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PROTEIN SUPPLEMENTATION OF LOW-QUALITY COOL-SEASON AND WARM-SEASON FORAGES

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ABSTRACT: An in situ study (Exp. 1) was performed using four ruminally cannulated steers (473 ± 13 kg BW) in a completely randomized design to compare the in situ degradation parameters of 2 low-quality, cool-season (C3) forages (Meadow foxtail, 4.5% CP; Reed canarygrass, 2.6% CP) and a warm-season (C4) forage (tallgrass prairie, 5.1% CP). Incubation time points were 0, 2, 8, 12, 24, 48, and 96 h. Means were separated using protected LSD ($P \leq 0.05$). Also, a digestion study (Exp. 2) utilizing 6 ruminally cannulated steers (428 ± 12 kg BW) in a 6 × 5 incomplete Latin square design was conducted to compare CP supplementation of low-quality C3 and C4 forages. Treatments included Meadow foxtail (MF; 4.6% CP), Reed canarygrass (RC; 2.6% CP) and tallgrass prairie (TG; 5.2% CP) hays with and without supplemental CP. Experimental periods were 20 d and soybean meal (50.2% CP) was used as the source of supplemental CP. Hays were offered at 120% of the previous 5 d average intake. No differences in lag time ($P = 0.83$) or rate of disappearance ($P = 0.58$) were noted for NDF in Exp. 1. However, NDF effective degradability was greatest for MF (49%; $P \leq 0.05$) while TG (45%; $P \leq 0.05$) was greater than RC (36%). Rate of N disappearance was similar for MF and RC ($P > 0.05$) with both C3 forages greater than TG ($P \leq 0.05$). Also, RDP as a proportion of total CP was greatest in MF (62%; $P \leq 0.05$) with RC (54%) greater than TG (41%; $P \leq 0.05$). However, effective degradability of N was greatest for MF (79%; $P \leq 0.05$) and for TG (64%; $P \leq 0.05$) compared with RC (58%). In Exp. 2, hay and total DMI was increased with supplementation ($P < 0.01$), for C3 compared with C4 ($P < 0.01$) and for MF compared with both RC ($P < 0.01$) and TG ($P < 0.05$). Also, apparent total tract DM digestibility was increased with supplementation ($P = 0.03$) while not affected by forage type ($P = 0.34$) but was greater for MF compared with both RC ($P < 0.01$) and TG ($P = 0.02$). These data indicate that low-quality C3 forages have greater RDP and N degradation rates than low-quality C4 forages. In addition, our data suggests that MF is better forage than RC and TG for beef cattle based on increased DMI and digestibility.

Key words: cattle, cool-season, forage, protein, supplementation, warm season

Introduction

Beef cattle in the Intermountain West normally consume low-quality cool-season (C3) forages (< 7% CP) for extended periods during the annual production cycle (Turner and DelCurto, 1992). In an effort to meet the nutritional needs of these animals, supplemental CP is often provided because it has been shown to increase forage OM intake (Lintzenich et al., 1995), forage DMD (DelCurto, 1990), and animal performance (Bodine et al., 2001). However, research suggests that CP supplementation of ruminants consuming low-quality C3 forages does not increase forage DMI in a manner similar to that observed with warm-season (C4) forages (Mathis et al., 2000; Bohnert et al., 2002; 2011). Therefore, the objective of this experiment was to compare DMI, digestibility, and ruminal fermentation of ruminants offered low-quality C4 (tall grass-prairie hay) and C3 hays (Meadow foxtail and Reed canarygrass) with and without supplemental CP in the hope of elucidating the reason(s) for the apparent difference in forage intake response.

Materials and Methods

All experimental procedures used in this study were approved by the Oregon State University Institutional Animal Care and Use Committee (ACUP# 4256).

