

Effects of monensin inclusion and level of intake in limit-feeding strategies for beef cows¹

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ABSTRACT: A study was conducted to evaluate the effects of intake management and ionophore inclusion on diet utilization under managed intake conditions in beef cattle. Two experiments utilized common diets fed at 120% (H) or 80% (L) of maintenance with either 0 or 200 mg/d monensin in a factorial arrangement. Forty cows were fed for 56 d (Exp. 1) to evaluate effects on diet utilization and energy retention; diets were fed to 16 ruminally cannulated steers (Exp. 2) to determine effects on digestion, energy value, and ruminal fermentation. Cows fed H had greater body weight (BW) gain ($P < 0.01$) and retained energy (RE; $P < 0.01$), although estimated heat production was also greater ($P < 0.01$). Monensin

had limited effects on overall BW gain ($P = 0.97$). Monensin had no effect on RE ($P = 0.94$) or calculated heat energy (HE; $P = 0.53$) despite effects on diet utilization observed in steers. In steers, L increased ($P < 0.01$) digestion of dry matter, organic matter, acid detergent fiber, and gross energy (GE) and reduced ($P < 0.01$) passage rate; monensin did not affect digestion ($P > 0.15$) but did reduce passage rate ($P < 0.03$). Monensin lowered ($P < 0.01$) acetate:propionate ratio and increased ($P < 0.05$) ruminal pH. Monensin did not alter feed required for maintenance; however, limit-feeding reduced apparent daily maintenance requirement to 62.85 kcal/kg BW^{0.75}, a 26% reduction from model-predicted values.

Key words: heat production, intensification, ionophore, maintenance requirement

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INTRODUCTION

Growth in global population and affluence are increasing global protein demand and competing for land resources. This competition, coupled with climatic variability, may limit forage availability for grazing cattle and thus limit the expansion of primary production to meet growing demand

(Steinfeld and Robinson, 2019). Shifting a portion of cow-calf production to more intensive management systems may allow the expansion of primary production in the face of limited forage supplies. Optimization of these systems is necessary if they are to be economically and environmentally resilient.

Managing the intake of more energy-dense diets has been shown to improve diet utilization and reduce the amount of feed required to maintain gestating beef cows (Loerch, 1996; Trubenbach et al., 2019). Cows fed diets below ad libitum intake levels exhibit a rapid (<7 d) decrease in heat production, followed by a longer

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but continued adaptation until a stable plane is achieved (Freetly et al., 2006). These adaptations suggest that feeding strategies may be used to reduce the maintenance requirements of beef cows (Trubenbach et al., 2019) and offer an opportunity to optimize management systems.

Ionophores improve the effective energy value of diets by altering the end products of fermentation (Richardson et al., 1976; Joyner et al., 1979) and protein utilization (Spears, 1990); these alterations may result in effective increases in diet NE_m of over 8% (Byers, 1980). In beef cows, monensin increased apparent feed values by 10–13% (Walker et al., 1980; Clanton et al., 1981). However, in pregnant heifers fed below ad libitum intake, monensin effects on energy balance (and thus apparent feed value) were less pronounced (Hemphill et al., 2018).

While the effects of intake restriction and ionophore consumption on energy utilization are well established, previous investigations typically have not considered their combined effects in limit-fed systems. We hypothesize that the effects of limiting the intake of a complete diet and those of feeding an ionophore are independent and, therefore, additive when these strategies are combined. The objective of these experiments was to test the hypotheses that limit-feeding and ionophore application will improve diet utilization and reduce feed requirements for the maintenance of pregnant cows.

MATERIALS AND METHODS

The experimental protocols were approved by the Agricultural Animal Care and Use Committee of Texas A&M Agrilife Research for research conducted at the McGregor Research Center or by the Institutional Animal Care and Use Committee at Texas A&M University for research conducted in College Station, TX.

Exp. 1: Cow Performance

Forty crossbred [three-fourth *Bos taurus*, one-fourth *Bos indicus*; body weight (BW) 385 ± 25 kg] cows in mid-gestation, 3 years of age, were used in an experiment designed to examine the effects of an ionophore (monensin; Rumensin 90, Elanco Animal Health, Indianapolis, IN) and dietary energy intake on cow maintenance and diet utilization. Cows were stratified by BW and four cows were assigned to each of 10 pens equipped with Calan-Broadbent feeders (American Calan, Northwood, NH) and

automatic waterers in a covered, open-sided barn. After a 14-d training period, cows were weighed, and this weight was used to determine individual maintenance requirements (NASEM, 2016) and subsequent feed amounts per individual.

Within each pen, cows were randomly assigned to receive a total mixed ration (TMR; Table 1) at one of two levels of NE_m intake, either 80% of calculated maintenance requirement (L; 51 g TMR/kg metabolic body weight [MBW]) or 120% maintenance requirement (H; 76 g TMR/kg MBW) without (0) or with monensin (200, 200 mg/d monensin) in a 2×2 factorial treatment arrangement. To ensure a constant dose of ionophore while allowing individual intake to vary according to treatment prescription, a supplement was constructed using dried distillers' grains with solubles (DDG) and monensin to contain 400 mg monensin per kilogram supplement. The corresponding amount (0.5 kg) of DDG was removed from the base TMR formulation on a percentage basis such that either 0.5 kg of the monensin containing supplement or 0.5 kg DDG was hand added to each animal's daily ration to achieve the final diet formulation and the appropriate dose of monensin (200 or 0 mg, respectively). Cows were fed individually at approximately 0730 h daily. Accumulated orts (if present) were collected weekly. Cows were allowed ad libitum access to freshwater from automatic fountain water troughs throughout the experiment.

