts, and feces were ground through a Wiley mill (1-mm screen).

Ground samples of hard fescue straw and N supplements were composited by period and daily orts composited by steer (within period) on an equal weight basis. Feed, orts, and feces were analyzed for DM, OM, N, NDF, and ADF as described in Experiment 1.

### 3. Statistical analysis

For all analyses, results are reported as LS Means, significance was set at  $P \leq 0.05$  and tendencies were determined if  $P > 0.05$  and  $\leq 0.10$ . Results are reported according to treatment effects if no interactions were significant or according to highest-order interaction detected.

#### 3.1. Experiment 1: N balance study

Data were analyzed as an incomplete  $5 \times 4$  Latin square (Cochran and Cox, 1957) using the MIXED procedure of SAS (SAS Inst. Inc., Cary, NC) and Satterwaite approximation to determine the denominator degrees of freedom for the test of fixed effects. The model included treatment and period as independent variables. Wether was used as the random variable. Because the treatment structure consisted of a  $2 \times 2$  factorial plus a negative control, orthogonal contrasts were used to partition specific treatment effects. Contrast statements included (1) CON vs. supplementation; (2) urea vs. SBM; (3) D vs. 2D supplementation; and (4) N source  $\times$  SF. The model used for PUN included treatment, day, and treatment  $\times$  day interaction, and period as the independent variable. Wether was used as the random variable. The specified term for the repeated statement was day, lamb (period  $\times$  treatment) was included as the subject, and autoregressive was used as the covariance structure based on it providing the lowest Akaike information criterion. The same contrasts noted above were used to partition treatment sums of squares.

#### 3.2. Experiment 2: digestion study

Data were analyzed as an incomplete  $5 \times 4$  Latin square using the MIXED procedure of SAS and Satterwaite approximation to determine the denominator degrees of freedom for the test of fixed effects. The model included treatment and period as independent variables. Steer was used as a random variable. The same orthogonal contrasts used in Experiment 1 were used to partition specific treatment effects.

## 4. Results

#### 4.1. Experiment 1: N balance study

Straw DM and OM intake by wethers were not affected by supplementation, N source, or SF ( $P \geq 0.15$ ; Table 2). Conversely, total DM and OM intake were increased with supplementation ( $P < 0.01$ ), but not by N source or SF ( $P \geq 0.15$ ). We noted a tendency for NDF intake to increase ( $P = 0.07$ ) with supplementation, but it was not influenced by N source or SF ( $P \geq 0.14$ ). Nitrogen intake was increased ( $P < 0.01$ ) with supplementation and tended ( $P = 0.06$ ) to be greater for urea supplemented steers than for SBM steers; however, SF had no effect on N intake ( $P \geq 0.35$ ).

Apparent total tract digestibility of DM, OM, ADF, and N were increased ( $P < 0.01$ ) with supplementation and not affected ( $P \geq 0.22$ ) by SF (Table 2). However, urea supplemented wethers had lower total tract OM ( $P = 0.05$ ) digestibility and tended to have lower total tract DM ( $P = 0.08$ ) digestibility compared with SBM supplemented wethers. Total tract ADF and N digestibility were not influenced by N source ( $P \geq 0.55$ ). Apparent total tract NDF digestibility tended to be greater with supplementation ( $P = 0.06$ ) and daily supplementation ( $P = 0.10$ ), but was not influenced ( $P = 0.82$ ) by N source.

Fecal and urinary N excretion were not altered ( $P \geq 0.16$ ) by supplementation, N source, or SF (Table 2). In contrast, N balance and digested N retained were increased ( $P < 0.01$ ) with supplementation while not affected by N source or SF ( $P \geq 0.40$ ).

Treatment  $\times$  day interactions ( $P < 0.01$ ) were observed for PUN. However, after considering the nature of the interactions, we concluded that discussing treatment means while providing the treatment  $\times$  day figure (Fig. 1) would aid in interpretation and discussion of the data. Plasma urea-N was increased with supplementation ( $P < 0.01$ ), greater for urea compared with SBM ( $P < 0.01$ ), and greater for D compared with 2D ( $P = 0.05$ ; Table 2; Fig. 1).

#### 4.2. Experiment 2: digestion study

Supplemented steers had greater straw and total DM and OM intake ( $P \leq 0.02$ ) compared with CON steers and consequently, N and NDF intake also increased with supplementation ( $P < 0.01$ ). Conversely, none of these parameters were further affected by N source or SF ( $P \geq 0.28$ ; Table 3).