Experiment 1. In Situ Degradation of C3 and a C4 low-quality forages

Four ruminally cannulated Angus × Hereford steers (473 ± 13 kg BW) were used in a completely randomized design to evaluate the ruminal degradation characteristics of 2 low-quality, C3 forages (Meadow foxtail; MF; Reed canarygrass; RC) and a C4 forage (tallgrass prairie; TG; Table 1). Steers had ad libitum access to low-quality meadow hay (6.5% CP; DM basis). A full description of meadow hay has been provided by Wenick et al. (2008). Steers were offered the low-quality meadow hay diet for at least 90 d prior to the start of Exp. 1.

Dacron bags (10 × 20 cm; Ankom Technology Corp., Fairport, NY) were labeled with a waterproof permanent marker, weighed, and 4 g (air equilibrated) of ground (2-mm; Wiley Mill; Model 4; Arthur H. Thomas, Philadelphia, PA) forage were added and the bags sealed with an impulse sealer (TISH-200; TEW Electric Heating Equipment CO., LTD, Taipei, Taiwan). Triplicate bags for each forage source were placed in a bucket containing warm water (39°C) and introduced into the rumen within 5 min. Bags were placed in a weighted polyester mesh bag within the rumen of each steer (0, 2, 8, 12, 24, 48, and 96 h) in reverse order, allowing all bags to be removed simultaneously. Three blank Dacron bags were incubated for 96 h and used to correct for microbial and feed contamination. Upon removal, Dacron bags were rinsed under tap water until the effluent was clear and dried at 55°C for 24 h. The dried triplicates were allowed to air equilibrate for 24 h at room temperature, weighed for residual DM, composited by steer, time and forage type, and analyzed for NDF (Robertson and Van Soest, 1981) using procedures modified for use in an Ankom 200 Fiber Analyzer (Ankom Technology Corp.). The NDF residue was then weighed and analyzed for N (Leco CN-2000; Leco Corp., St. Joseph, MI).

Kinetic variables for DM, NDF, and N digestibility were estimated with SAS (SAS Inst., Inc., Cary NC) using the modified nonlinear regression procedure described by Fadel (2004). Effective degradability of NDF and N was determined as described by Hoffman et al. (1993) using a ruminal passage rate of 2%/h (Mass et al., 1999). Rumen degradable protein (**RDP**) was calculated as described by Ørskov and McDonald (1979) with rumen undegradable protein (**RUP**) calculated as 1- RDP. Data were analyzed using the MIXED procedure of SAS. The model included forage source as the independent variable. Steer was used as random variable. Means were separated using LSD protected by a significant F-test ($P \leq 0.05$).

Experiment 2. Forage intake and nutrient digestibility of C3 and a C4 low-quality forages with and without supplemental CP

Six ruminally cannulated steers (428 ± 12 kg BW) were used in an incomplete 6 × 5 Latin square design (Cochran and Cox, 1957) and housed in individual pens (4 x 4 m) within an enclosed barn with continuous lighting. Steers were provided continuous access to fresh water and the 2 low-quality C3 forages and the single C4 forage used in Experiment 1; Table 1). Forage was provided daily (0700) at 120% of the average intake for the previous 5 d, with feed refusals from the previous day determined before feeding. A trace mineralized salt mix was provided daily. In addition, an intramuscular injection of vitamins A, D, and E was administered to each steer at the onset of the trial to safeguard against deficiency. Treatments were arranged in a 3 × 2 factorial design (3 forages with or without supplemental CP). Soybean meal (**SBM**) was placed directly into the rumen via the ruminal cannula for supplemented treatments. The amount of CP supplied by SBM was 0.11% of BW/d. The supplemented treatments were formulated, based on preliminary forage and SBM samples, to provide approximately 100% of the estimated DIP requirement assuming a microbial efficiency of 10%.