At the beginning (day 0) and end of the feeding period (day 56), animals were subjected to a series of measurements, including hip height, heart girth, body condition score (BCS), and ultrasound measurements of rib fat thickness (between 12th

Table 1. Ingredient and nutrient composition of diet

Ingredient	With supplement	Without supplement
	% As fed	
Wheat straw	34.52	38.11
Corn	29.46	32.52
Distillers grain	27.46	19.92
Urea	1.10	1.21
Molasses	5.00	5.52
Mineral	2.46	2.72
Diet components ^a	DM basis ^b	
Crude protein, %	16.30	
Total digestible nutrients, %	68.00	
ME, Mcal/kg	2.45	
Net energy (NE_m), Mcal/kg	1.54	
Net energy (NE_g), Mcal/kg	0.95	

^aFormulated values.

^bDM content: 89.8%.

and 13th rib), rump fat thickness, and ribeye area. Ultrasound measurements were collected for both direct comparison and estimation of body energy reserves. Body weight was measured prior to feeding on days 0, 14, 28, 42, and 56. Fecal grab samples were collected and immediately frozen on days 14, 28, 42, and 56 for the determination of fecal production using acid detergent insoluble ash (ADIA) as an internal marker (Kanani et al., 2014).

Exp. 2: Intake, Digestion, Ruminal Fermentation, and Ruminal Fill

Sixteen ruminally cannulated Angus × Hereford steers (BW 288 ± 20 kg) were used in an experiment designed to examine the effects of monensin inclusion and dietary energy intake on measures of digestibility, ruminal pH, volatile fatty acid (VFA) concentrations and ruminal fill. Steers were stratified by BW and housed in individual stalls (2.1 × 1.5 m) in an enclosed, climate-controlled barn. Treatments were applied using the same randomized 2 × 2 factorial arrangement as Exp. 1, with four steers assigned to each treatment.

Daily intake rate was designed to correspond to cow intakes in Exp. 1 such that steers fed H received 76 g TMR/kg MBW and those fed L received 51 g TMR/kg MBW. The same supplementation protocol was utilized for the delivery of 0 or 200 mg/d monensin. Steers were fed daily at 0700 h. Orts (if present) were collected daily prior to feeding. Steers were allowed ad libitum access to fresh-water throughout the experiment.

The first 14 d of the experiment served as an adaptation to treatments. Feed and ort samples were collected on days 14–17 to correspond with fecal samples collected on days 15–18. Fecal samples were collected per rectum three times daily over the 4-d period. Fecal samples were collected every 8 h, with the sampling time advanced by 2 h each day such that samples were represented in 2-h intervals across 24 h.

Ruminal pH level and VFA concentration were measured on day 19. A sample of ruminal fluid was collected immediately prior to feeding (0 h) and also at 2, 4, 6, 9, 12, and 16 h after feeding using a suction strainer (Raun and Burroughs, 1962; 19-mm diameter, 1.5-mm mesh). Immediately following collection, the pH of each sample was determined using a portable pH meter with a combined electrode (VWR SympHony). Subsamples of ruminal fluid (8 mL) were combined with 2 mL of 25% *m*-phosphoric acid and immediately frozen at –20 °C for future VFA analysis.

On day 20, ruminal contents were collected via ruminal evacuation at 0.5 h prior to feeding and 4 h after feeding. Ruminal fill was estimated as the mean weight of ruminal contents at the two evacuation events. Ruminal contents were collected into barrels, weighed, and three subsamples collected per steer at each evacuation. Ruminal contents were returned immediately following sampling.

Laboratory Analyses

Samples of feed, ruminal contents, and feces were processed and analyzed using the same techniques for both experiments. Samples were dried at 55 °C in a forced-air oven for 96 h and allowed to air equilibrate for 24 h for the determination of partial dry matter (DM). Samples were then ground to pass through a 1-mm screen (No. 4 Wiley Mill, Thomas Scientific, Swedesboro NJ). Ground samples were dried at 105 °C for final DM determination. Organic matter (OM) was determined as the loss in DM weight following combustion for 8 h at 450 °C in a muffle furnace. Acid detergent fiber (ADF) analysis was performed using an Ankom Fiber Analyzer (Ankom Technology Corp., Macedon, NY) with sodium sulfite and amylase omitted and without correction for residual ash. Sample ADIA was measured as the remaining DM upon the combustion of ADF residue in a muffle furnace. The energy content of samples was determined by direct calorimetry using a Parr 6300 Calorimeter (Parr Instrument Company, Moline, IL). Samples of ruminal fluid were thawed and centrifuged at 20,000 × g for 20 min. Volatile fatty acid concentrations were measured using a gas chromatograph with methods described by Vanzant and Cochran (1994).

