

Protein Supplementation for Beef Cattle

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Valuation of protein sources for beef cattle

For supplementing protein (or nitrogen [N]) to beef cattle, the primary concern is providing enough ruminally available N (**RAN**) to meet the needs of the ruminal microbes to ensure that ruminal fermentation maximizes energy availability. In short, we feed protein to optimize fermentation and maximize the energy availability from the diet. When the N/protein needs of the ruminal microbes are met, the flow of microbial protein to the small intestine along with the amount of ruminally undegraded protein (**RUP**) provided by common dietary ingredients will, in most cases, meet the needs of most beef cattle for absorbable amino acids.

Most models that calculate beef cattle performance predict the amount of microbial protein that flows out of the rumen, and this estimate is important for predicting the amount of RAN is required to meet the microbes needs. Ruminal microbes can obtain RAN either directly from ruminally degraded protein (**RDP**) in the diet or from recycled urea-N.

Recycling of urea-N to the gastrointestinal tract, and presumably that to the rumen, is generally similar when RDP or digestible RUP are included in the diet, at least under conditions where RAN is limiting (Wickersham et al., 2008, 2009). However, RDP will additionally provide its N directly to the microbes and, thus, it is better able to meet the microbes' needs than equivalent amounts of RUP. In other words, RDP provides the microbes with N from the degraded protein as well as N from recycled urea, whereas RUP only provides microbes with N from recycled urea. Therefore, in typical situations for most types of beef cattle production, RDP will be of greater value than RUP because RAN is the most critical nutrient provided by the dietary protein.

For most situations in the beef cattle industry, we can roughly equate the value of a protein source with its ability to provide RAN, either directly as RDP or via recycled urea-N. In this context, ruminal degradability and postruminal digestibility of RUP are the factors that will affect the ability of protein sources to provide RAN.

In the work of Wickersham et al. (2008, 2009), digestible RUP led to slightly more of the supplemental N being recycled to the rumen (98%) than did RDP (66%). Because the protein sources were provided on an equal total N basis, the RDP provided significantly more RAN to the cattle than did the RUP. The efficiency of urea recycling decreases as RAN supply increases, which might suggest, therefore, that recycling might be quite similar between RDP and RUP,

when providing equal amounts of RAN. Thus, for calculations presented herein, I simplified the relationship between protein supply and urea recycling by using an intermediate value of 80% to predict the recycling of urea from either RDP or RUP. As such, the RAN supply from a protein source could be calculated as: $RDP + (0.80 \times \text{digested N})$, regardless of where the N is digested.

Using the 80% estimate for urea recycling from either RDP or digestible RUP, the calculations in Table 1 are designed to consider the effects of ruminal degradability of protein as well as of the indigestible N content on the value of a protein source in providing RAN. Protein degradabilities were set to range from 20 to 80% of the feed's N, and indigestible protein ranged from 0 to 40% of the total N. Few feedstuffs would have values outside of these ranges. From the calculations in Table 1, there are clearly disadvantages to increasing the RUP content of a feedstuff, even if there is no detrimental effect on the amount of indigestible N. At the same time, there are additional detrimental effects on RAN if there are increases in indigestible protein, regardless of the ruminal degradability. Additionally, the effect on RAN of a 30% shift in ruminal N degradability (such as decreasing from 80% to 50%) is greater than the effect of a 30% change in indigestible protein. A 30% change in ruminal degradability is a real-world possibility if a feedstuff with highly degradable protein were treated to reduce ruminal degradation (e.g., heating of soybean meal). In contrast, 30% of total protein being indigestible would represent a rather poor quality feed, likely with extensive heat damage. Feedstuffs with large concentrations of indigestible protein are generally more likely to have large concentrations of RUP, so it would be unlikely to find a feedstuff with high RDP along with a large fraction of indigestible protein. In contrast, it is possible to find feeds, such as quality ring-dried blood meal, that would have a large fraction of RUP along with very small amounts of indigestible protein (i.e., the RUP is well digested in the small intestine).

With the viewpoint that the main goal of protein supplementation to most beef cattle diets should be to provide RAN, it is obvious that we primarily want to select protein sources that are extensively degraded in the rumen (high RDP) and that also have extensive small intestinal digestion of any RUP that is present. The question then becomes: How can we effectively measure these two characteristics in feedstuffs in a manner that is accurate, fast, and inexpensive?