Apparent total tract DM and OM digestibility were not affected by supplementation ( $P \geq 0.33$ ), for urea compared

**Table 2**  
Effects of supplemental N source and supplementation frequency on intake, diet digestibility, and N balance in lambs consuming low-quality forage.

Item	Treatment <sup>a</sup>					SEM <sup>b</sup>	P-value <sup>c</sup>			
	CON	UD	U2D	SBMD	SBM2D		Con vs. Supp	Urea vs. SBM	D vs. 2D	N source × SF
Daily DM intake, g/kg BW										
Straw	23.2	23.2	21.5	22.5	22.3	1.52	0.27	0.94	0.16	0.28
Supplement <sup>d</sup>	0.0	3.5	3.5	3.5	3.5					
Total	23.2	26.7	25.0	26.0	25.8	1.52	< 0.01	0.93	0.16	0.28
Daily OM intake, g/kg BW										
Straw	21.6	21.7	20.0	21.0	20.8	1.42	0.29	0.92	0.15	0.26
Supplement <sup>e</sup>	0.0	3.3	3.3	3.3	3.3					
Total	21.6	25.0	23.3	24.3	24.1	1.42	< 0.01	0.95	0.15	0.26
Daily NDF intake, g/kg BW	17.8	19.8	18.3	18.7	18.7	1.15	0.07	0.46	0.14	0.18
Daily N intake, g/kg BW	0.176	0.330	0.327	0.321	0.314	0.0121	< 0.01	0.06	0.35	0.73
Apparent total tract digestibility, %										
DM	37.9	42.4	42.0	43.9	43.4	0.93	< 0.01	0.08	0.56	0.95
OM	40.5	45.4	44.8	47.0	46.1	0.80	< 0.01	0.05	0.28	0.85
NDF	42.7	45.9	43.8	45.9	44.3	1.02	0.06	0.82	0.10	0.79
ADF	31.7	45.3	43.9	45.4	41.3	2.09	< 0.01	0.57	0.22	0.52
N	14.9	49.3	53.1	53.0	54.8	2.38	< 0.01	0.55	0.34	0.79
Daily N excretion, g/kg BW										
Fecal	0.151	0.165	0.154	0.151	0.142	0.0112	0.81	0.16	0.25	0.88
Urinary	0.070	0.087	0.107	0.085	0.069	0.0178	0.38	0.27	0.93	0.33
Daily N balance, g/kg BW	-0.042	0.080	0.064	0.085	0.101	0.0245	< 0.01	0.40	0.99	0.52
Daily digested N retained <sup>f</sup> , %	-178.3	46.4	35.7	42.5	57.5	40.48	< 0.01	0.83	0.96	0.76
Plasma urea-N, mM	3.7	7.0	6.1	5.0	4.9	0.22	< 0.01	< 0.01	0.05	0.15

<sup>a</sup> CON=control; UD=urea supplement every day; U2D=urea supplement every-other-day; SBMD=soybean meal supplement every day; SBM2D=soybean meal supplement every-other-day.

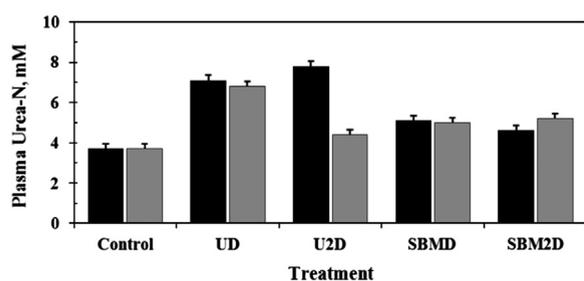
<sup>b</sup> n=4.

<sup>c</sup> Con vs. Supp=control vs. supplemented treatments; Urea vs. SBM=urea vs. soybean meal treatments; D vs. 2D=daily vs. alternate day supplementation; N Source × SF=interaction of N source vs. supplementation frequency.

<sup>d</sup> UD and SBMD received 3.47 g/kg BW daily; U2D and SBM2D received 6.94 g/kg BW every-other-day.

<sup>e</sup> UD received 3.29 g/kg BW daily; U2D received 6.58 g/kg BW every-other-day; SBMD received 3.25 g/kg BW daily; SBM2D received 6.50 g/kg BW every-other-day.

<sup>f</sup> Calculated as (Daily N retention, g/kg BW/Daily N digested, g/kg BW) × 100.



