

# Effect of metabolizable protein intake on growth performance, carcass characteristics, and feeding behavior in finishing steers

Leonardo G. Sitorski, Marc L. Bauer, and Kendall C. Swanson<sup>1,\*</sup>

Animal Sciences Department, North Dakota State University, Fargo, ND

**ABSTRACT:** One-hundred thirty-two finishing steers ( $300 \pm 2.7$  kg body weight [BW]) predominately of Angus, Simmental, and Shorthorn breeding were used to study the effect of metabolizable protein (MP) intake on growth performance, carcass characteristics, and feeding behavior. Steers were stratified by initial BW across five pens and randomly assigned to one of four dietary treatments to supply an average of 626, 906, 1,209, and 1,444 g MP/d ( $n = 33$  per treatment). Feed intake and feeding behavior were measured using radio frequency identification tags and the Insentec feeding system. For feeding behavior, a visit was defined as each time the Insentec system detected a steer at the feed bunk. A meal was defined as eating periods by intervals no longer than 7 min. Steers were fed until they reached an average BW of  $598 \pm 3.1$  kg. Average daily gain (ADG) responded quadratically ( $P < 0.01$ ) with ADG increasing in steers fed 906 g MP/d and plateauing thereafter. Dry-matter intake (DMI; kg) responded quadratically ( $P = 0.009$ ) with DMI increasing with MP intake up to 1,209 g/d MP and decreasing thereafter. Gain to feed ratio (G:F) increased linearly ( $P = 0.04$ ) and tended ( $P = 0.10$ ) to respond quadratically, as G:F increased up to

906 g MP/d and plateaued thereafter. A quadratic response ( $P = 0.04$  and  $P = 0.02$ , respectively) was observed for marbling score and 12th rib subcutaneous fat thickness with steers fed 1,209 g MP/d having the greatest marbling score and back fat thickness. A quadratic effect for visits and meals per day was observed ( $P < 0.01$ ) with steers fed the 1,209 g MP/d treatment having the least visits and meals per day. In addition, time eating per visit responded quadratically ( $P = 0.05$ ) with time increasing from 626 to 906 g MP/d. There was a linear increase ( $P \leq 0.02$ ) in time eating per meal and per day with increasing MP intake. A quadratic effect ( $P < 0.03$ ) was observed for DMI per visit, meal, and minute with steers fed 1,209 g MP/d having the greatest DMI. In summary, steers fed 626 g MP/d had increased visits and meals per day. However, DMI per visit, meal, and minute were greater in steers fed 1,209 g MP/d. A day  $\times$  treatment interaction ( $P < 0.001$ ) was observed for plasma urea N as concentrations increased to a greater extent over time in the higher MP treatments than in the lower MP treatments. These data indicate that MP supply (from deficient to excess) influences growth performance, carcass characteristics, and feeding behavior of finishing steers.

**Key Words:** feeding behavior, finishing cattle, growth performance, metabolizable protein

© The Author(s) 2019. Published by Oxford University Press on behalf of the American Society of Animal Science. This is an Open Access article distributed under the terms of the Creative Commons Attribution Non-Commercial License (<http://creativecommons.org/licenses/by-nc/4.0/>), which permits non-commercial re-use, distribution, and reproduction in any medium, provided the original work is properly cited. For commercial re-use, please contact [journals.permissions@oup.com](mailto:journals.permissions@oup.com)

Transl. Anim. Sci. 2019.3:1173–1181  
doi: 10.1093/tas/txz124

## INTRODUCTION

Feed costs represent the most expensive component in a feedlot production system (Lanna *et al.*, 1999). Protein supplementation to

<sup>1</sup>Corresponding author: [kendall.swanson@ndsu.edu](mailto:kendall.swanson@ndsu.edu)

Received April 15, 2019.

Accepted July 31, 2019.

ruminants often increases growth performance, which makes protein a limiting nutrient for production (Medeiros and Marino, 2015). However, excessive use of protein in finishing diets leads to economic losses and environmental implications due to excess of nitrogen excretion (Amaral et al., 2018). In addition, N excretion in ruminants is not only related to N intake but also microbial efficiency that influences metabolizable protein (MP) supply (Niu et al., 2016). MP is the true protein absorbed from the small intestine, supplied by microbial true protein, ruminally undegradable protein, and epithelial sloughed cells, which is available to the animal for maintenance, growth, and production (Das et al., 2014).

In the United States, the recommended percentage of crude protein (CP) and MP in finishing cattle diets ranged from 13.0% to 14.3% (average 13.4%), and 6.8% to 9.0% (average 8.15%; dry matter [DM] basis), respectively (Samuelson et al., 2016). These concentrations usually exceed the MP requirements for finishing cattle according to the Nutrient Requirements of Beef Cattle (NASEM, 2016). Furthermore, results from previous studies have shown that exceeding 13% CP (DM basis) in finishing diets does not improve growth performance (Gleghorn et al., 2004). In addition, increasing inclusion of CP in finishing cattle diets from 13% to 20% CP (DM basis) decreased energetic efficiency, which may also have a potential negative impact on growth performance (Hales et al., 2013) because of the energetic demand of metabolizing excess amino acids.

