Daily and alternate-day supplementation of urea or biuret to ruminants consuming low-quality forage: III. Effects on ruminal fermentation characteristics in steers[1,2]

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ABSTRACT: Five ruminally and duodenally cannulated steers (491 ± 21 kg BW) were used in an incomplete 5 × 4 Latin square with four 24-d periods to determine the influence of supplemental nonprotein N (NPN) source and supplementation frequency (SF) on the dynamics of ruminal fermentation in steers consuming low-quality grass straw (4% CP). Treatments (TRT) included an unsupplemented control (CON) and a urea or biuret supplement that were placed directly into the rumen at 0700 daily (D) or every other day (2D). The NPN treatments were formulated to provide 90% of the estimated degradable intake protein requirement; therefore, the urea and biuret treatments received the same amount of supplemental N over a 2-d period. Daily TRT were supplemented with CP at 0.04% of BW/d, whereas the 2D TRT were supplemented at 0.08% of BW every other day. Forage was provided at 120% of the previous 5-d average intake in two equal portions at 0715 and 1900. Ruminal fluid was collected 0, 3, 6, 9, 12, and 24 h after supplementation on a day of and a day before supplementation for all TRT. Ruminal NH_3-N increased ($P < 0.04$) with CP supplementation on the day all supplements were provided and on the day on which only daily supplements were provided compared with the CON. However, an NPN source × SF interaction ($P = 0.03$) on the day all supplements were provided indicated that NH_3-N increased at a greater rate for urea as SF decreased compared with biuret. Ruminal NH_3-N on the day only daily supplements were provided was greater ($P = 0.02$) for D compared with 2D. On the day all supplements were provided, D increased ($P = 0.05$) ruminal indigestible acid detergent fiber passage rate and ruminal fluid volume compared with 2D. These results suggest that urea or biuret can be used effectively as a supplemental N source by steers consuming low-quality forage without adversely affecting ruminal fermentation, even when provided every other day.

Key Words: Biuret, Forage, Frequency, Nonprotein Nitrogen, Supplementation, Urea

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Introduction

From late summer through winter, ruminants typically consume low-quality forage (<6% CP), resulting in low levels of ruminal NH_3-N that can hinder microbial protein synthesis and ruminal fermentation (Köster et al., 1996). Supplementation with degradable intake protein (**DIP**) has been shown to increase ruminal NH_3-N in ruminants consuming low-quality forage (Köster et al., 1996; Bohnert et al., 2002b). Ruminal NH_3-N has been estimated to provide 40 to 100% of the N used in the production of microbial protein (Stern and Hoover, 1979), the primary source of protein for ruminants consuming low-quality forage (Köster et al., 1996).

Decreasing the frequency of CP supplementation administered to ruminants consuming low-quality forage has been shown to result in acceptable levels of performance (Bohnert et al., 2002a) with only minimal impacts on nutrient intake and digestibility (Beaty et al., 1994; Köster et al., 1996). This supports the hypothesis that N recycling may support ruminal fermentation between supplementation events.

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Sources of nonprotein N (NPN) are attractive alternatives to most sources of natural protein because of their low cost per unit of N. However, the rapid hydrolysis of urea to NH_3-N can result in NH_3 toxicity if consumed in large quantities within a short period of time (Bartley et al., 1976). In contrast, biuret is comparatively nontoxic because it is less soluble in water and is ruminally degraded to NH_3-N at a rate slower than that of urea (Fonnesbeck et al., 1975). We are aware of limited data concerning ruminal fermentation in response to infrequent supplementation of NPN, with none comparing infrequent supplementation of urea and biuret. Therefore, the objective of this research was to compare ruminal fermentation in response to daily and alternate-day supplementation of urea or biuret to steers consuming low-quality forage.