**Fig. 1.** Effects of supplemental N source and supplementation frequency (SF) on lamb plasma urea-N (mM) 4 h post-feeding. Left and right columns for each treatment represent when all supplements were offered and when only daily supplements were offered, respectively. Treatments were: control; UD=urea supplement every day; U2D=urea supplement every-other-day; SBMD=soybean meal supplement every day; SBM2D=soybean meal supplement every-other-day. Treatment × SF interaction is ( $P < 0.01$ ). SEM for treatment × SF is 0.25.

with SBM supplementation ( $P \geq 0.51$ ), or for D compared with 2D supplementation ( $P \geq 0.25$ ; Table 3). In contrast, apparent total tract N digestibility was increased ( $P < 0.01$ ) with supplementation and not affected ( $P \geq 0.19$ ) by N

source or SF. Total tract NDF digestibility was not influenced by supplementation, N source, or SF ( $P \geq 0.11$ ).

## 5. Discussion

The primary goal of the present study was to investigate the effects of supplementation frequency (daily or every-other-day) and supplemental N source (SBM or urea) on forage intake, nutrient digestibility and N efficiency in ruminants consuming low-quality cool-season forage (< 5% CP; DM basis). Infrequent CP supplementation has been shown to be a viable option to reduce costs associated with supplementation (such as labor, fuel and feed) while maintaining adequate performance in cattle (Bohnert et al., 2002a, 2002b; Currier et al., 2004a, 2004b; Huston et al., 1999a, 1999b).

In agreement with previous research (DelCurto et al., 1990; Köster et al., 1996), straw DM and OM intake were increased by supplementation in steers (Experiment 2); however, N source and SF had no effect. The observed increase in forage intake with supplementation suggests that ruminal N may have been limiting in unsupplemented steers. In a companion paper, Cappellozza et al. (2013)

**Table 3**

Effects of supplemental N source and supplementation frequency on nutrient intake and disappearance in steers.

Item	Treatment <sup>a</sup>						P-value <sup>c</sup>			
	CON	UD	U2D	SBMD	SBM2D	SEM <sup>b</sup>	Con vs. Supp	Urea vs. SBM	D vs. 2D	N source × SF
Daily DM intake, g/kg BW										
Straw	15.7	17.1	17.3	17.6	17.0	0.88	0.02	0.90	0.68	0.37
Supplement <sup>d</sup>	0.0	1.3	1.3	1.3	1.3					
Total	15.7	18.4	18.6	18.9	18.3	0.88	< 0.01	0.90	0.68	0.37
Daily OM intake, g/kg BW										
Straw	14.6	16.0	16.2	16.4	15.9	0.83	0.02	0.89	0.66	0.38
Supplement <sup>e</sup>	0.0	1.2	1.2	1.2	1.2					
Total	14.6	17.2	17.4	17.6	17.1	0.83	0.02	0.89	0.66	0.38
Daily NDF intake, g/kg BW	12.4	14.2	14.4	14.4	13.9	0.71	< 0.01	0.79	0.64	0.37
Daily N intake, g/kg BW	0.12	0.19	0.19	0.19	0.18	0.006	< 0.01	0.28	0.88	0.59
Apparent total-tract digestibility, %										
DM	51.3	51.7	54.2	52.3	52.2	1.3	0.33	0.51	0.31	0.26
OM	53.4	53.7	56.2	54.3	54.4	1.3	0.34	0.61	0.25	0.29
NDF	56.1	55.5	57.7	54.8	56.4	1.3	0.99	0.37	0.11	0.73
N	28.5	43.5	48.9	44.7	42.9	1.7	< 0.01	0.19	0.31	0.06

<sup>a</sup> CON=control; UD=urea supplement every day; U2D=urea supplement every-other-day; SBMD=soybean meal supplement every day; SBM2D=soybean meal supplement every-other-day.

<sup>b</sup> n=4.

<sup>c</sup> Con vs Supp=control vs. supplemented treatments; Urea vs. SBM=urea vs. soybean meal treatments; D vs. 2D=daily vs. alternate day supplementation; N source × SF=interaction of N source vs. supplementation frequency.

<sup>d</sup> UD and SBMD received 1.30 g/kg BW daily; U2D and SBM2D received 2.60 g/kg BW every-other-day.