Under typical feeding conditions, decreasing CP concentrations in finishing diets could potentially cause adverse effects on DM intake (DMI) and growth (Galyean, 1996). Recent results have shown that increasing inclusion of CP in growing or finishing diets from 10% to 14% CP (DM basis) did not affect DMI (Amaral et al., 2018). However, Cole et al. (2003) reported that steers fed finishing diets containing 14% CP (DM basis) during the first 56 d had greater DMI than steers fed diets containing 12% CP (DM basis) during the same period. It is known that feeding behavior can be influenced by nutrition in cattle fed finishing diets (González et al., 2012); however, there is limited research about how specific nutrients, such as protein influence feeding behavior in finishing cattle. The objectives of this project were to determine the effect of MP intake on growth performance, carcass traits, and feeding behavior of finishing steers.

## MATERIALS AND METHODS

### *Animals, Experimental Design, and Dietary Treatments*

All procedures with animals were approved by the North Dakota State University Animal Care and Use Committee. One-hundred thirty-two steers ( $300 \pm 2.7$  kg body weight [BW]) predominantly of Angus, Simmental, and Shorthorn breeding were stratified by initial BW across five pens ( $n = 26$  to 32 steers/pen). Each pen contained eight automated feeders (Insentec; Hokofarm B.V. Repelweg 10, 8316 PV Marknesse, the Netherlands) and each diet was delivered to two troughs per pen. Steers were randomly assigned to one of four dietary treatments ( $n = 33$  steers/treatment) to supply 626, 906, 1,209, or 1,444 g MP/d (Table 1). The grain source used was dry-rolled corn. Diets were formulated using the Beef Cattle Nutrient Requirements Model (BCNRM) software (NASEM, 2016). The predicted estimates of MP intake before the initiation of the experiment of dietary treatments were 628, 912, 1,196, and 1,476 g MP/d. The average predicted MP requirement was 880 g MP/d (NASEM, 2016). Treatment 2 was formulated to meet the ruminally degradable protein (RDP) requirement (790 g RDP/d). The supply of predicted MP intake was formulated to differ by a similar amount between adjacent dietary treatments. After completion of the experiment, average MP intake was calculated as reported earlier using nutrient analysis and DMI data (NASEM, 2016). Diets were offered for ad libitum intake, and the steers had free access to water. Steers were adapted to experimental diets by transitioning from 60% to 90% concentrate diets over 28 d. Steers were implanted with 4 mg of estradiol and 20 mg of trenbolone acetate (Revalor-XS; Merck Animal Health, Whitehouse Station, N.J.) at d 0 of the experiment.

### *BW and Feed Intake Measurements*

BW measurements were taken on two consecutive days at the beginning of the experiment and on individual days every 28 d throughout the experiment in the AM before feed delivery. Final BW was estimated using linear regression (the slope of the days on feed  $\times$  predicted average daily gain [ADG] regression; average  $r^2 = 0.98$ ).

A radio frequency identification tag was placed in the right ear of each steer before the beginning of the experiment to allow for use of the Insentec automated feeding system (Hokofarm B.V.). As

**Table 1.** Diet and nutrient composition of the dietary treatments

Item	Treatment, g/d of MP intake			
	626	906	1,209	1,444
Ingredient, % of DM				
Corn	77.7	77.7	57.1	36.0
Corn silage	10.0	10.0	10.0	10.0
Wheat straw	5.00	5.00	5.00	5.00
DDGS	—	—	21.8	44.0
Corn oil	2.30	2.30	1.10	—
Urea	—	1.50	1.50	1.50
Limestone	1.80	1.80	1.80	1.80
Salt	0.24	0.24	0.24	0.24
Fine ground corn	2.87	1.37	1.37	1.37
Vitamin premix <sup>a</sup>	0.01	0.01	0.01	0.01
Trace mineral premix <sup>b</sup>	0.05	0.05	0.05	0.05
Monensin premix <sup>c</sup>	0.02	0.02	0.02	0.02
Tylosin premix <sup>d</sup>	0.01	0.01	0.01	0.01
Nutrient analyses <sup>e</sup>				
Dry matter (DM), %	73.3	73.8	74.2	74.1
Organic matter, % of DM	95.2	95.2	94.2	93.5
Crude protein, % of DM	7.84	11.7	17.2	20.9
Ruminally degradable protein, % of DM <sup>f</sup>	3.02	7.05	8.91	9.85
Neutral detergent fiber, % of DM	25.9	24.9	31.0	34.7
Acid detergent fiber, % of DM	10.7	10.3	12.6	13.8
Ether extract, % of DM	4.89	4.80	5.02	4.57
Calcium, % of DM	0.54	0.56	0.57	0.56
Phosphorus, % of DM	0.27	0.26	0.42	0.53

<sup>a</sup>Contained 48,510 kIU/kg vitamin A and 4,630 kIU/kg vitamin D.

<sup>b</sup>Contained 3.62% calcium (Ca), 2.56% copper (Cu), 16% zinc (Zn), 6.5% iron (Fe), 4% manganese (Mn), 1,050 mg/kg iodine (I) and 250 mg/kg cobalt (Co).

<sup>c</sup>Contained 176.4 g of monensin/kg premix.

<sup>d</sup>Contained 88.2 g of tylosin/kg premix.

<sup>e</sup>Average of weekly samples.