Calculations

Calculations followed those utilized by Trubenbach et al. (2019). Fecal production was calculated by dividing ADIA consumption by fecal ADIA concentration:

$$\text{Fecal production, kg} = \frac{\text{DMI} \times [\text{ADIA}_d]}{[\text{ADIA}_f]}$$

where:

DMI, kg

[ADIA_d] = Dietary ADIA concentration (%DM)

[ADIA_f] = Fecal ADIA concentration (%DM)

Digestibility of DM, OM, ADF, and gross energy (GE) was calculated as:

$$\text{Digestibility}_x, \% = \frac{\text{Intake}_x - \text{Fecal}_x}{\text{Intake}_x} \times 100\%$$

where:

$$\text{Intake}_x = \text{DMI (kg)} \times \text{dietary concentration of item } x \text{ (\% DM)}$$

$$\text{Fecal}_x = \text{Fecal production (kg)} \times \text{fecal concentration of item } x \text{ (\% DM)}$$

Measures of digestible energy (DE) and metabolizable energy (ME) were calculated as (NASEM, 2016):

$$\text{DE (Mcal/kg DM)} = \text{GE} \times \text{Digestibility}_{\text{GE}}$$

$$\text{ME (Mcal/kg DM)} = \text{DE} \times 0.82.$$

The maintenance level of intake for ME (ME_m) was estimated for both H and L using a linear regression of the means of retained energy (RE) on metabolizable energy intake (MEI). The linear functions representing each diet were solved for RE = 0; the solution of which represented ME_m for the respective diet.

Fasting heat production was estimated for H and L using the linear regression of the means of log (HE) on MEI. The linear functions representing each diet were solved for MEI = 0; the solution of which represented the estimate of fasting heat production (FHP) for each respective diet.

Body condition score was calculated at both the beginning and end of the trial using a regression equation derived from data presented by Herd and Sprott (1998):

$$\text{BCS} = -1.2927x^2 + 6.0916x + 2.2114$$

where:

$$x = \text{Rib fat thickness, cm}$$

Equations published in Nutrient Requirements of Beef Cattle (NASEM, 2016) were used to calculate empty body energy. Body composition was estimated using the following equations:

$$\text{AF} = 3.768 \times \text{BCS}$$

$$\text{AP} = 20.09 - 0.668 \times \text{BCS}$$

where:

$$\text{AF} = \text{proportion of empty body fat}$$

$$\text{AP} = \text{proportion of empty body protein}$$

Body components were calculated as:

$$\text{TF} = \text{AF} \times \text{EBW}$$

$$\text{TP} = \text{AP} \times \text{EBW}$$

$$\text{EBW} = \text{SBW} - \text{FL}$$

$$\text{FL} = \text{SBW} \times \alpha$$

$$\text{SBW} = \text{BW} \times 0.96$$

where:

$$\text{EBW} = \text{empty body weight,}$$

$$\text{TF} = \text{total fat, kg}$$

$$\text{TP} = \text{total protein, kg}$$

$$\text{FL} = \text{fill, kg}$$

α (% SBW) was estimated for each treatment based on rumen evacuation in Exp. 2.

Ruminal DM fill was calculated using:

$$\text{DM fill, kg} = \frac{\text{DM Fill}_0 + \text{DM Fill}_4}{2}$$

where:

$$\text{DM Fill}_0 = \text{Rumen evacuation DM contents before feeding}$$

$$\text{DM Fill}_4 = \text{Rumen evacuation DM contents 4 h after feeding}$$

Total body energy (TBE) was calculated as:

$$\text{TBE (Mcal)} = 9.4 \times \text{TF} + 5.7 \times \text{TP}$$

RE and HE were calculated as:

$$\text{RE} = \text{TBE}_f - \text{TBE}_i$$

$$\text{HE} = \text{ME} - \text{RE}$$

where:

$$\text{RE} = \text{retained energy, Mcal}$$

$$\text{TBE}_i = \text{total body energy on day 0, Mcal}$$

$$\text{TBE}_f = \text{total body energy on day 56, Mcal}$$

$$\text{HE} = \text{heat energy, Mcal}$$

$$\text{ME} = \text{metabolizable energy intake, Mcal.}$$

Molar proportions of VFA were calculated using:

$$\begin{aligned} \text{Molar proportion}_x, \% \\ = \frac{\text{Concentration}_x}{\text{VFA Concentration}} \times 100 \% \end{aligned}$$

where:

$$\text{Concentration}_x = \text{Individual VFA concentration (mM)}$$

$$\text{VFA Concentration} = \text{Sum of all concentration}_x \text{ (mM)}$$

Total ruminal VFA were calculated using:

$$\text{Total VFA}_x, \text{ mol} = \text{Concentration}_x \times \text{total ruminal liquid contents}$$

where:

$$\begin{aligned} &\text{Total ruminal liquid contents} \\ &= \text{Average ruminal contents (kg) DM fill (kg)} \end{aligned}$$

Statistical Analysis

Measures of digestibility, ultrasound measurements, RE, and HE were analyzed using the MIXED procedure in SAS 9.3 (SAS Institute Inc., Cary, NC). Model effects included diet, intake, and diet \times intake. Measures of VFA concentrations and ruminal fluid pH were analyzed as repeated measures using the MIXED procedure in SAS 9.3. The model effects included diet, intake, and time, with interactions of diet \times intake, diet \times time, intake \times time, and diet \times intake \times time. Animal nested within treatment served as the subject effect, and the covariance structure that minimized the Bayesian Information Criterion was selected.

RESULTS

Experiment 1

One cow from L0 was removed from the experiment and subsequent statistical analysis due to failure to accept training to the Calan gate system. No interactions were observed between intake level

and monensin inclusion ($P \geq 0.18$) for estimates of digestibility, dietary energy availability, or energy intake (Table 2) in cows. By design, DM intake (DMI), digestible OM, GE, DE, and ME were greater ($P < 0.01$) for cows fed H than those fed L. Also, by design, DMI and GE intake did not significantly differ between cows receiving 0 or 200 mg/d monensin ($P = 0.75$).

