

**COMPARISON OF UREA AND BIURET AS NITROGEN SUPPLEMENTS TO LOW-QUALITY FORAGE: DAILY AND ALTERNATE DAY SUPPLEMENTATION EFFECTS ON EFFICIENCY OF NITROGEN USE IN LAMBS**

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**ABSTRACT:** Five wethers ( $39 \pm 1$  kg BW) were used in an incomplete  $5 \times 4$  Latin square with four 24-d periods to determine the influence of supplemental non-protein nitrogen (NPN) source and supplementation frequency (SF) on efficiency of N use in lambs consuming low-quality forage (4.3% CP). Treatments included an unsupplemented control (CON) and a urea (28.7% CP) or biuret (28.6% CP) supplement provided daily (D) or every other day (2D) at 0700. Lambs were provided forage at 120% of the previous 5 d average intake in two equal portions at 0715 and 1900. Experimental periods were 24 d with an 18 d adaptation period. Feces and urine were collected on d 19 to 24. Blood samples were obtained 2, 4, and 6 h post-supplementation on d 19 to 24 for analysis of plasma urea-N (PUN). DMI, OM intake, N retention, DM, OM, and N digestibility, and digested N retained were greater ( $P < 0.02$ ) for supplemented wethers compared with CON with no difference ( $P > 0.05$ ) because of NPN source or SF. However, it is of interest to note that, even though no statistical difference was observed for digested N retained between NPN sources, digested N retained was 110% greater for biuret compared with urea. Supplemented lambs had increased PUN compared with CON ( $P < 0.01$ ) and urea treatments had greater PUN compared with biuret ( $P < 0.01$ ). Also, PUN was increased ( $P = 0.02$ ) for D compared with 2D treatments. In addition, data suggest that PUN exhibited less fluctuation on the day of a supplementation event for biuret compared with urea. These results suggest that supplements containing urea or biuret as the supplemental N source can be effectively used by lambs consuming low-quality forage without adversely affecting N efficiency, even when provided every other day. In addition, biuret should have greater utility for use in supplements offered infrequently to ruminants because it is comparatively nontoxic compared with urea.

**Key Words:** Urea, Biuret, Forage, Non-protein Nitrogen, Lamb

### Introduction

It has been 36 yr since Virtanen (1966) demonstrated that ruminants could convert non-protein nitrogen (NPN) to milk protein. Sources of NPN are an attractive protein replacement due to their low cost compared with natural proteins (per unit of nitrogen).

Consequently, numerous studies have been conducted evaluating NPN as a source of supplemental nitrogen. Urea, the most commonly used NPN source, is extremely soluble in water and is rapidly hydrolyzed to ammonia within the rumen. This can lead to ammonia toxicity if urea is consumed in large quantities within a short period of time (Raleigh and Wallace, 1963; Helmer and Bartley, 1971; Bartley et al., 1976). In contrast, biuret is not very soluble in water and is degraded to ammonia at a slower rate compared with urea (Fonnesbeck et al., 1975). As a result, biuret is comparatively non-toxic (Hatfield et al., 1959) and, therefore, can be incorporated into supplements at higher concentrations than urea. Also, biuret does not elicit the negative effects on palatability and intake often observed with urea (Fonnesbeck et al., 1975; Clanton, 1978).

Decreasing the frequency of supplementation is one management practice that decreases labor costs. Nolan and Leng (1972) suggested that recycling of absorbed N to the rumen may support fermentation between times of supplementation. In addition, research has shown that protein supplements can be fed at infrequent intervals and still maintain acceptable levels of performance (Hunt et al., 1989; Huston et al., 1997; Bohnert et al., 2001); however, data is limited comparing the effects of urea and biuret supplemented at infrequent intervals on forage intake, forage digestibility, and efficiency of N use. The objective of this research is to compare daily and alternate day supplementation of urea or biuret on utilization of low-quality forage by ruminants. This knowledge will assist in developing management strategies that help reduce winter feed costs while maintaining acceptable levels of production.