<sup>f</sup>Calculated using tabular values reported in [NASEM \(2016\)](#).

previously described by [Montanholi et al. \(2010\)](#) and [Wood et al. \(2011\)](#), the Insentec automated feeding system allows for provision of specific dietary treatments and monitoring of individual feed intake and feeding behavior characteristics. Feeding behavior measurements were quantified as described by [Montanholi et al. \(2010\)](#) as follows: events (number of daily visits and meals to the feed bunk), time eating in minutes (per visit, per meal, and per day), and feed intake in grams (per visit, per meal, and per minute). Feeding behavior data were summarized as the average of each individual steer over the entire experiment including the dietary adaptation period (28 d). A visit was defined as each time the Insentec system detected a steer at the feed bunk. A meal was defined as eating periods that might include short breaks separated by intervals no longer than 7 min ([Forbes, 1995](#); [Montanholi et al., 2010](#)).

### Feed Analysis

Diet samples were collected weekly throughout the experiment. Weekly samples were dried in a 55 °C oven for at least 48 h and ground to pass a 1-mm screen. Weekly samples were analyzed for DM, ash, N (Kjehldahl method), Ca, and P by standard procedures ([AOAC, 1990](#)). In addition, weekly samples also were analyzed for neutral detergent fiber and acid detergent fiber concentration by the method of [Robertson and Van Soest \(1981\)](#) using a fiber analyzer (Ankom Technology Corp., Fairport, NY). Percent CP was calculated by multiplying N concentration × 6.25. Samples also were analyzed for ether extract ([AOAC, 1990](#)). Average dietary composition results were entered into the feed composition table in the BCNRM software ([NASEM, 2016](#)). Updated feed nutrient composition and DMI values were used to calculate MP intake.

### *Carcass Characteristics*

Steers were fed until they achieved an average BW of  $598 \pm 3.1$  kg and marketed in five slaughter groups (treatments balanced within slaughter group). The first group was fed for 172 d ( $n = 15$ ), the second group for 179 days ( $n = 40$ ), the third group for 186 days ( $n = 44$ ), the fourth group for 195 days ( $n = 9$ ), and the fifth group for 200 days ( $n = 24$ ). After the fourth group was sent to slaughter, the remaining cattle were combined into one pen for the remainder of the experiment. Carcass characteristics were provided by the commercial slaughter facility; hot carcass weight (HCW) was measured after slaughter of the animal, whereas marbling score, subcutaneous fat thickness at the 12th rib (back fat), longissimus muscle area (LMA), and kidney, pelvic, and heart fat percentage (KPH) were measured via camera grading after carcass chilling.

### *Blood Collection and Plasma Glucose and Urea-N Analysis*

Blood samples were collected by jugular venipuncture into Vacutainer tubes containing sodium heparin (Becton Dickinson, Rutherford, NJ) on d 0, 86, and 172 before feeding. Plasma was isolated by centrifugation at  $3,000 \times g$  for 20 min at  $4^\circ\text{C}$  and stored at  $-20^\circ\text{C}$  until analysis. Plasma glucose analysis was performed using the hexokinase/glucose-6-phosphate dehydrogenase method (Farrance, 1987) with a kit from Thermo Scientific. Plasma urea-N was determined using the urease/Berthelot procedure (Fawcett and Scott, 1960; Chaney and Marbach, 1962). The intra-assay coefficient of variation was less than 5% for urea N and glucose assays.

### *Statistical Analysis*

Data were analyzed as a completely randomized block (slaughter group) design using the general linear model (GLM) procedure of SAS (SAS Inst. Inc., Cary, NC) for growth performance, carcass traits, and feeding behavior data. Linear and quadratic effects of MP intake were tested using orthogonal contrast statements. Contrast coefficients were determined using the IML procedure of SAS. For plasma glucose and urea-N concentrations, data were analyzed as a randomized block (slaughter group) design with repeated measures and tested for the effects of treatment, day, and treatment  $\times$  day using the Mixed procedure of SAS. Differences in DMI and plasma urea N observed

(see later) among treatments suggested differences over time in MP requirements. Therefore, DMI data (averages within each week of the experiment up until week 24, the week before the first group of steers were slaughtered) were also analyzed as a randomized block (slaughter group) design with repeated measures and tested for the effects of treatment, week, and treatment  $\times$  day using the Mixed procedure of SAS. Appropriate (minimize information criterion) covariance structures were used (Wang and Goonewardene, 2004). Data were considered statistically significant when  $P \leq 0.05$  and trends were discussed at  $0.05 < P \leq 0.10$ .

## RESULTS

ADG responded quadratically ( $P < 0.01$ ) with ADG increasing in steers fed 906 g MP/d and plateauing thereafter (Table 2). DMI (% of BW/day) was not different among treatments. However, DMI (kg/day) responded quadratically ( $P = 0.009$ ) with DMI increasing with MP intake up to 1,209 g MP/d and decreasing thereafter. When analyzing DMI over time, there was a week  $\times$  treatment interaction ( $P < 0.001$ ) as treatment influenced DMI differently depending on week (Figure 1). G:F increased linearly ( $P = 0.04$ ) and tended ( $P = 0.10$ ) to respond quadratically, as G:F increased up to 906 g MP/d and then plateaued. LMA was not affected by MP intake. HCW responded quadratically ( $P = 0.02$ ) as HCW increased to the greatest extent when MP intake increased from 626 g MP/d to 906 g MP/d with smaller increases thereafter. A quadratic effect ( $P = 0.04$ ) was observed for marbling score with steers fed 1,209 g MP/d having the greatest marbling score. In addition, there was a quadratic effect ( $P = 0.02$ ) for back fat thickness with steers fed 1,209 g MP/d having the greatest back fat thickness and plateauing thereafter. KPH fat percentage also responded quadratically ( $P = 0.01$ ) as KPH increased from 626 g MP/d to 906 g MP/d intake and plateaued thereafter.