Monensin inclusion did not affect the intake of digestible OM, DE, or ME ($P \geq 0.44$; Table 4). Digestibility of DM was greater ($P < 0.01$) for cows fed L, with corresponding increases in OM and GE digestibility ($P < 0.01$); however, ADF digestibility was not affected by the intake level ($P = 0.66$).

There were no differences observed in the digestibility of DM, OM, ADF, or GE ($P \geq 0.18$) due to monensin inclusion. Observed values of DE and, therefore, ME (Mcal/kg DM) were greater ($P < 0.01$) in cows fed L compared to those fed H but were not affected ($P = 0.44$) by monensin inclusion.

Body weight (Table 3) did not differ ($P \geq 0.77$) between levels of intake or monensin inclusion prior to treatment application. Cows fed H had greater BW gain ($P < 0.01$) than those fed L over the 56-d period (18.0 vs. -4.7 kg, respectively) such that BW on day 56 was greater for cows fed H than for those fed L. Neither final BW nor BW gain differed ($P \geq 0.36$) due to monensin inclusion.

Table 2. Observed intake, nutrient digestibility, and energy availability in cows fed high and low intakes with two levels of monensin inclusion

Item	Low ^a		High		SEM	Probability ^b	
	0	200	0	200		Monensin	Intake
Number of observations	9	10	10	10			
Intake, kg/d							
DMI	3.54	3.49	5.12	5.22	0.081	0.75	<0.01
DOMI	2.24	2.21	3.10	3.23	0.064	0.45	<0.01
Digestibility, %							
DM	68.0	67.8	65.1	66.4	0.72	0.43	<0.01
OM	69.2	69.0	66.1	67.5	0.78	0.41	<0.01
ADF	53.8	51.2	52.2	51.7	1.20	0.18	0.66
GE	66.9	66.6	63.5	65.2	0.80	0.37	<0.01
Energy availability, Mcal/kg DM							
DE	2.92	2.91	2.77	2.84	0.040	0.44	<0.01
ME	2.40	2.39	2.27	2.33	0.033	0.44	<0.01
Energy intake, Mcal/d							
GE	15.55	15.33	22.41	22.85	0.370	0.75	<0.01
DE	10.40	10.22	14.24	14.91	0.319	0.44	<0.01
ME	8.53	8.38	11.68	12.23	0.262	0.44	<0.01

DOMI = digestible organic matter intake.

^aLow = received 80% NRC requirements; high = received 120% NRC requirements; 0 = 0 mg/d monensin inclusion; 200 = 200 mg/d monensin inclusion.

^bMonensin = effect of 0 vs. 200; intake = effect of low vs. high; no treatment interactions ($P > 0.18$).

Table 3. Body weight and ultrasound measurements of cows fed high and low intakes with two levels of monensin inclusion

Item	Low ^a		High		SEM	Probability ^b	
	0	200	0	200		Monensin	Intake
Number of observations	9	10	10	10			
Initial measurements							
Body weight, kg	430.6	419.4	422.4	433.3	9.79	0.99	0.77
Hip fat, mm	2.46	2.79	2.09	1.83	0.427	0.93	0.11
Back fat, mm	2.57	2.34	1.75	1.68	0.357	0.66	0.04
Ribeye area, cm ²	58.19	56.13	56.27	58.97	2.286	0.89	0.84
Final measurements							
Body weight, kg	426.2	413.3	445.0	451.1	10.48	0.74	<0.01
Hip fat, mm	2.24	2.69	2.91	2.74	0.489	0.76	0.45
Back fat, mm	2.67	2.26	2.23	2.39	0.461	0.78	0.73
Ribeye area, cm ²	63.10	56.63	63.71	62.06	2.388	0.08	0.18
Change in measurements							
Body weight, kg	-3.9	-5.4	20.1	15.9	3.21	0.36	<0.01
Hip fat, mm	-0.23	-0.10	0.82	0.91	0.406	0.77	0.01
Back fat, mm	0.10	-0.08	0.48	0.71	0.350	0.94	0.09
Ribeye area, cm ²	4.90	0.29	6.94	3.10	1.950	0.03	0.19

^aLow = received 80% NRC requirements; high = received 120% NRC requirements; 0 = 0 mg/d monensin inclusion; 200 = 200 mg/d monensin inclusion.

^bMonensin = effect of 0 vs. 200; intake = effect of low vs. high; no treatment interactions ($P > 0.24$).

Table 4. Estimates of RE^a and HE^b in cows fed high or low levels of intake relative to maintenance with two levels of monensin inclusion

Item	Low ^c		High		SE	P-value ^d	
	0	200	0	200		Monensin	Intake
Number of observations	9	10	10	10			
RE, kcal/d/EBW ^{0.75}	-1.7	-3.2	16.1	17.0	4.61	0.94	<0.01
HE, kcal/d/EBW ^{0.75}	103.2	105.5	121.2	125.4	4.86	0.49	<0.01

^akcal/d/EBW^{0.75}, calculated as RE/d/EBW^{0.75}, where d = 56 d.

^bkcal/d/EBW^{0.75}, calculated as (ME - RE)/d/EBW^{0.75}, where d = 56 d.

^cLow = received 80% NRC requirements; high = received 120% NRC requirements; 0 = 0 mg/d monensin inclusion; 200 = 200 mg/d monensin inclusion.

^dMonensin = effect of 0 vs. 200; intake = effect of low vs. high; no treatment interactions ($P > 0.23$).