### Materials and Methods

Five wethers ( $39 \pm 1$  kg) were used in an incomplete  $5 \times 4$  Latin square design to evaluate the efficacy of N use in lambs supplemented with a urea or biuret supplement (Table 1) every day or every other day. Wethers were randomly allotted to treatments and housed in individual metabolism crates within an enclosed barn with continuous lighting.

Wethers had continuous access to fresh water and low-quality grass seed straw (Table 1). Treatments were arranged as a  $2 \times 2$  factorial, two sources of supplemental NPN and two supplementation frequencies

(SF), with a negative control (CON; no supplementation). Crude protein supplements were offered every day (D) or every other day (2D) at 0700. The urea and biuret 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. Urea and biuret intake was approximately .175, .350, .207, and .416 g/kg BW on each supplementation day for urea D, urea 2D, biuret D, and biuret 2D, respectively. The amount of CP supplied by each supplement was approximately 0.10% of BW/d (averaged over a 2 d period). Forage was provided daily at 120% of the average intake for the previous 5 d in two equal portions (0715 and 1900), with feed refusals from the previous day determined before the 0700 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. 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 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 grass seed straw and protein supplements were collected on d 17 to 22, while orts were collected on d 18 to 23. Samples of feed and orts were 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 (50% of total; weight basis) and stored at 4°C. Sufficient 6 N HCl (100 mL) was added daily to urinals to maintain urine pH < 3. A sub-sample of each daily fecal sample (7.5%; wet weight basis) was dried at 55°C for 96 h for calculation of fecal DM. On d 19 to 24, 12 mL of blood was collected via jugular venipuncture 2, 4, and 6 h after the 0700 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 (5000 × g, 4°C, 15 min), and plasma harvested and stored (-20°C).

Dried samples were ground through a Wiley mill (1-mm screen). Samples of ground grass seed straw and CP supplements were composited by period and daily orts composited by lamb (within period) on an equal weight basis (20% as-fed). 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 (Leco CN-2000, Leco Corporation, St. Joseph, MI). 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, Inc., Rochester, NY).

Data were analyzed as an incomplete 5 × 4 Latin square using the GLM procedure of SAS (1996). The model included period, wether, and treatment. Because the treatment structure consisted of a 2 × 2 factorial plus a negative control, orthogonal contrasts were used to partition specific treatment effects. Contrast statements included: 1) Control vs CP supplementation; 2) Urea vs Biuret; 3) D vs 2D; 4) NPN source × SF. Response variables included: 1) DM and OM intake; 2) total tract digestibility of DM, OM, and N; 3) N balance; and 4) digested N retained. Plasma urea-N was analyzed using the REPEATED statement with the MIXED procedure of SAS (1996). The model included lamb, period, treatment, hour, frequency, treatment × frequency, treatment × hour, and treatment × hour × frequency. In addition, lamb × period × treatment was used to specify variation between animals (using the RANDOM statement). Autoregression was used as the covariance structure. The same contrasts noted above were used to partition treatment sums of squares.

## Results and Discussion

Intake of hay DM and OM by lambs was not affected ( $P > 0.10$ ) by CP supplementation while there was a tendency ( $P = 0.08$ ) for hay DM and OM intake to decrease as SF decreased (Table 2). Total DM, OM, N, and NDF intake increased ( $P < 0.05$ ) with supplementation. Also, total DM and OM intake tended to decrease ( $P = 0.08$ ) as SF decreased, while NDF intake decreased ( $P = 0.04$ ) as SF decreased.

Total tract digestibility of DM, OM, N, NDF, and ADF were increased ( $P < 0.03$ ) with CP supplementation (Table 2). This agrees with other studies in which N supplementation of low-quality forage resulted in increased digestibility compared with an unsupplemented control (DeIurato et al., 1990; Scott and Hibberd, 1990). Daily fecal and urinary N excretion (g/kg BW) was increased ( $P < 0.01$ ) with CP supplementation. However, no differences were noted because of NPN source or SF. Daily N balance and digested N retained (g/kg BW) were greater ( $P < 0.03$ ) with CP supplementation with no difference because of NPN source. However, it is of interest to note that even though no statistical difference was observed between urea and biuret, average digested N retained with biuret supplementation was, numerically, 110% greater compared with urea.