Materials and Methods

A full description of experimental procedures (excluding ruminal fermentation measurement and analysis) and diet composition is given in a companion paper (Currier et al., 2004b). Briefly, five cannulated (ruminal and duodenal) beef steers (491 ± 21 kg) were allotted randomly to one of five treatments in an incomplete 5 × 4 Latin square (Cochran and Cox, 1957) and were housed in individual pens (4 × 8 m) within an enclosed barn with continuous lighting. Treatments consisted of an unsupplemented control and urea or biuret supplemented daily (**D**) or every other day (**2D**; **CON** = control, **UD** = urea supplement every day, **U2D** = urea supplement every other day, **BD** = biuret supplement every day, and **B2D** = biuret supplement every other day). Supplemented treatments were formulated to provide 90% of the estimated degradable intake protein requirement assuming a microbial efficiency of 11% (NRC, 1996). 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.

Experimental periods were 24 d long, with 10 d of diet adaptation and 14 d of sampling. On d 13 and 18, treatment effects on ruminal DM and indigestible ADF (**IADF**) fill were determined by manually removing reticulorumen contents 4 h after feeding. This allowed sampling on the day all supplements were provided and the day on which only daily supplements were provided, respectively. Total ruminal contents were weighed, mixed by hand, and subsampled in triplicate (approximately 400 g). The remaining ruminal contents were replaced immediately into the steer. Ruminal samples were weighed, dried in a forced-air oven (55°C; 96 h), reweighed for DM, ground to pass a 1-mm screen in a Wiley mill, and composited within period and day by steer.

On d 19 and 24, each steer was intraruminally pulse-dosed with 5 g of Co-EDTA in a 150-mL aqueous solution (Uden et al., 1980) at 0700 (the time at which supplements were provided). As described above for ruminal evacuations, this allowed sampling on the day all supplements were provided and the day on which only daily supplements were provided. The Co marker was administered throughout the rumen using a stainless steel probe with a perforated tip. Ruminal fluid (approximately 100 mL) was collected by suction strainer (Raun and Burroughs, 1962; 19-mm diameter, 1.6-mm mesh) immediately before dosing and at 3, 6, 9, 12, and 24 h after dosing. Ruminal fluid pH was measured immediately after collection (model SA 520, Orion Research, Inc., Boston, MA). Twenty milliliters was stored (–20°C) for later analysis of Co concentration and 5 mL was acidified with 1 mL of 25% (wt/vol) metaphosphoric acid and stored (–20°C) for subsequent analysis of VFA and NH_3-N. Frozen (–20°C) ruminal samples were prepared for analysis by thawing, centrifuging (15,000 × *g* for 10 min for VFA and NH_3-N; 2,000 × *g* for 20 min for Co), and collecting the supernatant. Cobalt concentration in ruminal fluid was analyzed by atomic absorption using an air/acetylene flame (Model 351 AA/AE Spectrophotometer, Instrumentation Laboratory, Inc., Lexington, MA). Ruminal fluid fill and fluid dilution rate were estimated by regressing the natural logarithm of Co concentration against sampling time as described by Warner and Stacy (1968). Volatile fatty acids were analyzed as described by Horney et al. (1996) and NH_3-N by a modification (sodium salicylate substituted for phenol) of the procedure described by Broderick and Kang (1980) using a UV/VIS spectrophotometer (Spectronic 710 Spectrophotometer, Bausch & Lomb, Inc., Rochester, NY).

Ground samples of hard fescue straw and CP supplements were composited by period and daily orts composited by steer (within period) on an equal-weight basis (5% as-fed). Feed, orts, and ruminal particulate were analyzed for DM and OM (AOAC, 1990), and, except for ruminal particulate, 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, ruminal particulate, and fecal samples (from Currier et al., 2004a) were analyzed for IADF as described by Bohnert et al. (2002b). Fecal recovery of IADF was 87.0 ± 1.0%. Digesta kinetics techniques described by Van Soest (1982) were used to determine IADF passage by dividing IADF intake by the quantity of IADF in the rumen 4 h after feeding.