No interactions between the level of intake and monensin inclusion were observed ($P \geq 0.24$) relative to ultrasound measurements collected on days 0 or 56. Likewise, no interactions were observed relative to changes in ultrasound measurements between days 0 and 56 ($P \geq 0.24$). Back fat thickness was greater in L than H on day 0 ($P = 0.04$), but no other differences in ultrasound measurements due to feeding level were observed ($P \geq 0.18$) on days 0 or 56. No differences in back fat thickness, hip fat thickness, or longissimus area due to monensin inclusion were observed on day 0 ($P \geq 0.66$). On day 56, no differences in hip fat or rib fat ($P \geq 0.76$) were observed between 0 and 200, although longissimus tended ($P = 0.08$) to be larger in 0 than 200.

Changes in hip fat thickness were greater ($P = 0.01$) and changes in back fat thickness tended to be greater

($P = 0.09$) in cows fed H than those fed L. It is important to note that changes in hip fat and back fat for cows fed H were positive and nonzero ($P < 0.05$), while, in cows fed L, these changes were not different from zero ($P \geq 0.45$). Longissimus area increased in both L and H ($P < 0.05$) over the 56-d period, but no differences in the change in longissimus area were observed due to the level of intake ($P = 0.19$).

No differences ($P \geq 0.77$) in the 56-d change in back fat thickness or hip fat thickness were detected between the 0 and 200 mg/d inclusion of monensin; none of these measures differed than zero. Change in longissimus area was greater ($P < 0.01$) in cows fed 0 compared to those fed 200 mg/d monensin.

No interactions were observed ($P \geq 0.65$) between the level of intake and monensin inclusion

for calculated RE (kcal/d/kg EBW^{0.75}). Retained energy was greater ($P < 0.01$; Table 4) in cows fed H compared to those fed L (16.57 vs. -2.48 kcal/d/kg EBW^{0.75}, respectively). No differences ($P \geq 0.23$) in RE were observed due to monensin inclusion.

No ($P \geq 0.23$) interactions between the level of intake and monensin inclusion were observed for the estimates of HE (kcal/d/kg EBW^{0.75}). Estimates of HE were 21.4% greater ($P < 0.01$) in cows fed H than those fed L. No differences in HE in response to monensin inclusion were observed ($P \geq 0.19$).

Experiment 2

There were no interactions observed between intake level and monensin inclusion ($P \geq 0.22$) for estimates of digestibility, dietary energy availability, energy intake, passage rate, or ruminal fill (Table 5). By design, the intake of DM, digestible OM, GE, DE, and ME were greater ($P < 0.01$) in steers fed H than those fed L, but were not affected by the inclusion of monensin ($P \geq 0.80$).

Passage rate was 28% slower ($P < 0.01$) for steers fed L compared to those fed H, and 12% slower ($P = 0.03$) for steers fed 200 compared to

0 mg/d monensin. Total DM in the rumen (expressed as total kilogram or as a percentage of BW) was greater in steers fed H ($P < 0.01$) than those fed L and was greater ($P = 0.05$) in steers fed 200 compared to 0 mg/d monensin.

Digestion of DM, OM, ADF, and GE was greater ($P < 0.01$) in steers fed L than those fed H. No differences were observed in the digestion of DM, OM, ADF, or GE ($P \geq 0.16$) due to monensin inclusion. Observed values of DE (and, therefore, ME) per unit of feed DM were greater ($P < 0.01$) for steers fed L compared to those fed H, but DE and ME values were not affected ($P = 0.74$) by monensin inclusion.

A monensin inclusion \times time ($P < 0.03$; Table 6) interaction was observed for the molar proportions of acetate, propionate, butyrate, isobutyrate, isovalerate, and valerate, as well as for the acetate to propionate ratio and ruminal pH. This interaction resulted from the variation in the magnitude of differences among 0 and 200 mg/d monensin inclusion rates of monensin at different times and not from a re-ranking of treatments (data not shown).

An intake level \times monensin \times time ($P = 0.03$) interaction was observed for molar proportion of acetate in ruminal fluid (data not shown). There were monensin \times time and intake level \times time

Table 5. Observed intakes, nutrient digestibility, energy availability, passage rate, and ruminal fill of steers fed high and low intakes with two levels of monensin inclusion

Item	Low ^a		High		SEM ^c	Probability ^b	
	0	200	0	200		Monensin	Intake
Intake, kg/d							
DMI	2.92	2.91	4.34	4.36	0.112	0.98	<0.01
DOMI	2.00	2.00	2.77	2.73	0.094	0.80	<0.01
Digestibility, %							
DM	74.2	74.4	69.7	68.5	1.18	0.64	<0.01
OM	76.7	76.9	71.9	70.8	1.11	0.69	<0.01
ADF	59.4	58.1	53.8	51.2	1.30	0.16	<0.01
GE	75.2	75.4	70.7	69.8	1.19	0.74	<0.01
Energy availability, Mcal/kg DM							
DE	2.90	2.90	2.70	2.66	0.052	0.74	<0.01
ME	2.38	2.38	2.21	2.18	0.043	0.74	<0.01
Energy intake, Mcal/d							
GE	11.66	11.64	17.14	17.21	0.438	0.96	<0.01
DE	8.49	8.49	11.63	11.49	0.408	0.86	<0.01
ME	6.96	6.96	9.54	9.42	0.335	0.86	<0.01
Passage rate, %/h	1.88	1.51	2.44	2.29	0.107	0.03	<0.01
Ruminal DM fill, kg	3.51	4.18	4.54	4.90	0.236	0.05	<0.01
Ruminal DM fill, % of BW	1.15	1.39	1.54	1.64	0.053	<0.01	<0.01

DOMI = digestible organic matter intake.