Experimental periods were 20 d, with intake measured beginning d 13 and concluding d 18. On d 14, treatment effects on ruminal DM, indigestible ADF (**IADF**), and fluid contents were determined by manually removing the contents from each steer's reticulo-rumen 4 h after feeding. The total ruminal contents were weighed, mixed by hand, and sub-sampled in triplicate (approximately 400 g). The remaining ruminal contents were immediately replaced into the animal. 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 steer.

Samples of forages and SBM were collected d 13 through d18 and orts were collected on d 14 through 19. Forages, SBM, and orts were dried at 55°C for 48 h and ground in a Wiley mill (1-mm screen). On d 15 through 20, fecal grab samples were collected 2 times/day at 12-h intervals with a 2-h increment added between days to shift sampling times. This allowed sampling on every even hour of the 24-h day. Fecal subsamples (200 g) were composited by steer, stored (-20°C), dried at 55°C for 96 h, and ground as described above.

On d 20, each steer was intra-ruminally pulse-dosed with 5 g of Co-EDTA in a 150-ml aqueous solution. The Co marker was administered throughout the rumen by injecting through a stainless steel probe with a perforated tip. Ruminal fluid (approximately 100 mL) was collected by suction strainer immediately prior to dosing and at 3, 6, 9, 12, 18, and 24 h post-dosing. Ruminal fluid pH was measured immediately after collection. Twenty milliliters was stored (-20°C) for later analysis of Co concentration and 5 mL was acidified with 1 mL of 25% (wt/vol) meta-phosphoric acid and stored (-20°C) for subsequent analysis of VFA and NH3-N. Frozen (-20°C) ruminal samples were prepared for analysis by thawing, centrifuging, and collecting the supernatant. Cobalt was analyzed by atomic absorption using an air/acetylene flame.

Ground samples were analyzed for DM and OM (AOAC, 1990), N (Leco CN-2000), 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. Also, samples were analyzed for IADF as described by Bohnert et al. (2002).

Data were analyzed as an incomplete 6 × 5 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 random variable. Contrasts used were: 1) supplemented vs not supplemented; 2) C3 vs C4; 3) contrast 1 × contrast 2; 4) meadow foxtail vs reed canarygrass; 5) meadow foxtail vs tall grass prairie.

Results and Discussion

We had a wider range in forage CP than anticipated for the hays used in Experiments 1 and 2, with the greatest values occurring with TG (5.1 and 5.2%, respectively) and the least occurring with RC (2.6 in both experiments; Table 1).

Experiment 1

The A fraction (total pool disappearing at a rate to rapid to measure) of NDF was greater for MF compared with RC and TG ($P \leq 0.05$) with no difference noted between RC and TG ($P > 0.05$; Table 2). The B fraction (degradable fraction disappearing at a measurable rate) was comparable between MF and TG ($P > 0.05$) while the proportion of degradable NDF in RC was approximately 22% less than TG and MF ($P \leq 0.05$). The undegradable fraction of NDF (C fraction) was approximately 70% greater ($P \leq 0.05$) for RC compared with TG and MF. Also, though no differences ($P = 0.58$) were noted in the rate of NDF degradation, the effective degradability of NDF was greatest ($P \leq 0.05$) for MF (48.7%) followed by TG (45.2%) which was more digestible than RC (35.6%; $P \leq 0.05$).

The A fraction of N was greatest with RC ($P \leq 0.05$) followed by MF. Also, both C3 forages had a greater A fraction than TG ($P \leq 0.05$; Table 2). However, when evaluating the B fraction, MF and TG were similar in ruminal degradable N ($P > 0.05$) but greater than RC ($P \leq 0.05$). Consequently, the undegradable N fraction was greatest for RC ($P \leq 0.05$) while TG was greater than MF ($P \leq 0.05$). The rate of N degradation was similar for the C3 forages ($P > 0.05$) which were almost 75% greater than that observed with the C4 forage ($P \leq 0.05$). This agrees with work by Bohnert et al. (2011) in which the N degradation rate of TG was almost 70% less than that observed with Kentucky bluegrass straw (C3; *Poa pratensis*). The proportion of RDP, as well as the effective degradability of N, was greatest for MF ($P \leq 0.05$). Also, RC contained a greater proportion of RDP than TG ($P \leq 0.05$). This agrees with a results reported by Bohnert et al. (2011) in which a low-quality C3 forage had approximately 28% greater RDP than a C4 forage with comparable CP concentration. The effective degradability of N was greater for TG compared with RC ($P \leq 0.05$).