Statistical Analysis

Ruminal fluid fill, fluid dilution rate, DM fill, IADF fill, and IADF passage rate were analyzed as an incomplete 5 × 4 Latin square using the GLM procedure of SAS (SAS Inst. Inc., Cary, NC). The model included period, steer, 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 were 1)

Table 1. Effects of nonprotein nitrogen (NPN) source and supplementation frequency on ruminal DM fill, indigestible acid detergent fiber (IADF) fill, fluid fill, and fluid and IADF passage rates in steers fed hard fescue straw

	Treatment[a]						*P*-value[c]			
Item	CON	UD	U2D	BD	B2D	SEM[b]	CON vs. Suppl.	Urea vs. Biuret	D vs. 2D	NPN source × SF
Day all supplements provided										
DM fill, g/kg BW	33.9	35.3	35.3	34.0	36.4	0.8	0.14	0.87	0.16	0.16
IADF fill, g/kg BW	8.40	8.76	8.76	8.61	8.82	0.19	0.16	0.81	0.62	0.58
IADF passage, %/h	1.62	1.62	1.58	1.71	1.59	0.03	0.99	0.20	0.05	0.22
Fluid fill, mL/kg BW	209	228	194	248	223	13	0.35	0.09	0.05	0.72
Fluid dilution rate, %/h	7.8	7.6	8.1	7.3	7.6	0.3	0.71	0.19	0.15	0.79
Day only daily supplements provided										
DM fill, g/kg BW	35.7	37.2	35.1	37.6	35.6	1.0	0.59	0.67	0.09	0.93
IADF fill, g/kg BW	8.80	9.34	8.86	9.53	8.94	0.24	0.21	0.59	0.06	0.81
IADF passage, %/h	1.55	1.52	1.57	1.53	1.56	0.04	0.89	0.96	0.39	0.76
Fluid fill, mL/kg BW	207	222	226	232	221	10	0.14	0.77	0.70	0.46
Fluid dilution rate, %/h	8.8	8.5	8.7	8.7	9.0	0.3	0.93	0.46	0.45	0.92

[a]CON = control; UD = urea supplement provided every day; U2D = urea supplement provided every other day; BD = biuret supplement provided every day; B2D = biuret supplement provided every other day.
[b]n = 4.
[c]CON vs. Suppl. = control vs. supplemented treatments; urea vs. biuret = urea vs. biuret treatments; D vs. 2D = daily vs. alternate-day supplementation; NPN source × SF = interaction of NPN source vs. supplementation frequency.

CON vs. CP supplementation, 2) urea vs. biuret, 3) D vs. 2D supplementation, and 4) NPN source × supplementation frequency (**SF**).

Ruminal pH, NH_3-N, and VFA data, collected at the fixed times after feeding on the day all supplements were provided and the day on which only daily supplements were provided (d 19 and 24, respectively) were analyzed using the REPEATED statement with the MIXED procedure of SAS. The model included steer, period, treatment, time, and treatment × time. In addition, steer × period × treatment was used to specify variation between steers (using the RANDOM statement). Steer × period × treatment was used as the SUBJECT, and autoregression was used as the covariance structure. The same contrasts noted above were used to partition the treatment sums of squares.

Results

On the day all supplements were provided, ruminal DM fill and IADF fill were not affected ($P > 0.13$) by CP supplementation, NPN source, or SF (Table 1). However, ruminal IADF passage rate was greater ($P = 0.05$) for D treatments compared with 2D treatments, with no difference ($P > 0.19$) because of CP supplementation or NPN source.

Ruminal fluid fill was greater ($P = 0.05$) for D treatments compared with 2D treatments, whereas no difference ($P = 0.35$) was noted because of CP supplementation on the day all supplements were provided (Table 1). However, there was a tendency ($P = 0.09$) for biuret treatments to have higher ruminal fluid fill than urea treatments. Ruminal fluid dilution rate was not affected ($P > 0.14$) by CP supplementation, NPN source, or SF on the day all supplements were provided.

On the day on which only daily supplements were provided, ruminal DM fill tended ($P = 0.09$) to be greater for D compared with 2D treatments (Table 1). Similarly, ruminal IADF fill tended ($P = 0.06$) to be greater for D compared with 2D treatments. There were no differences ($P > 0.20$) in ruminal DM fill or IADF fill because of CP supplementation or NPN source. Likewise, there was no effect ($P > 0.13$) of CP supplementation, NPN source, or SF on ruminal IADF passage rate, fluid fill, or fluid dilution rate on the day only daily supplements were provided.