Many protein systems consider the *in situ* Dacron bag method as an acceptable way to assess ruminal degradation of feed proteins. Most routine analyses with this approach use a single time point for the incubation to improve throughput and reduce cost. By increasing the number of time points, it is possible to more thoroughly fractionate a feedstuff's protein and determine the rate of degradation for the potentially degraded fraction; this allows RDP to be calculated across a range of passage rates. If a single time point is used, the time of incubation is critical. For example, companies marketing to the dairy industry, where high RUP is valued, are likely to utilize a shorter incubation time to elevate the estimate of their product's RUP concentration. In the beef industry where greater RDP should be valued, longer incubation times might be preferred for marketing purposes, but few feedstuffs are specifically marketed on the basis of a high ruminal protein degradability. This may be because few feeds have greater RDP than the commonly available solvent soybean meal, alfalfa, and urea.

About 50 years ago, Goering et al. (1972) identified acid detergent insoluble N (**ADIN**) as a useful measure of indigestible protein in heat-damaged forages. Based on the success of ADIN as a measure of indigestible protein in heated forages, a number of researchers have assessed ADIN as a measure of indigestible protein in various feedstuffs, and this concept still remains in some models. For non-forage protein sources, there is not a direct relationship between ADIN content and indigestible protein, suggesting that ADIN cannot be used as an accurate assessment of indigestible protein. For example, Nakamura et al. (1994) measured total tract N digestibilities of various sources of distillers grains in lambs, and they found no relationship between ADIN content of the distillers grains and the N digestibility. This agreed with previous work from Nebraska where the ADIN fraction of the feed was not found to be indigestible (Britton et al., 1987). Visual assessment of the color of SBM or distillers grains can provide some qualitative information about heat damage in feeds. Several studies have verified the expected conclusion that DDGS that have experienced more heating have a greater ADIN concentration and a darker color. Cromwell et al. (1993) showed a general relationship between dark color and ADIN concentration of dried distillers grains, although most of the samples in that study were from beverage plants and not from fuel alcohol manufacturers. Cromwell et al. (1993) demonstrated that darker DDGS had lower lysine contents and led to worse performance of pigs fed protein-limiting diets. Lower lysine concentrations reflect irreversible binding of lysine in Maillard reaction products, which would be expected to increase both RUP and indigestible N. In contrast, Nakamura et al. (1994) observed different colors among their distillers grains as well as large differences in ADIN concentrations, yet total tract digestibility of N did not differ appreciably among sources, suggesting that color and ADIN may not be perfect predictors of the ability of distillers grains to provide RAN to cattle.

In my opinion, the best option for assessing RUP concentration and postruminal digestion remains the three-step procedure described by Calsamiglia and Stern (1995). This procedure estimates ruminal digestion using a 16-hour in situ ruminal fermentation followed by sequential treatment with acid-pepsin and pancreatin to determine small intestinal digestion of the RUP. For reasons noted above, some users of this approach select shorter time points for the ruminal fermentation to better reflect rapid ruminal passage from cattle with high feed intakes. The three-step procedure does not directly estimate indigestible protein, but large intestinal disappearance of N from a supplemental protein source is unlikely to be large, so the estimate of indigestible RUP from the three-step procedure should be a reasonable estimate of unavailable N.

Certainly there are some aspects of the three-step procedure that are not ideal. Most importantly, ruminally cannulated cattle are required, which increases complexity of the assay as well as run-to-run variation. One could argue that the data are directly applicable only to feeding conditions that match the diet fed to the cannulated cattle. Moreover, the cost and length of the assay are concerns. Some commercial labs will provide data from the three-step procedure; most commercial analyses are conducted for feeds destined for use in the dairy industry where high RUP concentrations are valued, but it may be worthwhile for feedstuffs destined for beef cattle as well (although for different reasons). Using data collected from the

three-step procedure, one could compare the value of protein sources for the beef industry as $RDP + 0.8 \times \text{total tract digestible protein}$.

Lysine supplementation for growing cattle limit fed corn-based diets

Although most beef cattle will have their metabolizable protein requirements met by supplies of microbial protein and RUP contained in common dietary ingredients, there may be cases where beef cattle require protein/amino acid supplementation to achieve optimal performance. Limit-fed, rapidly growing cattle might be a situation where responses to protein supplementation might be expected. Growing cattle have protein deposition rates that are greater than finishing cattle. Moreover, when growing cattle are limit fed, the goal is typically to achieve near maximal rates of protein deposition, while limiting the amount of fat deposition. To limit fat deposition, energy intake is restricted, either by feeding a diet with a low energy concentration or by restrictedly feeding a more energy-dense diet. In cases where energy intake is restricted, microbial protein synthesis will be limited by the availability of fermentable energy. This in turn will decrease supplies of microbial protein. In addition, corn protein is known to be particularly deficient in lysine. Thus, if protein supply is limiting in calves fed corn-based diets, then lysine might be the most limiting amino acid.