Treatment × hour and treatment × SF 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 × hour figure would aid in interpretation and discussion of the data. Lamb plasma urea-N was greater ( $P < 0.01$ ) for CP supplemented lambs and urea had greater ( $P < 0.01$ ) plasma urea-N than biuret (Table 2). Also, plasma urea-N decreased ( $P = 0.02$ ) as SF decreased. This agrees with the findings of Bohnert et

al. (2001). They supplemented lambs consuming low-quality forage with degradable or undegradable intake protein every day, every third day, or every sixth day. They noted that plasma urea-N was increased with CP supplementation and decreased as SF decreased. Figure 1 provides an illustration of plasma urea-N means for the day of and day before supplementation over the six day collection period. Interestingly, plasma urea-N was fairly constant on the day of supplementation for UD but increased from 2 to 6 h post-supplementation for U2D. In contrast, plasma urea-N was similar over the collection period on the day of supplementation for BD and B2D. On the day before supplementation, plasma urea-N responded in a like manner for the U2D and B2D treatments (decreasing over the collection period). However, the difference between daily and alternate day treatments was less for BD and B2D compared with UD and U2D.

### Implications

Ruminants consuming low-quality forage (< 6% crude protein) can effectively use supplemental non-protein nitrogen. In addition, daily and alternate day supplementation of non-protein nitrogen results in similar efficiency of nitrogen use. Ruminants appear to have the ability to conserve nitrogen over extended periods, thereby storing it for use between supplementation events. Infrequent supplementation of non-protein nitrogen, primarily urea, should be conducted with caution because of the potential for ammonia toxicity. However, biuret is safer compared with urea because of its decreased solubility and slower hydrolysis to ammonia.

### Literature Cited

- AOAC. 1990. Official Methods of Analysis (15th Ed.). Association of Official Analytical chemists, Arlington, VA.
- Bartley, E. E., A. D. Davidovich, G. W. Barr, G. W. Griffel, A. D. Dayton, C. W. Deyoe, and R. M. Bechtel. 1976. Ammonia toxicity in cattle. I. Rumen and blood changes associated with toxicity and treatment methods. *J. Anim. Sci.* 43:835-841.
- 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.
- Bohnert, D. W., C. S. Schauer, and T. DelCurto. 2001. 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. *Proc. West. Sec. Amer. Soc. Anim. Sci.* 52:305-310.
- Clanton, D. C. 1978. Non-protein nitrogen in range supplements. *J. Anim. Sci.* 47:765-779.
- 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.
- Fonnesbeck, P. V., L. C. Kearn, and L. E. Harris. 1975. Feed grade biuret as a protein replacement for ruminants - A Review. *J. Anim. Sci.* 40:1150-1184.
- Goering, H. K., and P. J. Van Soest. 1970. Forage fiber analysis (apparatus, reagents, procedures and some applications). *Agric. Handbook No. 379.* ARS, USDA, Washington, DC.
- Hatfield, E. E., U. S. Garrigus, R. M. Forbes, A. L. Neumann, and W. Gaither. 1959. Biuret - A source of NPN for ruminants. *J. Anim. Sci.* 18:1208-1219.
- Helmer, L. G., and E. E. Bartley. 1971. Progress in the utilization of urea as a protein replacer for ruminants. A review. *J. Dairy Sci.* 54:25-51.
- 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., H. Lippke, T. D. A. Forbes, J. W. Holloway, R. V. Machen, B. G. Warrington, K. Bales, S. Engdahl, C. Hensg, P. Thompson, and D. Spiller. 1997. Effects of frequency of supplementation of adult cows in western Texas. *Proc. Western Sect. Amer. Soc. Anim. Sci.* 48:236-238.
- Mass, R. A., G. P. Lardy, R. J. Grant, and T. J. Klopfenstein. 1999. In situ neutral detergent nitrogen as a method for measuring forage protein degradability. *J. Anim. Sci.* 77:1656-1571.
- Nolan, J. V., and R. A. Leng. 1972. Dynamic aspects of ammonia and urea metabolism in sheep. *Br. J. Nutr.* 27:597-600.