Treatment × time interactions ($P < 0.01$) were noted for ruminal NH_3-N on the day all supplements were provided and on the day on which only daily supplements were provided. However, after considering the nature of the interactions, we concluded that discussing treatment means while providing the treatment × time figures would aid in the interpretation and discussion of the data. No treatment × time interactions ($P > 0.10$) were detected for ruminal pH and VFA data. Therefore, overall treatment means are discussed.

On the day all supplements were provided, ruminal NH_3-N increased ($P < 0.01$; Table 2; Figure 1) over twofold with CP supplementation. In addition, a NPN source × SF interaction ($P = 0.03$) occurred, indicating ruminal NH_3-N increased at a greater rate, and magnitude, with urea supplementation as SF decreased compared with biuret supplementation. Ruminal pH and total VFA were not affected ($P > 0.21$) by CP supplementation, NPN source, or SF on the day all supplements were provided (Table 2). Also, molar proportions of individual VFA, and the acetate:propionate ratio, were not affected ($P > 0.06$) by CP supplementation, NPN source, or SF.

Table 2. Effects of nonprotein nitrogen (NPN) source and supplementation frequency on steer ruminal fermentation characteristics on the day all supplements were provided

	Treatment[a]						*P*-value[c]			
Item	CON	UD	U2D	BD	B2D	SEM[b]	CON vs. Suppl.	Urea vs. Biuret	D vs. 2D	NPN source × SF
Ammonia N, m*M*	1.36	2.57	4.47	2.72	3.30	0.24	<0.001	0.07	0.001	0.03
pH	6.45	6.49	6.50	6.54	6.50	0.04	0.22	0.49	0.75	0.56
Total VFA, m*M*	68.8	71.1	70.5	67.6	70.7	2.2	0.64	0.47	0.59	0.43
VFA, mol/100 mol										
Acetate	75.8	75.9	75.8	76.2	75.6	0.31	0.84	0.89	0.36	0.37
Propionate	15.8	16.3	16.3	15.6	16.3	0.19	0.24	0.11	0.12	0.08
Isobutyrate	0.27	0.26	0.28	0.26	0.26	0.12	0.90	0.43	0.60	0.66
Butyrate	7.1	6.6	6.5	6.9	6.9	0.2	0.35	0.20	0.83	0.99
Isovalerate	0.40	0.40	0.46	0.42	0.38	0.02	0.54	0.28	0.63	0.07
Valerate	0.64	0.60	0.62	0.61	0.59	0.03	0.29	0.81	0.99	0.45
Acetate:propionate	4.81	4.71	4.72	4.93	4.68	0.07	0.52	0.25	0.11	0.10

[a]CON = control; UD = urea supplement provided every day; U2D = urea supplement provided every other day; BD = biuret supplement provided every day; B2D = biuret supplement provided every other day.
[b]n = 4.
[c]CON vs. Suppl. = control vs. supplemented treatments; urea vs. Biuret = urea vs. biuret treatments; D vs. 2D = daily vs. alternate day supplementation; NPN source × SF = interaction of NPN source vs. supplementation frequency.

On the day only daily supplements were provided, ruminal NH_3-N was increased ($P = 0.03$) with CP supplementation (Table 2; Figure 1). In addition, ruminal NH_3-N was greater for D compared with 2D treatments ($P = 0.02$) but not affected ($P = 0.41$) by NPN source. Ruminal pH, total VFA, molar proportions of individual VFA, and acetate:propionate ratio were not affected ($P > 0.16$) by CP supplementation, NPN source, or SF.

Discussion

In our review of research concerning NPN supplementation of ruminants consuming low-quality forage, we are aware of limited data comparing the effects of urea and biuret on ruminal fermentation (Oltjen et al., 1969; Chicco et al., 1971; Bond and Rumsey, 1973; Löest et al., 2001) and none that has compared the effects of infrequent supplementation of urea and biuret on ruminal fermentation.