^aLow = received 80% NRC requirements; high = received 120% NRC requirements; 0 = 0 mg/d monensin inclusion; 200 = 200 mg/d monensin inclusion.

^bMonensin = effect of 0 vs. 200; intake = effect of low vs. high; no treatment interactions ($P > 0.22$).

^c $n = 4$.

Table 6. Rumen pH and VFA profile of steers fed high and low intakes with two levels of monensin inclusion

Item	Low ^a		High		SEM ^c	Probability ^b					
	0	200	0	200		Monensin	Intake	Time	M × I	M × T	I × T
Ruminal pH	6.4	6.58	6.27	6.39	0.054	0.02	0.01	<0.01	0.62	0.13	<0.01
Molar proportion											
Acetate	63.87	59.74	65.66	58.01	0.863	<0.01	0.94	<0.01	0.07	0.03	0.07
Propionate	21.20	24.02	21.02	26.05	0.921	<0.01	0.33	<0.01	0.25	<0.01	0.14
Butyrate	10.84	11.09	9.62	11.31	0.653	0.16	0.46	<0.01	0.29	0.01	0.24
Isobutyrate	1.22	1.54	1.08	1.38	0.036	<0.01	<0.01	<0.01	0.70	<0.01	0.01
Isovalerate	1.88	2.64	1.64	2.13	0.183	<0.01	0.06	<0.01	0.48	<0.01	<0.01
Valerate	0.99	0.96	0.98	1.04	0.047	0.76	0.49	<0.01	0.36	<0.01	0.07
Acetate:propionate	3.04	2.51	3.16	2.28	0.136	<0.01	0.71	<0.01	0.21	<0.01	0.04
Concentration, mM											
Acetate	52.07	41.89	57.01	44.93	1.589	<0.01	0.03	<0.01	0.56	<0.01	<0.01
Propionate	17.32	16.82	18.28	20.38	1.019	0.45	0.05	<0.01	0.23	<0.01	<0.01
Total VFA	81.67	69.98	86.79	77.54	2.595	<0.01	0.03	<0.01	0.65	<0.01	<0.01

^aLow = received 80% NRC requirements; high = received 120% NRC requirements; 0 = 0 mg/d monensin inclusion; 200 = 200 mg/d monensin inclusion.

^bMonensin = effect of 0 vs. 200; intake = effect of low vs. high; M × I × T interaction for acetate proportion ($P = 0.03$); all others ($P > 0.10$).

^c $n = 4$.

($P < 0.01$) interactions for acetate, propionate, and total VFA concentrations (data not shown). In all cases, interactions resulted from changes in the magnitude of differences between treatments at different times rather than a re-ranking of treatments. Across sampling times (Table 6), acetate concentration was lower ($P = 0.03$) in steers fed L compared to those fed H and was also lower ($P < 0.01$) for steers receiving 200 compared to 0 mg/d monensin. Propionate concentration was lower ($P = 0.05$) in steers fed L compared to those fed H but was unaffected ($P = 0.45$) by monensin inclusion. Total VFA concentration was lower ($P = 0.03$) for steers fed L than H, and lower ($P < 0.01$) for steers receiving 200 than for those receiving 0 mg/d monensin.

DISCUSSION

Our objective was to test the hypotheses that limit-feeding and ionophore inclusion improve diet digestion and reduce maintenance requirements in pregnant cows.

Effects of Intake

Restricting intake (L) increased the digestion of DM, OM, and GE in both experiments. This effect is likely related to reduced passage rate as observed in steers fed L compared to H in Exp. 2 (Baldwin et al., 1977; Bull et al., 1979). The improved digestibility of GE observed in both experiments with a reduced level of feed intake resulted in increased dietary DE and ME concentration for cattle fed L compared to

H. While the magnitude in improvement in apparent dietary energy availability from feeding at 80% of maintenance was greater in Exp. 2 (8% increase in DE) than in Exp. 1 (4% increase), the observed DE and ME values are lower than formulated targets based on NASEM (2016) tabular values (2.45 Mcal ME/kg DM), likely due to the variation in individual ingredients from tabular means. The energy values observed in these studies are slightly higher than those observed by Trubenbach et al. (2019) for similar diets and appear to result from greater GE digestion in the current study.

Importantly, the reductions in energy availability resulting from reduced digestion when feeding at a higher intake level imply that the diet provides less NE_m /kg for cows fed H (mean of 8.5% below formulated target) than those fed L (3.1% below formulated target). Changes in dietary energy utilization that result from changes in digestion might be important considerations when feeding beef cows diets designed to be fed below ad libitum consumption, as dietary energy value is consistently modified by the level of intake (Trubenbach et al., 2019).

Positive cow BW change and increases in ultrasound measurements of fat depots were expected in cows fed H, as they were fed to exceed estimated maintenance requirements. Cows fed L lost modest amounts of BW, but changes in fat measures via ultrasound were not different from zero. Body weight and ultrasound measures were used to estimate body energy content according to accepted equations (NASEM, 2016). For cows fed above maintenance

(i.e., cows fed H in the current study), RE is expected to be positive (Table 4). Although the L feeding level was designed to supply NE_m at 80% of NASEM (2016) requirements, the observed changes in BW and ultrasound measures of fat deposits resulted in estimates of RE that were not different from zero, suggesting that energy intake approximated maintenance requirements. The apparent achievement of maintenance at L intake suggests either greater efficiency of ME utilization for maintenance when intake is less than expected maintenance requirement (Byers, 1980) or adaptation to the constrained plane of nutrition such that a new maintenance equilibrium may have been reached (Freetly and Nienaber, 1998; Trubenbach et al., 2019).