Raleigh, R. J., and J. D. Wallace. 1963. Effect of urea at different nitrogen levels on digestibility and on performance of growing steers fed low quality flood meadow roughage. *J. Anim. Sci.* 22:330-334.

Robertson, J. B., and P. J. Van Soest. 1981. The detergent system of analysis and its application to human foods. In: W. P. T. James and O. Theander (Eds.) *The Analysis of Dietary Fiber*. pp. 123-158. Marcell Dekker, New York.

SAS. 1996. *SAS/STAT Software Changes and Enhancements Through Release 6.11*. SAS Inst. Inc., Cary, NC.

Scott, R. R., and C. A. Hibberd. 1990. Incremental levels of supplemental ruminal degradable protein for beef cows fed low quality native grass hay. MP-129, p. 57. Oklahoma State Univ. Animal Sci. Res. Rep.

Virtanen, A. I. 1966. Milk production of cows on protein-free feed. *Science*. 153:1603-1614.

Table 1. Supplement composition and feedstuff nutrient content

Item	Hard Fescue Straw	Urea Supplement <sup>a</sup>	Biuret Supplement <sup>a</sup>
Urea	-	5.3	-
Biuret	-	-	6.1
Soy Hulls	-	91.0	90.2
Dried Molasses	-	3.7	3.7
Nutrient Composition			
CP, % DM	4.3	28.7	28.6
DIP <sup>b</sup> , %CP	76.0	83.0	84.2
OM, % DM	93.6	90.2	92.4
NDF, % DM	73.8	57.9	55.4
ADF, %DM	32.0	38.1	38.2

<sup>a</sup> Pelleted supplements were provided by ADM Alliance Nutrition, Inc., Quincy, IL.

<sup>b</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.

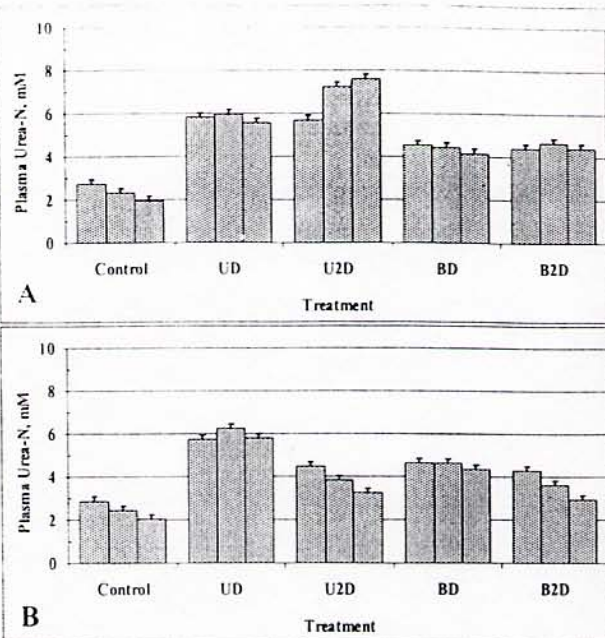


Figure 1. Effect of non-protein nitrogen source and supplementation frequency on lamb plasma urea-N (mM) on the day of (A) and the day before (B) supplementation. Columns from left to right for each treatment represent lamb plasma urea-N at 2, 4 and 6 h post-supplementation. Treatments were: Control; UD = Urea supplement every day; U2D = Urea supplement every other day; BD = Biuret supplement every day; B2D = Biuret supplement every other day

Table 2. Effect of non-protein nitrogen source and supplementation frequency on intake, diet digestibility, and nitrogen balance by lambs consuming low-quality forage