Variable results have been reported for ruminal particulate and fluid fill and passage rates with CP supplementation of ruminants consuming low-quality forage. Most research has resulted in increased (DelCurto et al., 1990; Hannah et al., 1991; Köster et al., 1996), or no difference in (Caton et al., 1988; Krysl et al., 1989; Beaty et al., 1994), fluid and particulate fill and/or passage rates. Faichney (1993) reported in his review that rate of passage in ruminants is affected by dietary factors including intake. He suggested that, generally, increased intake is associated with increased passage rate. This may explain much of the inconsistency noted with ruminal fluid and particulate dynamics in CP-supplemented ruminants consuming low-quality forage. Therefore, our observation that CP supplementation did not affect ruminal DM, IADF, and fluid dynamics compared with the CON on the day all supplements were provided and the day on which only daily supplements were provided coincides with the lack of a CP-supplementation effect on forage DM and OM intake reported in a companion paper (Currier et al., 2004b).

Kropp et al. (1977) substituted urea for 0, 25, 50, or 75% of the total supplemental N provided by soybean meal to steers fed 3% CP forage and noted that increasing the proportion of supplemental N provided by urea did not affect ruminal fluid dilution rate. In addition, Köster et al. (1997) supplemented steers consuming dormant tallgrass prairie forage with supplements in which urea provided 0, 25, 50, 75, or 100% of the supplemental N, with casein providing the remainder of the supplemental N, and did not alter ruminal DM fill, fluid fill, or fluid dilution rate as the proportion of urea increased. These results coincide with our observation that NPN source did not affect ruminal DM fill, particulate fill and passage rate, and fluid fill and dilution rate on the day all supplements were provided and the day on which only daily supplements were provided. Therefore, it appears that urea and biuret elicit similar effects on ruminal fluid and particulate dynamics in ruminants consuming low-quality forage.

Our observation that ruminal IADF passage rate and ruminal fluid fill decreased as SF decreased on the day all supplements were provided disagrees with the results of Beaty et al. (1994) and Bohnert et al. (2002b). Beaty et al. (1994) provided supplemental protein 7 d/wk and 3 d/wk to steers consuming wheat straw and noted no difference in ruminal IADF passage rate or fluid volume as SF decreased. Bohnert et al. (2002b) supplemented steers fed low-quality meadow hay with low-DIP (40% DIP; CP basis) or high-DIP (82% DIP; CP basis) supplements daily, once every 3 d, or once