Recently, we conducted a trial to assess the benefit of supplementing ruminally protected lysine to limit-fed steers (255 kg). The steers were predominantly Angus-cross and were implanted with Revalor G. The control diet contained 10% dry-rolled corn, 29.5% steam-flaked corn, 40% Sweet Bran, and 13% hay. Treatments included: control, 0.129% Smartamine-ML (**Lys-3**, providing roughly 3 g/d metabolizable lysine), 0.259% Smartamine-ML (**Lys-6**, providing roughly 6 g/d metabolizable lysine), and 0.89% blood meal (**BM**, providing roughly 3 g/d metabolizable lysine). Calves were limit-fed once daily at 2.4% of body weight (dry matter basis). Relative to control over the 77-day growing phase, supplementing Lys-3 increased body weight gain 8.7 kg, whereas Lys-6 increased body weight gain by 4.7 kg (Table 2). The BM treatment, which should have provided the same amount of lysine as Lys-3, did not increase body weight gain.

Following the growing phase where the treatments were applied, steers were shipped to a commercial feedyard where they were finished on a common diet for an average of 195 days. At slaughter relative to control, Lys-3 steers had 3.4 kg greater carcass weights, Lys-6 steers had 7.1 kg greater carcass weights, whereas BM steers had carcass weights no greater than control. This data provides for some interesting observations. During the growing phase, 3 g/d lysine was more effective than 6 g/d lysine in improving performance. Yet, when the cattle were finished on a common diet, steers fed Lys-3 maintained their advantage over the controls, but the higher level of lysine (Lys-6) during the growing phase led to better finishing performance and the heaviest carcasses. Also interesting was the inability of BM, which was designed to provide the same amount of lysine as Lys-3, to modify either growing phase or finishing phase performance. These results raise the possibility that Lys-6 somehow programmed the cattle for better performance during the finishing phase when the identical diets were fed. We were unfortunately unable to measure feed intake by treatment during the finishing phase, so it is possible that finishing-phase feed intake was different among treatments. However, ribeye

areas were slightly larger and back fat depth was slightly less for cattle that received Smartamine-ML during the growing period, and the slight decreases in back fat might suggest that feed intake was not greatly increased by lysine supplementation during the finishing phase.

Methionine and choline effects on health of receiving cattle

We have recently been studying supplementation to growing cattle of methionine and other compounds containing methyl groups. Some data would suggest that the amino acid methionine or the methyl-containing compound choline could reduce inflammation and fatty liver in periparturient dairy cows (Grummer, 2008; Zhou et al., 2016a,b). In a growth study with receiving beef heifers, Grant (2020) supplemented ruminally protected methionine as Smartamine-M. Methionine supplementation did not affect performance, which was an expected result because the corn-based diet was predicted to provide adequate amounts of methionine. We were most interested in evaluating effects on health performance, but unfortunately, from a research perspective, morbidity rates were extremely low and therefore could not be assessed. However, over time, plasma haptoglobin, a measure of hepatic inflammation, became lower ($P = 0.05$) for heifers that received supplemental methionine than for control heifers.

We are now in the midst of replicated growth studies with receiving heifers to assess effects of supplementation with ruminally protected methionine or ruminally protected choline. Our hypothesis is that either methionine or choline might improve immune response of heifers, leading to less morbidity and/or better responsiveness of sick heifers to treatment. Although pathogens cause respiratory disease, an animal's overactive immune response can sometimes be more detrimental to health than the pathogen itself. Thus, taming of an overstimulated immune system could be of value.

We conducted an experiment to evaluate the effects of choline supplementation to steers maintained under conditions where methyl group supply was designed to be either increased and decreased relative to control. The methyl group status of the steers did not appear to affect our measures of immune function, but choline supplementation tended to reduce plasma haptoglobin as well as in vitro neutrophil phagocytosis after a lipopolysaccharide challenge. These responses suggest a modification of the immune response that might lead to less self-damage in response to an overly activated immune system.

Supplementation of guanidinoacetic acid to growing cattle

Methionine is often a limiting amino acid for lactating dairy cattle, and it has been shown to be the most limiting amino acid in ruminal microbial protein. Across a number of research projects, we have shown that supplemental methionine is used with a lower efficiency than are various other essential amino acids (Titgemeyer, 2012). Over time, this led us to consider the role that methionine plays as a methyl group donor, with the thought that methionine's use as a methyl group donor might lead to a catabolism rate greater than for other amino acids. There are hundreds of reactions for which methionine serves as a methyl group donor, but the two quantitatively most important reactions are synthesis of creatine and choline.