<sup>e</sup> UD received 1.23 g/kg BW daily; U2D received 2.46 g/kg BW every-other-day; SBMD received 1.22 g/kg BW daily; SBM2D received 2.44 g/kg BW every-other-day.

reported average ruminal NH<sub>3</sub>-N concentrations of 1.32 mM in unsupplemented steers. This concentration is near the lower end of the levels shown to be needed for maximal growth of ruminal microbes in vivo (1.18–2.94 mM; Slyter et al., 1979). Also, ruminal IADF passage rate was increased with supplementation (Cappellozza et al., 2013). Therefore increased ruminally available N may have resulted in increased microbial growth and the subsequent increase in straw intake.

Conversely, straw DM and OM intake in lambs were not influenced by supplementation, N source or SF. These findings are in agreement with results from Currier et al. (2004a), who reported no effects of N supplementation on straw DM and OM intake in a similarly designed study. It has been suggested that DMI is maximized and will not respond to supplementation when daily NDF intake is roughly ≥12.5 g/kg BW (Mertens, 1994). In the current study, lamb daily NDF intake was 17.8 g/kg BW for the CON group and ranged from 18.3 to 19.8 g/kg BW in supplemented lambs. Thus, forage intake was not expected to increase. This coincides with other work related to a lack of forage intake following N supplementation of low-quality cool-season forages (Bohnert et al., 2002a, 2002b; Currier et al., 2004a, 2004b; Schauer et al., 2010). It should be noted that NDF intake increased in Experiments 1 and 2, likely a result of NDF provided by the supplements (which ranged from 46% to 59%; Table 1). Also, the lack of a supplementation effect on straw intake in Experiment 1 may have resulted from adequate protein intake without supplementation. Preston et al. (1965) reported that a PUN in excess of 3.57 mM indicates adequate protein intake for lambs consuming high-fiber based diets. In the current

study, CON lambs had a PUN concentration of 3.7 mM suggesting that protein intake may have been adequate prior to supplementation. Conversely, the negative N balance and digested N retained with CON along with improvement in N balance and digested N retained with supplementation suggest that lambs were deficient in N prior to supplementation.

Protein supplementation has been shown to increase total tract digestibility in ruminants consuming low-quality forage (Bohnert et al., 2002a, 2002b; Currier et al., 2004c; Wickersham et al., 2008b). As expected, supplementation increased DM, OM, ADF and N digestibility, and tended to increase NDF digestibility, in lambs. Conversely, N supplementation, regardless of source, failed to increase DM, OM, and NDF digestibility in steers. Similarly, Wickersham et al. (2008b) reported that increasing amounts of DIP (casein) had no effect on total tract digestibility of OM and NDF in steers. The authors suggest that the lack of a supplementation effect may have been caused by an increased intake and passage rate in response to increased ruminally available N supply (Guthrie and Wagner, 1988; Olson et al., 1999). This agrees with our observation of increased straw intake as well as ruminal IADF passage rate in supplemented steers from Experiment 2 reported by Cappellozza et al. (2013). As a result, the length of time during which ruminal microbes had access to the substrate was reduced, thereby, potentially masking the positive effects on digestibility often observed due to supplementation of low-quality forage.

Apparent total-tract N digestibility increased significantly with supplementation in both experiments. This is in agreement with other results observed with ruminants

consuming low-quality forage and supplemented with CP (Bohnert et al., 2002a, 2002b; Currier et al., 2004a; Ferrell et al., 1999). The low N digestibility observed in the CON groups (28.5% in steers and 14.9% in lambs) is probably a result of the high fiber and low CP of the forage. Additionally, metabolic fecal N, the portion of fecal N that is not directly from the diet (Strozinski and Chandler, 1972), can constitute a significant portion of total fecal N in unsupplemented ruminants, resulting in low apparent N digestibility (Ferrell et al., 1999) due to the relatively consistent quantity of metabolic fecal N (5.35 g N/kg DMI; NRC 1985) and low N intake in ruminants consuming low-quality forages (Currier et al., 2004a). The greater N digestibility with supplementation is likely due to the greater digestibility of supplements compared to low-quality forage and decreased metabolic fecal N as a percentage of N intake (Currier et al., 2004a; Wickersham et al., 2008a).