A quadratic effect for visits ( $P = 0.002$ ) and meals ( $P = 0.005$ ) per day was observed with steers fed 1,209 g MP/d having the least visits and meals per day (Table 3). Time eating per visit responded quadratically ( $P = 0.05$ ) with time increasing from 626 to 906 g MP/d and plateauing thereafter. There was a linear increase in time eating per meal ( $P < 0.001$ ) and per day ( $P = 0.02$ ) with increasing MP intake. DMI (g) responded quadratically per visit ( $P < 0.001$ ), per meal ( $P < 0.001$ ), and per minute ( $P = 0.03$ ) with DMI being greatest when MP intake was 1,209 g MP/d.

A day  $\times$  treatment interaction ( $P < 0.001$ ) was observed for plasma urea N as concentrations increased to a greater extent over time in the higher MP treatments than in the lower MP treatments (Table 4; Figure 2). There was a day effect ( $P < 0.001$ ) for plasma glucose concentration but no day  $\times$  treatment interaction or dietary treatment effect.

## DISCUSSION

Optimization of MP intake in finishing cattle diets is important for maximizing production and

economic efficiency as well as minimizing environmental implications. The quadratic effect on ADG, HCW, marbling score, back fat, and KPH and the tendency for a quadratic effect on G:F likely occurred because steers fed 626 g MP/d were markedly deficient in RDP and thus MP. These results are similar to past research in finishing bulls (Amaral *et al.*, 2018) and in finishing lambs (Ebrahimi *et al.*, 2007). Efficient ruminal microbial growth and optimum microbial protein synthesis are influenced by ruminal synchronization between nitrogen and energy availability from dietary protein and

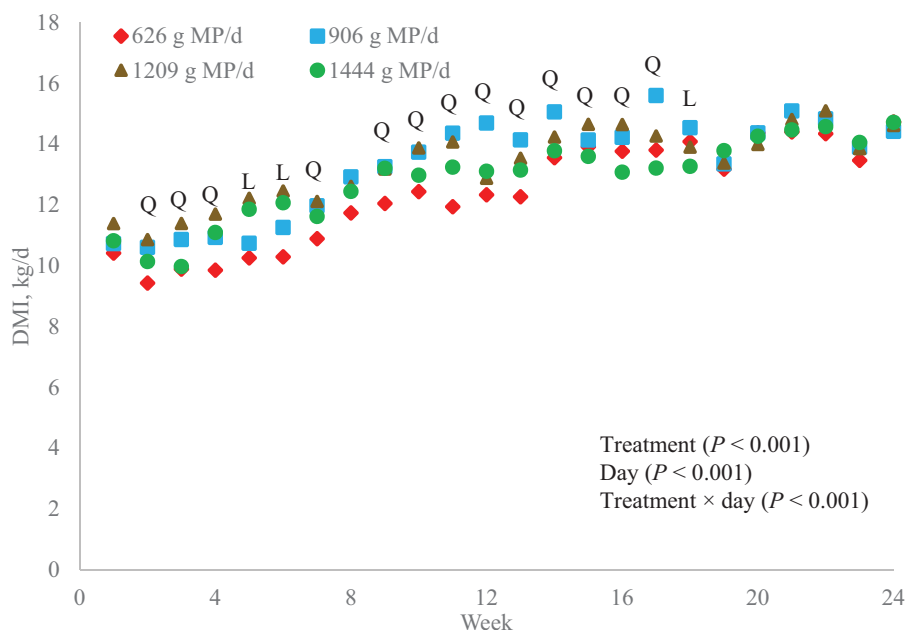
**Table 2.** Effect of metabolizable protein intake on growth performance and carcass characteristics of finishing steers

Item	Treatment, g/d of MP intake				SEM <sup>a</sup>	Contrast <i>P</i> -value	
	626	906	1,209	1,444		Linear	Quadratic
Initial BW, kg	295	299	303	304	5.4	0.18	0.77
Final BW, kg	566	610	607	607	6.2	<0.001	<0.001
Average daily gain, kg/d	1.46	1.67	1.63	1.63	0.027	<0.001	<0.001
Dry-matter intake, kg/d	9.19	9.79	9.92	9.64	0.170	0.03	0.009
Dry-matter intake, % BW/d	3.05	2.98	3.03	2.98	0.045	0.34	0.81
Gain:feed, g/kg	0.15	0.17	0.16	0.17	0.003	0.04	0.10
Hot carcass weight, kg	341	362	366	370	3.668	<0.001	0.02
Dressing percentage, %	60.4	59.5	60.4	61.0	0.52	0.26	0.11
Marbling score <sup>b</sup>	431	481	500	479	17.25	0.02	0.04
Back fat, cm	0.91	1.22	1.34	1.32	0.068	<0.001	0.02
Longissimus muscle area, cm <sup>2</sup>	83.9	84.6	85.4	86.4	1.437	0.17	0.89
KPH fat <sup>c</sup> , %	1.81	1.92	1.98	1.92	0.033	0.004	0.01

<sup>a</sup>Standard error of the mean where  $n = 33$ /treatment.