Item	Treatment <sup>a</sup>										P-Value <sup>c</sup>									
	CON		UD		U2D		BD		B2D		SEM <sup>b</sup>		Con vs Urea vs Biuret		Daily vs Alternate D		NPN Source × SF			
Daily DM Intake, g/kg BW																				
Hay	26.4	28.2	26.1	28.2	26.1	28.2	26.1	28.2	26.1	28.2	1.1	1.1	0.56	0.99	0.08	0.99	0.99			
Supplement <sup>d</sup>	0.0	3.3	3.3	3.4	3.3	3.4	3.4	3.4	3.4	3.4										
Total	26.4	31.5	29.4	31.6	29.4	31.6	29.4	31.6	29.4	31.6	1.1	1.1	0.009	0.97	0.08	0.99	0.99			
Daily OM Intake, g/kg BW																				
Hay	24.8	26.5	24.5	26.4	24.5	26.4	24.4	26.4	24.4	26.4	1.0	1.0	0.56	0.97	0.08	0.97	0.97			
Supplement <sup>d</sup>	0.0	3.0	3.0	3.1	3.0	3.1	3.1	3.1	3.1	3.1										
Total	24.8	29.5	27.5	29.5	27.5	29.5	27.5	29.5	27.5	29.5	1.0	1.0	0.01	0.96	0.08	0.97	0.97			
Daily N Intake, g/kg BW	0.183	0.347	0.331	0.343	0.331	0.343	0.347	0.343	0.347	0.347	0.012	0.012	<0.001	0.58	0.63	0.42	0.42			
Daily NDF Intake, g/kg BW	19.6	22.9	21.3	22.8	21.3	22.8	20.8	22.8	20.8	20.8	0.7	0.7	0.02	0.68	0.04	0.75	0.75			
Total Tract Digestibility, %																				
DM	39.2	48.0	47.9	47.8	47.9	47.8	45.3	47.8	45.3	45.3	1.9	1.9	0.006	0.50	0.51	0.55	0.55			
OM	42.8	51.4	51.2	51.0	51.2	51.0	48.7	51.0	48.7	48.7	2	2	0.009	0.49	0.56	0.63	0.63			
N	24.3	53.0	48.5	51.9	48.5	51.9	52.3	51.9	52.3	52.3	2.9	2.9	<0.001	0.66	0.51	0.43	0.43			
NDF	42.2	50.8	51.1	49.5	51.1	49.5	46.1	49.5	46.1	46.1	2.3	2.3	0.02	0.20	0.51	0.44	0.44			
ADF	42.9	52.5	52.0	51.8	52.0	51.8	46.9	51.8	46.9	46.9	2.4	2.4	0.02	0.27	0.31	0.40	0.40			
Daily N excretion, g/kg BW																				
Fecal	0.136	0.160	0.170	0.167	0.170	0.167	0.164	0.167	0.164	0.164	0.008	0.008	0.01	0.92	0.71	0.46	0.46			
Urinary	0.059	0.144	0.148	0.135	0.148	0.135	0.142	0.135	0.142	0.142	0.007	0.007	<0.001	0.27	0.45	0.79	0.79			
Daily N balance, g/kg BW	-0.012	0.042	0.013	0.041	0.013	0.041	0.041	0.041	0.041	0.041	0.014	0.014	0.02	0.36	0.34	0.35	0.35			
Daily digested N retained <sup>f</sup> , %	-54.4	16.3	6.5	28.6	6.5	28.6	19.4	28.6	19.4	19.4	15.6	15.6	0.003	0.45	0.56	0.98	0.98			
Plasma urea-N, mM	2.40	5.86	5.36	4.46	5.36	4.46	4.05	4.46	4.05	4.05	0.16	0.16	<0.001	<0.001	0.02	0.78	0.78			

<sup>a</sup> CON = control; UD = urea supplement every other day; U2D = urea supplement every other day; BD = biuret supplement every day; B2D = biuret supplement every other day. <sup>b</sup> n = 4.

<sup>c</sup> Con vs Supp = control vs supplemented treatments; Urea vs Biuret = urea vs biuret treatments; Daily vs Alternate D = daily vs alternate day supplementation; NPN Source × SF = interaction of NPN source vs supplementation frequency.

<sup>d</sup> UD received 3.3 g/kg BW daily; U2D received 6.6 g/kg BW every other day; BD received 3.4 g/kg BW daily; B2D received 6.8 g/kg BW every other day.

<sup>e</sup> UD received 3.0 g/kg BW daily; U2D received 6.0 g/kg BW every other day; BD received 3.1 g/kg BW daily; B2D received 6.2 g/kg BW every other day.

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