The increased N balance and digested N retained observed with supplementation agrees with previous research in which supplemental N was provided to ruminants consuming low-quality forage (Bohnert et al., 2002b; Currier et al., 2004a; Egan, 1965). The lack of a N source effect agrees with previous work by Ammerman et al. (1972), who reported no differences in N retention or balance between lambs supplemented with either SBM or an NPN source. Also, infrequent CP supplementation of ruminants consuming low-quality forages has resulted in similar N balance and digested N retained compared with daily supplementation (Bohnert et al., 2002b; Currier et al., 2004a; Romero et al., 1976). Supporting our results, Romero et al. (1976) supplemented steers consuming 2% CP forage with an oral drench of urea twice daily, once daily or once every 2 d. Nitrogen balance was  $-5.0$  g/d for unsupplemented steers and increased to 7.0, 5.7 and 5.7 g/d for twice daily, once daily and once every 2 d, respectively. Supplementation frequency did not affect N balance. Similarly, Bohnert et al. (2002b) supplemented wethers consuming 5.2% CP meadow hay with low- or high-DIP supplements daily, once every 3 d, or once every 6 d and increased N balance compared to an unsupplemented control. However, the authors reported a linear decrease in N balance as SF decreased, which they attributed to a similar decrease in N intake as SF decreased which was supported by no effect of SF on digested N retained. These findings suggest that infrequent N supplementation to ruminants consuming low-quality forage has minimal effects on N balance and digested N retained compared with daily supplementation.

Plasma urea concentration has been shown to be positively correlated with N intake (Harmeyer and Martens, 1980). Likewise, we observed increases of 83% and 55% in N intake and PUN, respectively, with supplementation compared with the CON. Other research has shown increased PUN with CP supplementation of low-quality forages (Bohnert et al., 2002b; Ferrell et al., 1999; Krehbiel et al., 1998). An effect of N source on PUN was also observed, with increased PUN in lambs supplemented with urea compared with SBM. Urea is quickly hydrolyzed to  $\text{NH}_3\text{-N}$  in the rumen, which is then either utilized by the ruminal microbes, absorbed across the rumen wall, or flows to the duodenum for absorption in the small

intestine. Ammonia absorbed from the gastrointestinal tract is rapidly converted to urea by the liver (Van Soest, 1982) and subsequently released into the circulation. While a portion of supplemental N from SBM is converted to  $\text{NH}_3\text{-N}$  in the rumen, a lower, delayed peak in ruminal  $\text{NH}_3\text{-N}$  concentrations is typically observed when compared to NPN sources (Owens and Zinn, 1988). Consequently, lower ruminal  $\text{NH}_3\text{-N}$  levels, and thus lower PUN concentrations, should be expected with SBM supplementation when compared to urea supplementation.

Decreased PUN has been reported with decreased SF in other studies with ruminants consuming foraged-based diets (Bohnert et al., 2002b; Currier et al., 2004a; Huston et al., 1999a). In general, the response to infrequent supplementation is a larger peak in PUN following supplementation events compared with daily treatments. This typically occurs within 24 h and is proportional to the quantity of supplement provided (Currier et al., 2004a; Schauer et al., 2010). We noted an almost 100% decrease in PUN on the day after supplementation for the urea 2D treatment (Fig. 1). This response for urea 2D is similar to the response reported by Currier et al. (2004a) for lambs consuming the same treatment and comparable quality forage. In contrast, we noted a small increase in PUN for SBM 2D compared with D, most likely a consequence of delayed ruminal/intestinal digestion of protein in SBM. As a result, the decreased average PUN observed with the 2D compared with D treatments appears primarily due to the aforementioned decrease in PUN for urea 2D on the day following supplementation (Fig. 1).

## 6. Conclusion

In conclusion, supplementation of steers consuming low-quality forage increased total and forage DMI compared with unsupplemented steers. Conversely, supplementation had no influence on DMI by lambs, likely a result of high NDF intake, greater N intake as a percentage of BW, and sufficient ruminal available N. Additionally, N source and supplementation frequency had little effect on intake and digestibility of low-quality hard fescue straw by lambs and steers. These results suggest that supplements containing urea or SBM as the supplemental N source can be effectively used by lambs and steers consuming cool-season, low-quality forage without adversely affecting N efficiency, nutrient intake, or nutrient digestibility, even when provided every-other-day. As a result, reducing supplementation frequency to every-other-day would result in a 50% savings in the costs associated with supplementation (fuel, labor, etc.). Also, supplementation costs can be further reduced by incorporating urea, normally cheaper per unit N than sources of natural protein, into supplementation programs for use by mature ruminants consuming low-quality forages.

## Conflict of interest

None.

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