<sup>b</sup>For marbling score 400 = slight, 500 = small, 600 = moderate.

<sup>c</sup>Kidney, pelvic, and heart fat.



**Figure 1.** Effect of MP intake on DMI in finishing steers (pooled SEM = 0.356; L = linear effect of MP intake if  $P < 0.05$ , Q = quadratic effect of MP intake within day if  $P < 0.05$ ).

**Table 3.** Effect of metabolizable protein intake on feeding behavior of finishing steers

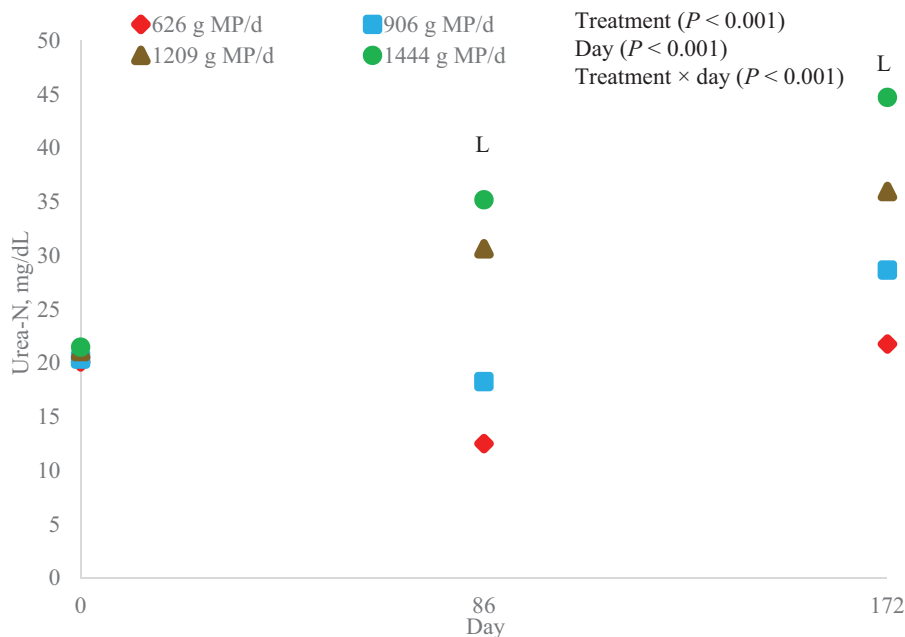
Item	Treatments, g/d of MP intake				SEM <sup>a</sup>	Contrast <i>P</i> -value	
	626	906	1,209	1,444		Linear	Quadratic
Events, per d							
Visits	36.1	29.5	24.4	27.0	1.45	<0.001	0.002
Meals	10.36	9.54	8.92	9.38	0.223	<0.001	0.005
Time eating, min							
Per visit	2.84	3.80	4.63	4.57	0.243	<0.001	0.05
Per meal	9.87	10.95	11.79	12.10	0.423	<0.001	0.43
Per day	101	103	104	112	3.2	0.02	0.27
Dry-matter intake, g							
Per visit	257	362	447	400	20.8	<0.001	<0.001
Per meal	906	1,053	1,140	1,053	30.4	<0.001	<0.001
Per min.	94.7	97.8	98.7	88.7	3.12	0.23	0.03

<sup>a</sup>Standard error of the mean where  $n = 33$ /treatment.

**Table 4.** Effect of metabolizable protein intake on jugular plasma metabolite concentration in finishing steers

Item	Treatment, g/d of MP intake				SEM <sup>a</sup>	<i>P</i> -value		
	626	906	1,209	1,444		Treatment	Day	Treatment × Day
Glucose, mg/dl	90.4	92.8	91.4	96.0	2.36	0.35	<0.001	0.74
Urea-N, mg/dL	18.1	22.4	29.2	33.8	0.58	<0.001	<0.001	<0.001

<sup>a</sup>Standard error of the mean where  $n = 33$ /treatment.



**Figure 2.** Effect of MP intake on jugular plasma urea-N concentration in finishing steers (pooled SEM = 0.97; L = linear effect of MP intake if  $P < 0.05$ ).

carbohydrates (Kebreab *et al.*, 2002; Neto *et al.*, 2007). The 626 g MP/d diet likely supplied too little dietary nitrogen for optimal fermentation, which may have also resulted in reduced energy utilization and volatile fatty acid production. In addition, the low intake of RDP likely reduced microbial protein synthesis and amino acid supply (MP) to the small

intestine. Although there were lesser differences in growth and carcass characteristics when MP intake increased from 906 to 1,444 g/d, there were differences between treatments and it differed depending on the output trait (ADG, HCW, marbling score, back fat, and KPH). When MP is provided in excess, there is a need to excrete the excess N. It

is estimated that a portion of energy lost as heat from the body can be attributed to urea synthesis in the liver and this may increase as MP intake increases (Huntington and Archibeque, 2000; Reed *et al.*, 2017). Therefore, increasing MP supply may result in increased ureagenesis potentially influencing metabolizable energy (ME) use by ruminants. Interestingly, there were quadratic effects in marbling score, back fat thickness, and KPH where steers fed 1,209 g MP/d had greater fat deposition than steers fed 1,444 g MP/d. This could suggest that steers fed 1,444 g MP/d had greater ME expenditure toward urea synthesis and nitrogen excretion, having a negative effect on energy availability towards growth. However, the magnitude of the response was relatively small suggesting only minor effects on growth and efficiency.