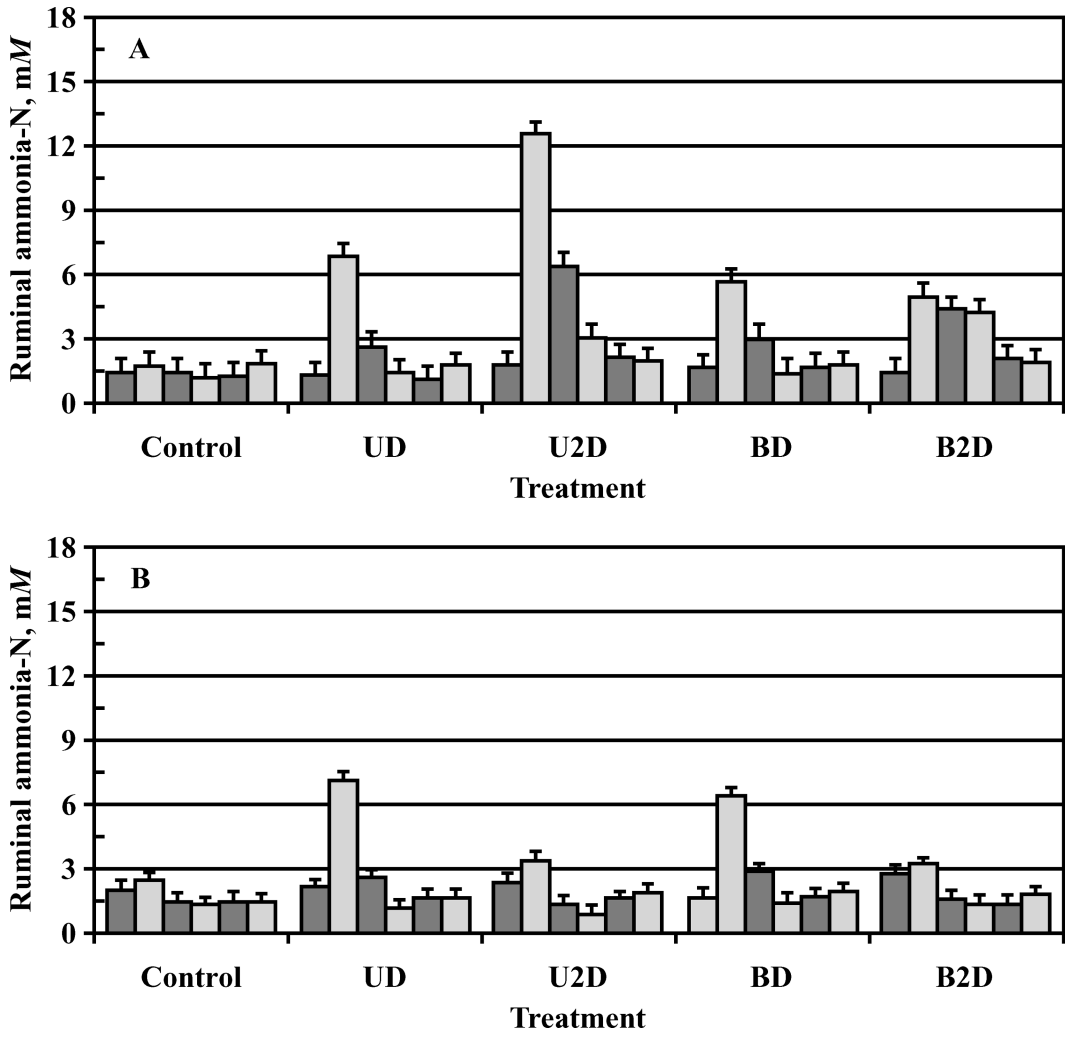

Figure 1. Effects of nonprotein nitrogen source and supplementation frequency on steer ruminal ammonia-N on the day all supplements were provided (A) and on the day on which only daily supplements were provided (B). Columns from left to right for each treatment represent 0, 3, 6, 9, 12, and 24 h after feeding, respectively. Treatments were Control; UD = urea supplement provided every day; U2D = urea supplement provided every other day; BD = biuret supplement provided every day; and B2D = biuret supplement provided every other day. Treatment × time interactions for A and B are ($P < 0.001$). SEM for A and B are 0.61 and 0.39, respectively.

every 6 d and noted that IADF passage rate was not affected by SF. It is probable that the decreased IADF passage rate as SF decreased on the day all supplements were provided in the current study is related to the tendency for forage and total OM intake to decrease as SF decreased (Currier et al., 2004b). In addition, this could explain the tendency for IADF fill to decrease as SF decreased on the day only daily supplements were provided. Bohnert et al. (2002b) noted that ruminal fluid fill increased as SF decreased from daily to once every 6 d with the high-DIP supplement but was not altered with the low-DIP supplement. They suggested that, on the day all supplements were provided, an increased ruminal fluid fill as SF decreased with the high-DIP supplement might have been caused by a disruption in rumen function caused by the large quantity of ruminally degradable supplement provided during a supplementation event for the infrequently supplemented groups compared with the daily treatment. This appears reasonable because infrequent supplementation of the low-DIP supplement (low rumen degradability) did not affect ruminal fluid fill on the day all supplements were provided. Furthermore, they noted that as SF of the high-DIP supplement decreased from daily to once every 6 d, fluid dilution rate decreased approximately 16% on the day all supplements were provided and approximately 22% on the day only daily supplements were provided. Results similar to ours were reported by Farmer et al. (2001). These results suggest that infrequently (> once every 3 d) providing a large quantity of a rumen-degradable CP supplement to ruminants consuming low-quality forage may disrupt rumen function (fluid and particulate fill and passage rates). However, our results suggest that alternate-day

Table 3. Effects of nonprotein nitrogen (NPN) source and supplementation frequency on steer ruminal fermentation characteristics on the day only daily supplements were provided