Experiment 2

Hay and total DMI were increased with supplementation ($P < 0.01$; Table 3) and were greater for the C3 forages compared with the C4 ($P < 0.01$). These results agree with Bohnert et al. (2011) who reported similar results when comparing CP supplementation of TG with Kentucky bluegrass straw. However, in contrast to Bohnert et al. (2011), we did not note a supplementation × forage type interaction for hay intake ($P = 0.65$). Also, MF had greater hay and total DMI than RC ($P < 0.01$) or TG ($P < 0.01$). The differences between the current study and Bohnert et al. (2011) are probably due to differing nutritional quality profiles of the forages and/or the different C3 forage species used.

Apparent GIT digestibility of DM and N was increased with supplementation ($P \leq 0.03$; Table 3) but not affected by forage type ($P \geq .34$). However, digestibility of DM and N was greater with MF compared to RC ($P < 0.01$) and digestibility of DM was greater ($P = 0.02$) and N digestibility tended to be greater ($P = 0.06$) for MF compared with TG.

Ruminal IADF fill was not influenced by supplementation ($P = 0.11$; Table 3) but was decreased for MF compared to RC ($P < 0.01$) and tended to be less compared with TG ($P = 0.08$). However, supplementation increased both ruminal passage rate ($P < 0.01$) and outflow rate ($P < 0.01$) of IADF with no influence of forage type ($P \geq 0.80$) or for MF compared with RC ($P \geq 0.21$) and TG ($P \geq 0.38$).

Implications

The data reported here adds to the growing body of evidence that intake of low-quality C3 forages by ruminants is greater than intake of C4 forages. However, it is not evident what specific nutritional quality factors are causing the increased forage intake with C3 compared with C4. Consequently, further research is warranted to help ascertain the indices that will assist nutritionists to better predict forage intake of ruminants consuming low-quality forages. Also, our data indicates that MF is better forage than RC and TG for beef cattle based on increased DMI and digestibility.

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Table 1. Feedstuff[a] nutrient content (DM basis)

Nutrient,%	MF	RC	TG	SBM
Exp. 1				
CP	4.6	2.6	5.1	--
NDF	63.6	68.9	77.7	--
Exp. 2				
CP	4.6	2.6	5.2	50.2
NDF	64.1	69.0	77.1	16.1
ADF	33.6	41.5	42.0	5.6
IADF	21.3	32.8	28.6	0.0

[a] MF = Meadow foxtail hay (cool-season forage); RC = Reed Canarygrass hay (cool-season forage); TG = tallgrass prairie hay (warm-season forage); SBM = soybean meal.

Table 2. Ruminal degradation parameters of two cool-season forages (meadow foxtail and reed canarygrass) and one warm season forage (tallgrass prairie).

Degradation Parameters	Meadow Foxtail	Reed Canarygrass	Tallgrass Prairie	SEM[a]	P-Value
NDF					
Fractions, %[b]					
A	7.21[x]	4.51[y]	4.74[y]	0.41	0.006
B	68.7[x]	55.0[y]	72.1[x]	3.5	0.03
C	24.2[x]	40.6[y]	23.1[x]	3.29	0.02
Kd[c], /h	0.031	0.028	0.026	0.0043	0.58
Effective Degradability, %[d]	48.7[x]	35.6[y]	45.2[z]	0.98	< 0.001
N					
Fractions, %					
A	28.2[x]	36.4[y]	11.7[z]	0.53	< 0 .001
B	51.1[x]	21.8[y]	52.2[x]	1.41	< 0.001
C	20.7[x]	41.9[y]	36.2[z]	1.23	< 0.001
Kd[c], /h	.0613[x]	.0738[x]	.0388[y]	0.0078	0.02
RDP[e], % of CP	62.4[x]	54.2[y]	41.4[z]	1.03	< 0.001
RUP[f]	37.6[x]	45.8[y]	58.6[z]	1.03	< 0.001
Effective Degradability, %[d]	79.3[x]	58.1[y]	63.8[z]	1.23	< 0.001