Reducing intake from H to L (i.e., a 29% reduction in ME intake) reduced estimated heat production by 16%. When Freetly et al. (2006) imposed a 26% reduction in DM intake per unit of $BW^{0.75}$, they observed a 15% decrease in heat production occurring within 7 d of restriction and an 18% reduction 56 d after restriction. Trubenbach et al. (2019) observed a 22% reduction in heat production feeding a similar diet at levels similar to those in the present study. These results are consistent with observed patterns of BW change in the present study and support the hypothesis that rapid adjustments in maintenance energy demand occurs in cows fed below model-predicted requirements. The consistency of this response suggests that an adjustment to current models should be made to better predict outcomes related to limit-feeding of cows.

Effects of Monensin

Ionophores are known to alter the VFA profile and improve the capture of feed energy during ruminal fermentation (Richardson et al., 1976). Accordingly, NRC (2000) recommends increasing NE_m values by 12% with the inclusion of an ionophore, and NASEM (2016) suggests a more moderate 2.3% increase in diet ME value when monensin is fed. Based on these recommendations, it was hypothesized that feeding cows monensin would increase RE for either intake level, as increasing the effective NE_m (or ME) value of the diet should have either minimized the energy deficit and spared body tissue loss (L200 vs. L0) or reduced feed used for maintenance and thus increased energy available for retention (H200 vs. H0). However, these effects were not observed in the current study; inclusion of monensin did not affect cow BW change over the 56-d trial, nor did it affect measures of fat

deposition or RE. Linneen et al. (2015) did not observe changes in cow BW or condition score due to monensin supplementation in beef cows, while Walker et al. (1980) reported 3.4% to 11.7% decreases in feed required for BW stasis in pregnant cows fed monensin, suggesting improved diet utilization or energy yield.

If monensin increased effective NE_m by 12% as suggested by NRC (2000), cows on L200 would have theoretically been receiving 89.6% of maintenance requirements rather than the 80% that L0 received and would be expected to exhibit differential rates of BW and RE change; these effects were not observed. While adaptation to energy restriction in cows may occur very rapidly (7 d; Freetly et al. 2006; Trubenbach et al., 2019), the rate and magnitude of adaptation may be related to the severity of the restriction. If so, improving effective dietary energy value with monensin may have resulted in slower adaptation, obviating the energetic advantage and resulting in similar BW change.

In spite of minimal apparent effects of monensin inclusion on cow RE, the results of Exp. 2 are consistent with the well-documented effects of monensin inclusion on ruminal fermentation (Richardson et al., 1976; Lemenager et al., 1978a); monensin reduced molar proportion of acetate and increased molar proportion of propionate, reducing acetate:propionate ratio. However, the reductions in acetate production resulted in lower total VFA concentration in cows fed monensin such that any efficiency gains due to altered VFA profile may have been offset by a reduction in total VFA yield. Lemenager et al. (1978b) reported that total VFA concentrations were decreased with the inclusion of 200 mg/d monensin, but others have shown no change in total VFA concentrations (Dinius et al., 1976).

Reported increases in NE_m from monensin are primarily attributed to alterations in fermentation products rather than large changes in digestibility (Spears, 1990). The present study found no increase in digestibility from monensin inclusion, although the rate of passage was decreased, which has been reported in steers limit-fed a high-concentrate diet (Lemenager et al., 1978b), perhaps due to a decrease in the number of daily ruminal contractions (Deswysen et al., 1987). Reported monensin effects on diet digestibility are varied. There may be minimal effects on ADF digestion (Bell et al., 2017; Benz and Johnson, 1982) or on DM digestion once adaptation has occurred (Poos et al., 1979). Others report that monensin increased DM digestion in grain-fed (Dinius et al., 1976) and grazing cattle

(Pond and Ellis, 1981), but these effects may result from reductions in the intake and corresponding changes in passage rate. Because cattle in the present study were limit-fed, effects of monensin on digestibility may have been muted as monensin was not allowed to exert an independent effect on voluntary intake.

Using mean MEI and RE data, estimated ME_m values were calculated by regressing RE on MEI and solving for $RE = 0$ (Fig. 1). The resulting ME_m values were estimated to be 102 and 105 kcal/EBW^{0.75}/d, respectively, for 0 and 200. These values are very similar to the 104 ME_m kcal/EBW^{0.75}/d reported by Freetly and Nienaber (1998)

when intake was restricted to 65% of maintenance requirements. Although values from the current study are greater than the 93 kcal/EBW^{0.75}/d reported by Trubenbach et al. (2019) in a similar study (likely due to higher digestibility estimates in the current study), they are 35% lower than the 160 kcal/EBW^{0.75}/d calculated from NASEM (2016), suggesting that standard equations overestimate ME_m required in cows limit-fed moderately high-concentrate diets.