There have been mixed results on the effect of increasing CP inclusion in finishing diets on DMI (kg/day) in finishing cattle (Galyean, 1996; Menezes *et al.*, 2016; Amaral *et al.*, 2018). In the current experiment, increasing MP intake in finishing diets quadratically influenced DMI (kg/day) with the steers fed 626 g/d MP having the least DMI with smaller differences between the other three treatments. This may be partly because of the observed decreased ADG of the steers fed 626 g/d MP, and thus average BW throughout the feeding period, when compared with the other treatments. In addition, previous research has suggested that low supply of dietary N limits ruminal fermentation as well as passage rate of the digesta, which results in decreased feed intake (Campling, 1970).

Less is known about how MP influences feeding behavior of steers. Quadratic effects for visits and meals per day were observed with steers fed 626 g MP/d having the greatest number of visits and meals among the treatments. However, DMI per visit, per meal, and per minute were greater in steers fed 1,209 g MP/d. The lower ruminal supply of N to the microbes in steers fed 626 g MP/d likely affected microbial efficiency, decreasing ruminal digestibility and passage rate as well as lower ruminal ammonia release resulting in a lesser buffered ruminal environment. Therefore, steers fed 626 g MP/d may have had greater visits and meals in an attempt to compensate for the RDP deficiency; however, DMI was lesser per visit and meal. Ruminal pH is thought to be associated with feeding behavior (González *et al.*, 2012) and feeding behavior is influenced by feeding diets with different particle size (Swanson *et al.*, 2014). In the current experiment, increasing MP intake quadratically influenced time eating per visit, and resulted in linear increases in

time eating per meal and per day. Concomitantly with increasing total time eating, increasing MP intake of steers quadratically affected DMI (g/d). Interestingly, steers fed 1,444 g MP/d consumed 10 g less per minute when compared with steers fed 1,209 g MP/d. This could be partially attributed to palatability and physical characteristics of the 1,444 g MP/d diet, since the inclusion of 44% DDGS (DM basis) likely resulted in a diet having a smaller particle size (Swanson *et al.*, 2014) or greater S concentrations (Felix and Loerch, 2014; Felix *et al.*, 2015) when compared with the other diets. Alternatively, MP could influence feed intake through chemostatic mechanisms, resulting from changes in the amount and form of absorbed nutrients, regulated through the central nervous system (Hackmann and Spain, 2010; Allen, 2014). The transport of substrates such as VFA, glucose, and amino acids to the liver triggers brain feeding centers, which is thought to affect feeding behavior in ruminants (Allen, 2014). Our results showed that there were no effects on plasma glucose concentration among different MP intake treatments suggesting that changes in plasma glucose concentration were not responsible for the observed differences in feeding behavior.

It should be pointed out that diets containing greater MP also contained greater RDP as the identical supplement containing urea was fed in diets that supplied 906, 1,209, and 1,274 g/d MP. Also, starch concentrations of the diets decreased when MP supply increased as corn was replaced with DDGS to supply greater MP. Diets fed to supply 906, 1,209, and 1,274 g/d were formulated to meet or exceed RDP requirements and increasing RDP above requirements is not expected to increase MP supply (NASEM, 2016). We are confident that MP supply increased in the diets formulated to supply greater MP, even if the predicted RDP requirements happened to be inaccurate. The increased starch concentrations in the diets fed to supply lesser amounts of MP, especially in diets fed to supply 626 g/d MP, could result in increased incidence of subacute acidosis, which can influence DMI and feeding behavior (González *et al.*, 2012). However, no visual differences were observed among treatments in the incidence of subacute acidosis as indicated by fecal consistency and color. As increased incidence of subacute acidosis can be associated with differences in the variance of DMI throughout the experiment, we also analyzed DMI variance using repeated measures using weekly DMI variance and steers fed diets to supply lower MP did not consistently have greater DMI

variance (data not shown). In fact, steers fed diets to supply 626 g/d had the least DMI variance during 16 of the 24 weeks. Taken together, this would suggest that the differences observed in DMI were the results of differences in MP rather than starch in the diets.

Interestingly, there was an interaction between dietary treatment and day for plasma urea N with urea N decreasing between day 0 and 84 and then increasing until d 172 in steers fed 626 and 906 g MP/d, whereas urea N increased from d 0 until 172 in steers fed 1,209 and 1,444 g MP/d. This may suggest that MP requirements were not met for steers fed 626 or 906 g MP/d early in the feeding period but then were met as steers increased in BW. To gain more insight into this hypothesis, we also analyzed DMI over time using repeated measures by week and found an interaction between treatment and time ( $P < 0.001$ ). Interestingly, when analyzed within each week, there were quadratic or linear effects for 12 of the first 14 weeks with steers fed 626 g MP/d having the least DMI. After 12 weeks (d 84), the order of treatments from smallest to greatest changed and by 19 weeks no differences in DMI were observed among treatments.