[a] n = 4.
[b] A = soluble fraction (total pool disappearing at a rate too rapid to measure); B = degradable fraction (total pool disappearing at a measurable rate); C = undegradable fraction (total pool unavailable in the rumen).
[c] Fractional rate constant.
[d] Calculated as A + {B × [(*K*d/(*K*d + *K*p)]}, where *K*p was the ruminal passage rate, which was set at 2%/h (Hoffman et al., 1993). The units used for *K*d in the equation were per hour.
[e] Calculated as described by Ørskov and McDonald (1979).
[f] Calculated as 1 − RDP.
[x,y,z] Means in a row without a common superscript are different ($P < 0.05$).

Table 3. Intake, digestibility, and ruminal IADF dynamics by beef steers consuming low-quality cool-season (C3; Meadow foxtail; Reed canarygrass) and warm-season (C4; tallgrass prairie) grass hays with or without soybean meal (CP) supplementation

								P-Value[b]				
Item	MF	MF+	RC	RC+	TG	TG+	SEM[a]	Con vs Supp.	C3 vs C4	Supp. × Type	MF vs RC	MF vs TG
Intake, g/kg BW												
Hay DMI	17.4	22.5	14.8	20.0	14.6	19.2	0.724	< 0.001	0.004	0.65	< 0.001	< 0.001
Supplement DMI	0.00	1.09	0.00	1.09	0.00	1.09						
Total DMI	17.4	23.6	14.8	21.1	4.6	20.3	0.72	< 0.001	0.004	0.65	< 0.001	< 0.001
N	0.127	0.256	0.066	0.176	0.121	0.247	0.0061	< 0.001	< 0.001	0.44	< 0.001	0.17
NDF	11.3	14.8	10.4	14.1	11.5	15.2	0.572	< 0.001	0.13	0.92	0.16	0.52
IADF	4.0	5.0	4.3	5.8	3.8	5.2	0.46	0.002	0.52	0.83	0.21	0.94
Apparent GIT Digestibility, %												
DM	50.0	59.4	37.7	45.1	41.7	47.7	4.23	0.03	0.34	0.73	0.004	0.02
N	26.8	51.8	-22.5	33.2	11.4	36.9	7.86	< 0.001	0.79	0.27	< 0.001	0.06
NDF	46.2	56.6	36.6	36.9	47.9	48.6	6.80	0.50	0.48	0.70	0.04	0.65
ADF	36.8	45.0	26.7	29.1	38.1	37.0	8.71	0.66	0.68	0.68	0.15	0.70
Ruminal IADF												
Fill, g/kg BW	8.9	8.3	11.2	10.0	10.2	9.4	0.67	0.11	0.73	0.93	0.006	0.08
Passage rate, %/h	1.82	2.63	1.64	2.49	1.63	2.34	0.243	0.002	0.49	0.80	0.55	0.38
Outflow, (g/kg BW)/h	0.165	0.208	0.178	0.240	0.158	0.217	0.0192	0.002	0.52	0.83	0.21	0.94

[a] n = 5.

[b] Con vs Supp. = non-supplemented vs supplemented treatments; C3 vs C4 = cool-season vs warm-season forages; Supp. × Type = interaction of supplementation and forage type; MF vs RC = Meadow foxtail vs Reed canarygrass; MF vs TG = Meadow foxtail vs tallgrass prairie.