Creatine is a vitamin-like compound that can be synthesized by the body in a two-step process. In the first step, glycine and arginine (two amino acids) are used to synthesize guanidinoacetic acid (**GAA**). The GAA is then methylated to form creatine. The regulatory step in this process is the synthesis of GAA, whereas all of the available GAA is methylated to creatine, independent of the body's needs. Thus, we started studying GAA supplementation as a potential means of modifying methyl group availability because the supplemental GAA would consume methyl groups from methionine. Our initial goal was to create a methyl group deficiency. Although GAA supplementation to cattle led to some minor increases in plasma homocysteine (Ardalan et al., 2020, 2021), which is a hallmark of methyl group deficiency, we never generated an extreme methyl group deficiency with GAA supplementation.

Recently, some research from China has demonstrated huge improvements in performance of finishing Angus bulls in response to GAA supplementation. Bulls started the trials at 400-450 kg, and were fed diets containing on average 36% corn silage and 29% ground corn (13.5% CP, 40% NDF, 36% NFC). Across three studies, 0.6 g GAA/kg dry matter increased average daily gains during 60- to 90-day feeding periods (Li et al., 2020; Liu et al., 2021a,b). Gains increased by an average of 24%, whereas efficiency was improved by an average of 16% when 0.6 g GAA/kg dry matter was added to the diet. Presumably this response relates to the conversion of GAA to creatine, which was a limiting factor for growth of muscle tissues. If translatable to the U.S. beef finishing industry, this response to GAA supplementation would be a game changer.

Although we have not supplemented GAA to finishing cattle, we have observed some small changes in nitrogen retention (a measure of whole body protein deposition) in growing cattle. In one study, GAA was able to slightly increase N retention when steers were provided adequate amounts of methionine, but not when they were methionine deficient (Ardalan et al., 2021). This makes sense, because methionine is required for the methylation of GAA to creatine. In another study, GAA led to small decreases in N retention, independent of methionine status (Speer, 2019). In a third study (Grant et al., 2021), N retention was slightly increased when GAA was supplemented, independent of methionine status. Taken as whole, we have not observed large growth responses to GAA supplementation, but our models have been designed more to effect a methyl group deficiency than to assess growth responses. Future research in this area will be particularly interesting.

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Table 1. Amount of ruminally available N (RAN) provided by protein sources with different proportions of ruminally degradable protein (RDP) and indigestible protein

Indigestible protein, % of total N	RDP, % of total N		
	20	50	80
	----- RAN, % of feed N -----		
0	100	130	160
10	92	122	152
20	84	114	144
30	76	106	NA
40	68	98	NA

RAN was estimated as: $RDP + 0.80 \times (\text{digestible protein})$, where digestible protein equals RDP plus intestinally digested RUP. The 0.80 coefficient is based on the assumption that 80% of RDP as well as 80% of digestible RUP will be recycled to the rumen as urea.

NA: more than 20% indigestible protein is not compatible with 80% of total protein as RDP.

Table 2. Response of growing cattle to lysine supplementation during the growing phase

Item	Treatment ¹				SEM	Lysine (<i>P</i> -value)	
	Control	Lys-3	Lys-6	BM		Linear	Quad
Bodyweight, kg							
Day 0	249.1	247.9	248.6	248.7	1.45	0.83	0.60
Day 77	393.7	401.3	397.9	392.8	3.87	0.45	0.26
DM intake, kg/d	7.66	7.73	7.68	7.63	0.061	0.77	0.41
Daily gain, kg/d	1.88	1.99	1.94	1.87	0.042	0.32	0.12
Gain:feed, kg/kg	0.247	0.259	0.254	0.247	0.0040	0.25	0.08

¹Lys-3 = 0.129% of diet as Smartamine ML. Lys-6 = 0.259% of diet as Smartamine ML. BM = 0.89% of diet as blood meal.

Table 3. Response of finishing cattle to lysine supplementation during the growing phase

Item	Treatment ¹				SEM	Lysine (<i>P</i> -value)	
	Control	Lys-3	Lys-6	BM		Linear	Quad
Daily gain, kg/d	1.37	1.37	1.42	1.38	0.06	0.17	0.39
Slaughter wt ² , kg	672.8	678.1	683.8	672.1	5.9	0.20	0.98
Carcass weight, kg	434.2	437.6	441.3	433.7	3.8	0.20	0.99
Ribeye area, cm ²	94.7	97.7	97.5	96.0	1.6	0.05	0.18
Back fat, cm	1.87	1.68	1.80	1.79	0.060	0.36	0.04
Choice + Prime, %	98.3	97.1	99.2	95.5	2.3	0.75	0.53

¹Cattle received treatments only through the 77-day growing phase. See Table 2.

²Calculated from hot carcass weights and average dressing percentages.