Although our primary goal of the experiment was not to predict MP requirements in finishing cattle, the design of the experiment allows for such estimates. Because of the observed quadratic effects of MP intake on ADG and G:F, regression analysis, using treatment means fitting a quadratic equation and calculation of the vertex, was conducted with the predicted maximal response in ADG and G:F to be 1,152 ( $r^2 = 0.85$ ) and 1,274 g ( $r^2 = 0.54$ ) MP/d, respectively, both of which are greater than the predicted requirement of 880 g MP/d (NASEM, 2016). However, more research is needed to determine if MP requirements could be greater than predicted because the sample size was relatively small for adequate prediction of requirements and there is much variability in requirements between different animals.

In conclusion, increasing from insufficient to excess levels of MP intake of steers in finishing diets quadratically influenced growth performance, carcass characteristics, and feeding behavior. Steers fed 906 g MP/d had the greatest ADG whereas steers fed 1,209 g MP/d had the greatest DMI. HCW was greatest in steers fed 1,444 g MP/d; however, marbling score, back fat thickness, and KPH was greatest in steers fed 1,209 g MP/d. Steers that were fed 1,209 g MP/d had the least visits and meals per day but had larger meals than steers from the other treatments. In addition, steers that were fed 1,444 g MP/d spent more time eating per day and

had the slowest eating rate per minute. Additional research is needed to better understand how feeding behavior is affected by MP intake in finishing cattle, which potentially could contribute to the development of new feeding strategies.

## ACKNOWLEDGMENTS

The authors thank the staff at the NDSU Beef Cattle Research Complex and Nutrition Laboratory for their assistance.

*Conflict of interest statement.* None declared.

## LITERATURE CITED

- Allen, M. S. Drives and limits to feed intake in ruminants. 2014. *Anim. Prod. Sci.* 54:1513–1524. doi:10.1071/AN14478.
- Amaral, P. M., L. D. S. Mariz, D. Zanetti, L. F. Prados, M. I. Marcondes, S.A. Santos, E. Detmann, A. P. Faciola, and S. C. Valadares Filho. 2018. Effect of dietary protein content on performance, feed efficiency and carcass traits of feedlot Nellore and Angus  $\times$  Nellore cross cattle at different growth stages. *J. Agric. Sci.* 156:110–117. doi:10.1017/S0021859617000958.
- AOAC (Association of Official Analytical Chemists). 1990. *Official methods of analysis*, 15th ed. Arlington (VA): AOAC.
- Campling, R. C. 1970. Physical regulation of voluntary feed intake. In: A. T. Phillipson, editor, *Physiology of digestion and metabolism in the ruminant*. Newcastle Upon Tyne, UK: Oriel Press, p. 226–234.
- Chaney, A. L., and E. P. Marbach. 1962. Modified reagents for determination of urea and ammonia. *Clin. Chem.* 8:130–132.
- Cole, N. A., L. W. Greene, F. T. McCollum, T. Montgomery, and K. McBride. 2003. Influence of oscillating dietary crude protein concentration on performance, acid-base balance, and nitrogen excretion of steers. *J. Anim. Sci.* 81:2660–2668. doi:10.2527/2003.81112660x.
- Das, L. K., S. S. Kundu, D. Kumar, and C. Datt. 2014. Metabolizable protein systems in ruminant nutrition: a review. *Vet. World.* 7:622–629. doi:10.14202/vetworld.2014.622–629.
- Ebrahimi, R., H. R. Ahmadi, M. J. Zamiri, and E. Rowghani. 2007. Effect of energy and protein levels on feedlot performance and carcass characteristics of Mehraban ram lambs. *Pak. J. Biol. Sci.* 10:1679–1684. doi:10.3923/pjbs.2007.1679.1684
- Farrance, I. 1987. Plasma glucose methods, a review. *Clin. Biochem. Rev.* 8:55–68.
- Fawcett, J. K., and J. E. Scott. 1960. A rapid and precise method for the determination of urea. *J. Clin. Pathol.* 13:156–159. doi:10.1136/jcp.13.2.156.
- Felix, T. L., and S. C. Loerch. 2014. Effects of haylage and monensin supplementation on performance, carcass characteristics, and ruminal metabolism of feedlot cattle fed diets containing 60% dried distillers grains. *J. Anim. Sci.* 89:2614–2623. doi:10.2527/jas.2010–3716.
- Felix, T. L., T. A. Murphy, and S. C. Loerch. 2015. Effects of dietary inclusion and NaOH treatment of dried distillers grains with solubles on ruminal metabolism of