	Treatment[a]						*P*-value[c]			
Item	CON	UD	U2D	BD	B2D	SEM[b]	CON vs. Suppl.	Urea vs. Biuret	D vs. 2D	NPN source × SF
Ammonia N, m*M*	1.59	2.78	1.80	2.88	2.18	0.27	0.03	0.41	0.02	0.61
pH	6.48	6.53	6.54	6.47	6.53	0.05	0.50	0.52	0.48	0.68
Total VFA, m*M*	65.4	69.3	69.1	72.1	67.2	2.2	0.19	0.87	0.33	0.37
VFA, mol/100 mol										
Acetate	77.2	76.6	76.9	76.7	76.5	0.4	0.28	0.75	0.98	0.60
Propionate	14.6	15.1	15.0	15.1	15.1	0.3	0.18	0.95	0.92	0.90
Isobutyrate	0.28	0.28	0.34	0.28	0.30	0.02	0.48	0.44	0.17	0.61
Butyrate	6.8	6.9	6.7	6.9	7.1	0.2	0.54	0.36	0.86	0.23
Isovalerate	0.39	0.42	0.51	0.40	0.44	0.05	0.34	0.42	0.20	0.65
Valerate	0.67	0.62	0.56	0.62	0.61	0.04	0.17	0.59	0.46	0.70
Acetate:propionate	5.32	5.12	5.17	5.14	5.11	0.13	0.23	0.87	0.95	0.80

[a]CON = control; UD = urea supplement provided every day; U2D = urea supplement provided every other day; BD = biuret supplement provided every day; B2D = biuret supplement provided every other day.
[b]n = 4.
[c]CON vs. Suppl. = control vs. supplemented treatments; urea vs. biuret = urea vs. biuret treatments; D vs. 2D = daily vs. alternate day supplementation; NPN source × SF = interaction of NPN source vs. supplementation frequency.

supplementation of urea or biuret to ruminants consuming low-quality forage has minimal effects on ruminal fluid and particulate dynamics compared with daily supplementation.

Increased ruminal NH_3-N with CP supplementation of ruminants consuming low-quality forage has been demonstrated in numerous studies (Caton et al., 1988; Köster et al., 1996; Weder et al., 1999). This agrees with our observation that CP supplementation increased ruminal NH_3-N by approximately 240% on the day all supplements were provided and by 152% on the day on which only daily supplements were provided compared with the CON. Bohnert et al. (2002b) reported similar results with steers fed 5% CP meadow hay and provided a low- or high-DIP supplement daily, once every 3 d, or once every 6 d. They noted that ruminal NH_3-N was increased, on average, by 267 and 173% on the day all supplements were provided and the day only daily supplements were provided, respectively, compared with an unsupplemented control.

The NPN source × SF interaction observed for ruminal NH_3-N on the day all supplements were provided coincides with the ruminal CP degradability × SF interaction reported by Bohnert et al. (2002b) for ruminal NH_3-N. They noted that ruminal NH_3-N increased at a greater rate, and peaked at an elevated concentration, as SF decreased with a high-DIP supplement compared with a low-DIP supplement on the day all supplements were provided. In the current study, peak ruminal NH_3-N on the day all supplements were provided increased from approximately 7 m*M* with UD to almost 13 m*M* with U2D compared with peaks of approximately 5 m*M* for both BD and B2D (Figure 1). This is indicative of the lower ruminal solubility and slower enzymatic hydrolysis associated with biuret compared with urea (Fonnesbeck et al., 1975). In addition, other research has demonstrated higher ruminal NH_3-N concentrations for urea supplementation compared with biuret (Chicco et al., 1971; Bartle et al., 1998; Löest et al., 2001). These results can be interpreted to suggest that ruminal hydrolysis of biuret to NH_3-N is slower than hydrolysis of urea to NH_3-N. Therefore, biuret should be safer than urea when supplemented infrequently to ruminants. Also, early work with biuret suggested that an adaptation period is required to allow ruminal microorganisms to develop adequate biuretolytic activity (Schröder and Gilchrist, 1969) and this activity is rapidly lost when biuret supplementation is halted (Clemens and Johnson, 1973). However, research reported here and in the two companion papers suggests that adequate biuretolytic activity can be obtained after at least 18 d of supplementation and is not lost with every-other-day supplementation. This is based on our observation that ruminal NH_3-N concentration, N balance and cow performance (Currier et al., 2004a), and rumen microbial protein production (Currier et al., 2004b) were comparable to urea supplementation and not affected by SF.