The lack of an observed difference in heat production from the inclusion of monensin was expected. Thornton and Owens (1981) reported no significant differences in heat production from the inclusion of

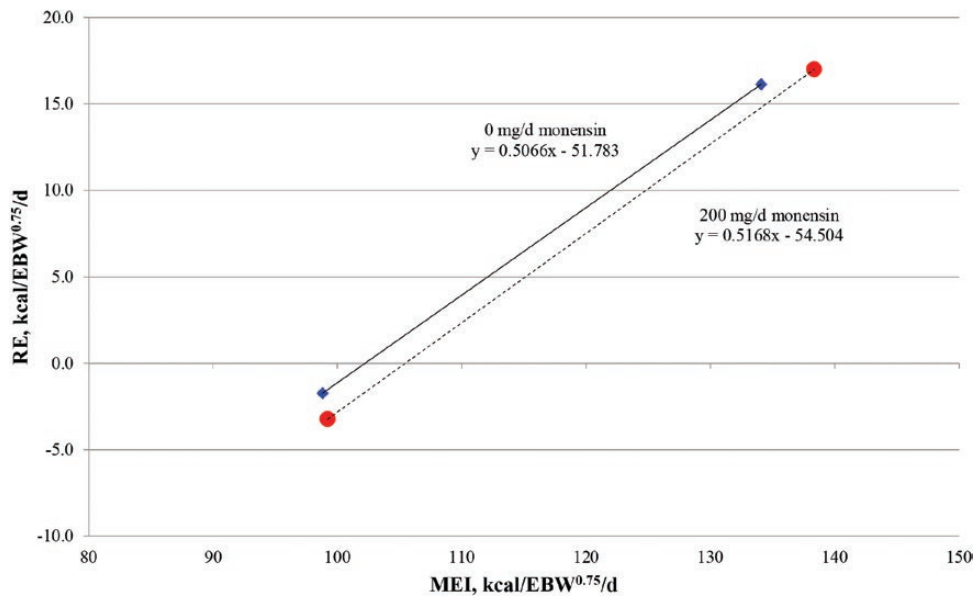


Figure 1. The effect of MEI on RE in cows fed two levels of monensin. 0 = received 0 mg/d monensin; 200 = received 200 mg/d monensin.

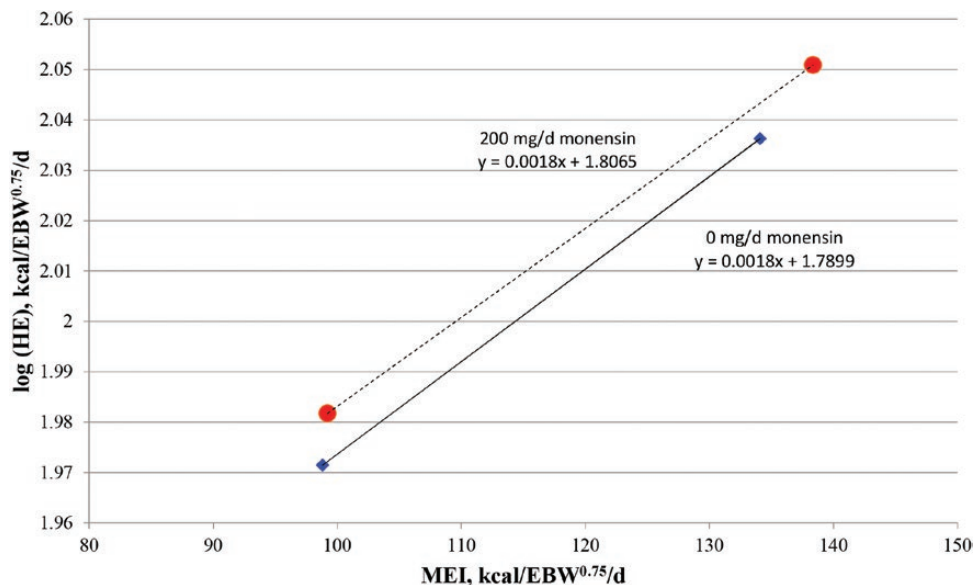


Figure 2. Logarithmic transformation of the effect of MEI on HE in control cows or fed monensin. 0 = received 0 mg/d monensin; 200 = received 200 mg/d monensin.

monensin in finishing steers, and Byers (1980) suggests that monensin may change the efficiency of energy use for maintenance, but this may be manifest as changes in diet energy value rather than requirement.

To account for the nonlinearity of heat production as MEI increases, FHP was estimated by regressing log (HE) on MEI (Fig. 2; Byers, 1980). Although estimates for 0 and 200 were not different (62 and 64 kcal/EBW^{0.75}/d, respectively), these figures are an 18.4% reduction from the 77 kcal/EBW^{0.75}/d considered the base estimate for FHP in beef cattle (NASEM, 2016), and a 26.1% reduction from the 85 kcal/EBW^{0.75}/d estimated for pregnant cows 8 months past calving, similar to cows in our study.

The results of these experiments suggest that limiting the intake of moderate energy density diets improves the efficiency of diet utilization in beef cows. Cows adapt to these strategies by reducing apparent maintenance energy demand such that the combination of improved diet utilization and cow adaptation may result in lower amounts of feed required for maintenance under limit-feeding scenarios than predicted by models with static estimates of NE_m required (Ferrell and Oltjen, 2008). The use of ionophores in conjunction with limit-feeding conferred expected outcomes for measures of ruminal fermentation and may have altered cow adaptation to restriction below targeted maintenance, but these effects were not pronounced in overall outcomes. Further exploration of the role of ionophores, such as the timing of application, may be warranted. Adjustments to standard nutritional models or adoption of new models that incorporate enhanced diet utilization and dynamic maintenance requirements for cows fed in these systems may allow the developing of sustainable intensification strategies for beef cow management by reducing the proportion of total energy consumed in production systems that is allocated to the maintenance of the cowherd.

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