- feedlot cattle. *J. Anim. Sci.* 90:4951–4961. doi:10.2527/jas2011-4736.
- Forbes, J. M. 1995. Feeding behavior. In: J. M. Forbes, editor, *Voluntary food intake and diet selection in farm animals*. Wallingford, UK: CAB International, p. 11–37.
- Galyean, M. L. 1996. Protein levels in beef cattle finishing diets: industry application, university research, and systems results. *J. Anim. Sci.* 74:2860–2870. doi:10.2527/1996.74112860x.
- Gleghorn, J. F., N. A. Elam, M. L. Galyean, G. C. Duff, N. A. Cole, and J. D. Rivera. 2004. Effects of crude protein concentration and degradability on performance, carcass characteristics, and serum urea nitrogen concentrations in finishing beef steers. *J. Anim. Sci.* 82:2705–2717. doi:10.2527/2004.8292705x.
- González, L. A., X. Manteca, S. Calsamiglia, K. S. Schwartzkopf-Genswein, and A. Ferret. 2012. Ruminant acidosis in feedlot cattle: interplay between feed ingredients, rumen function and feeding behavior (a review). *Anim. Feed Sci. Technol.* 172:66–79. doi:10.1016/j.anifeedsci.2011.12.009.
- Hackmann, T. J., and J. N. Spain. 2010. A mechanistic model for predicting intake of forage diets by ruminants. *J. Anim. Sci.* 88:1108–1124. doi:10.2527/jas.2008-1378.
- Hales, K. E., N. A. Cole, and J. C. MacDonald. 2013. Effects of increasing concentrations of wet distillers grains with solubles in steam-flaked corn-based diets on energy metabolism, carbon-nitrogen balance, and methane emissions of cattle. *J. Anim. Sci.* 91:819–828. doi:10.2527/jas2012-5418.
- Huntington, G. B., and S. L. Archibeque. 2000. Practical aspects of urea and ammonia metabolism in ruminants. *Proc. Am. Soc. Anim. Sci.* 77:(E-Suppl):1–11. doi:10.2527/jas2000.&&E-Suppl1y.
- Kebreab, E., J. France, J. A. Mills, R. Allison, and J. Dijkstra. 2002. A dynamic model of N metabolism in the lactating dairy cow and an assessment of impact of N excretion on the environment. *J. Anim. Sci.* 80:248–259. doi:10.2527/2002.801248x.
- Lanna, D. P. D., L. O. Tedeschi, and J. A. B. Filho. 1999. Modelos lineares e nao-lineares de uso de nutrientes para formulacao de dietas de ruminantes. *Sci. Agric.* 56:479–488. doi:10.1590/S0103-90161999000200031.
- Medeiros, S. R., and C. T. Marino. 2015. Proteínas na nutrição de bovinos de corte. In: S. R. Medeiros, editor, *Nutrição de bovinos de corte: fundamentos e aplicações*. Brasília (BR): EMBRAPA, p. 29–44.
- Menezes, A. C. B., S. C. Valadares Filho, L. F. Costa e Silva, M. V. C. Pacheco, J. M. V. Pereira, P. P. Rotta, D. Zanetti, E. Detmann, F. A. S. Silva, L. A. Godoi, and L. N. Rennó. 2016. Does a reduction in dietary crude protein content affect performance, nutrient requirements, nitrogen losses, and methane emissions in finishing Nellore bulls? *Agric. Ecosyst. Environ.* 223:239–249. doi:10.1016/j.agee.2016.03.015.
- Montanholi, Y. R., K. C. Swanson, R. Palme, F. S. Schenkel, B. W. McBride, D. Lu, and S. P. Miller. 2010. Assessing feed efficiency in beef steers through feeding behavior, infrared thermography and glucocorticoids. *Animal* 4:692–701. doi:10.1017/S1751731109991522.
- NASEM (National Academies of Sciences, Engineering, and Medicine). 2016. *Nutrient requirements of beef cattle: eighth revised edition*. Washington, DC: The National Academies.
- Neto, S. F. C., L. M. Zeoula, R. Kazama, I. N. Prado, L. J. V. Geron, F. C. L. Oliveira, and O. P. P. Prado. 2007. Proteína degradável no rúmen associada a fontes de amido de alta ou baixa degradabilidade: digestibilidade in vitro e desempenho de novilhos em crescimento. *R. Bras. Zootec.* 36:452–460. doi:10.1590/S1516-35982007000200024.
- Niu, M., J. A. D. R. N. Appuhamy, A. B. Leytem, R. S. Dungan, and E. Kebreab. 2016. Effect of dietary crude protein and forage contents on enteric methane emissions and nitrogen excretion from dairy cows simultaneously. *Anim. Prod. Sci.* 56:312–321. doi:10.1071/AN15498.
- Reed, K. F., H. C. Bonfá, J. Dijkstra, D. P. Casper, and E. Kebreab. 2017. Estimating the energetic cost of feeding excess dietary nitrogen to dairy cows. *J. Dairy Sci.* 100:7116–7126. doi:10.3168/jds.2017-12584.
- 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, editors, *The analysis of dietary fiber*. New York (NY): Marcell Dekker, p. 123–158.
- Samuelson, K. L., M. E. Hubbert, and C. A. Löest. 2016. Nutritional recommendations of feedlot consulting nutritionists: the 2015 New Mexico State and Texas Tech University survey. *J. Anim. Sci.* 94:2648–2668. doi:10.2527/jas.2016-0282.
- Swanson, K. C., A. Islas, Z. E. Carlson, R. S. Goulart, T. C. Gilbery, and M. L. Bauer. 2014. Influence of dry-rolled corn processing and increasing dried corn distillers grains plus solubles inclusion for finishing cattle on growth performance and feeding behavior. *J. Anim. Sci.* 92:2531–2537. doi:10.2527/jas.2013-7547.
- Wang, Z., and L. A. Goonewardene. 2004. The used of MIXED models in the analysis of animal experiments with repeated measures data. *Can. J. Anim. Sci.* 84:1–11. doi:10.4141/A03-123.
- Wood, K. M., H. Salim, P. L. McEwen, I. B. Mandell, S. P. Miller, and K. C. Swanson. 2011. The effect of corn or sorghum dried distillers grains plus solubles on growth performance and carcass characteristics of cross-bred beef steers. *Anim. Feed Sci. Technol.* 165:23–30. doi:10.1016/j.anifeedsci.2011.02.011.