Ruminal pH averaged approximately 6.5 for all treatments on the day all supplements were provided and on the day on which only daily supplements were provided. In addition, ruminal pH never fell below 6.3 (data not shown) and should have been sufficient to support adequate fiber digestion (Yokoyama and Johnson, 1988). This is supported by results reported in a companion paper that ruminal OM and NDF disappearance were not affected by CP supplementation, NPN source, or SF (Currier et al., 2004b).

Supplemental CP has been shown to increase total VFA and molar proportions of branched-chain VFA in ruminants consuming low-quality forage (Hannah et al., 1991; Köster et al., 1996); however, this did not occur on the day all supplements were provided or the day only daily supplements were provided in the current study. This can be at least partially explained by the type of supplemental DIP (NPN) used in the current study. Hannah et al. (1991) and Köster et al. (1996) offered sources of natural protein to ruminants consuming low-quality tallgrass prairie forage. These authors attributed increased VFA concentrations with CP supplementation to increased ruminal fermentation brought about by improving ruminal N status. However, sources of natural protein contain branched-chain amino acids that are precursors to branched-chain VFA. In contrast to the aforementioned studies, we used NPN (urea or biuret) as a source of supplemental CP. Consequently, we were not providing branched-chain amino acids as precursors for branched-chain VFA. Also, even though OM intake was greater with CP supplementation, CP supplementation did not affect ruminal disappearance of OM and NDF, ruminal fluid fill, or ruminal fluid dilution rate. Therefore, it is not surprising that CP supplementation did not affect total or branched-chain VFA concentrations in the current study.

Our observation that NPN source did not affect total and branched-chain VFA agrees with the work of Bond and Rumsey (1973). In contrast, studies by Chicco et al. (1971) and Löest et al. (2001) noted increased total VFA with urea compared with biuret supplementation of ruminants consuming low-quality forage. However, Chicco et al. (1971) collected rumen fluid 2 h after supplementation. This may not have been sufficient time to accurately determine the influence of supplemental biuret and urea on ruminal fermentation. Past research has demonstrated that ruminal fermentation of biuret is delayed compared with urea (Fonnesbeck et al., 1975; Bartle et al., 1998); therefore, it is probable that collecting rumen fluid 2 h after dosing did not allow sufficient time for biuret hydrolysis and ruminal fermentation, which may explain the increased VFA for urea compared with biuret reported by Chicco et al. (1971). Löest et al. (2001) increased diet CP of steers from 5.5% without supplementation (low-quality forage only) to 10.3% with supplementation (urea or urea/biuret molasses blocks) and collected rumen fluid on d 3, 7, 14, and 21. Early work by Schröder and Gilchrist (1969) demonstrated that the number of days required to develop maximum biuretolytic activity was a function of the CP content of the basal diet. Their data suggested that peak activity was attained after approximately 71 d with a diet CP concentration of approximately 10.3%. Therefore, Löest et al. (2001) may not have allowed for a sufficient adaptation period to adequately determine the affect of biuret on ruminal fermentation. Furthermore, this was verified by their determination that no major adaptation to biuret occurred by d 21.

Supplementation intervals of 2 d or less have had little to no effect on VFA compared with daily supplementation (Hunt et al., 1989; Farmer et al., 2001). This coincides with the lack of a SF effect on VFA observed in the current study. In addition, our data suggest that CP supplements containing urea or biuret as the source of supplemental N can be expected to elicit similar effects on ruminal VFA when supplemented daily or every other day.

Implications

Daily and alternate-day supplementation of nonprotein nitrogen can be an effective means of providing supplemental nitrogen to ruminants consuming low-quality forage (<6% crude protein). Alternate-day supplementation of nonprotein nitrogen may provide ruminant livestock producers with a management alternative that may decrease crude protein supplementation costs and improve economic sustainability while maintaining performance similar to daily supplementation.

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