

PROTEIN REQUIREMENT AND NON PROTEIN NITROGEN UTILIZATION

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Factors governing nitrogen utilization by dairy cattle include the following: (a) Maintenance of ruminal ammonia nitrogen in excess of 5 mg/100 ml rumen fluid has no effect on microbial protein production; (b) Supplemental NPN is not utilized in typical dairy and feedlot beef rations containing, more than 12 to 13% CP (DM basis); (c) NPN is approximately equal to true protein as a nitrogen source in typical dairy and feedlot rations containing not more than 12 to 13% CP; (d) A scheme based upon metabolizable protein (absorbable protein) for calculating requirements and comparing protein sources is superior to the crude or digestible protein designations. Ultimate expression of the requirement may be in terms of CP for the sake of simplicity; (e) One kg of CP regardless of nitrogen source, equals about .75 kg metabolizable protein in typical dairy and feedlot beef rations containing not more than 12 to 13% CP. One kg of plant protein (true protein) fed in excess of an amount equivalent to 12 to 13% dietary protein equals about 0.3 kg metabolizable protein; (f) Protein supplementation of lactating cows might better be related to stage of lactation than level of milk production; (g) Lactating cows having above-average lactation AI ability may benefit from dietary protein levels reaching as high as 16 to 18% (DM basis) during the first third of lactation; (h) Cows in the latter two-thirds of lactation appear to require 12.5% dietary protein or less; (i) Plant protein (true protein) should be the supplemental source of nitrogen during the first third of lactation, with NPN providing most, if not all, the supplemental nitrogen during the last two-thirds of lactation.

INTRODUCTION

It has long been recognized that supplemental nonprotein nitrogen (NPN) is most efficiently utilized in rations low in protein and relatively high in digestible energy. It is also widely accepted that supplemental NPN is utilized better when small rather than large amounts are added to ruminant rations. A clear understanding of the limits to NPN utilization is necessary for profitable use of NPN in ruminant rations.

It can reasonably be assumed that dietary NPN will be of little benefit to the ruminant unless it is first converted into ammonia, and then utilized for microbial protein synthesis in the rumen. If this is true, it then is important to know the concentration of ruminal ammonia necessary for maximal microbial growth rates. Maintenance of ruminal concentration in excess of the bacterial requirement would result in waste of the nitrogen and thus add cost but no benefit.

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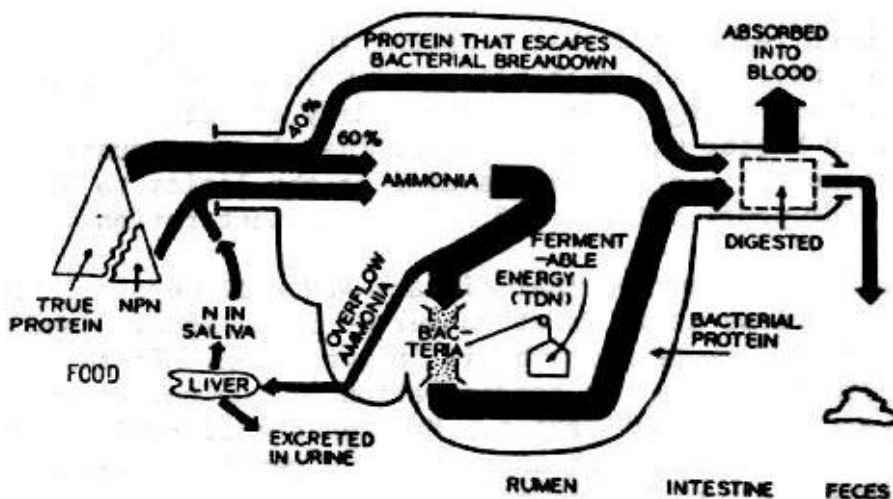
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The purpose of this paper is to outline a scheme that can be used to evaluate protein and NPN utilization by ruminants. This approach relates crude protein and total digestible nutrient (TDN) content of the ration to absorbable or metabolizable protein. This is discussed in relation to what is currently understood about protein requirements of lactating cows.

UTILIZATION OF AMMONIA BY RUMEN MICROORGANISMS

A schematic summary of nitrogen utilization by ruminants is presented in figure 1. Ingested true protein may either be degraded by the ruminal microorganisms, or may escape degradation and pass to the lower gut to be digested or excreted in the faeces. The amount of true protein that escapes degradation may vary considerably, but with most management and feeding conditions employed in the dairy and feedlot industry in the USA, an escape rate of 40% for the dietary protein probably represents an acceptable average. The remaining 60% of dietary protein is degraded almost entirely to ammonia. Dietary NPN, salivary nitrogen and possibly a small amount of urea entering across the rumen wall are converted almost totally to ammonia. Dietary NPN, salivary nitrogen and possibly a small amount of urea entering across the rumen wall are converted almost totally to ammonia.

Figure 1:
Schematic summary of nitrogen utilization by the ruminant



The amount of ammonia that can be utilized by the bacteria will depend on how many bacteria there are, and how rapidly they are growing. In other words, it will depend on the amount of fermentable feed consumed. Feeds high in TDN are more fermentable than those low in TDN. Therefore, more ammonia (NPN) can be utilized when feeds

high in TDN are used. This is illustrated in figure 1 where an increase in TDN "opens the gate" and allows greater ammonia use by supporting greater bacterial numbers.

Figure 1 illustrates a situation where the bacteria are unable to utilize all of the ammonia produced and there is an "ammonia overflow". This excess ammonia is absorbed from the reticulorumen, or passed to the lower gut, where it is absorbed and eventually converted to urea by the liver. A fraction of this may be recycled via saliva to the rumen.

It is apparent that being able to predict the point of "ammonia overflow" in the rumen would be helpful in determining when or when not to expect benefit from addition of NPN to the ration. To accomplish this, two pieces of information were considered necessary. The first was to know what concentration of ruminal ammonia was necessary to support maximal growth rates of rumen bacteria. Secondly, it was necessary to know the mean or prevailing concentration of ruminal ammonia under typical feeding management conditions. The desired objective, therefore, was to relate mean ruminal ammonia concentration to readily measured and understood characteristics of the ration, namely crude protein and TDN content.

CONCENTRATION OF RUMINAL AMMONIA NECESSARY FOR MAXIMAL MICROBIAL GROWTH

The effect of ruminal ammonia concentration on microbial protein production was determined in continuous culture fermenters (Satter and Slyter 1974). Fermenters were charged with ruminal ingesta obtained from steers fed either a protein-free purified, or a maize-based all-concentrate, or a 77% concentrate ration. The cultures were fed at the rate of 16 - 1.4 g dry matter/500 ml culture volume per day. Diets were similar to those fed steers supplying the ingesta except that the crude protein level of the culture diets ranged from 4 to 28%. Urea was the source of supplemental nitrogen and was continuously infused into the fermenters to maintain constant ammonia concentrations.

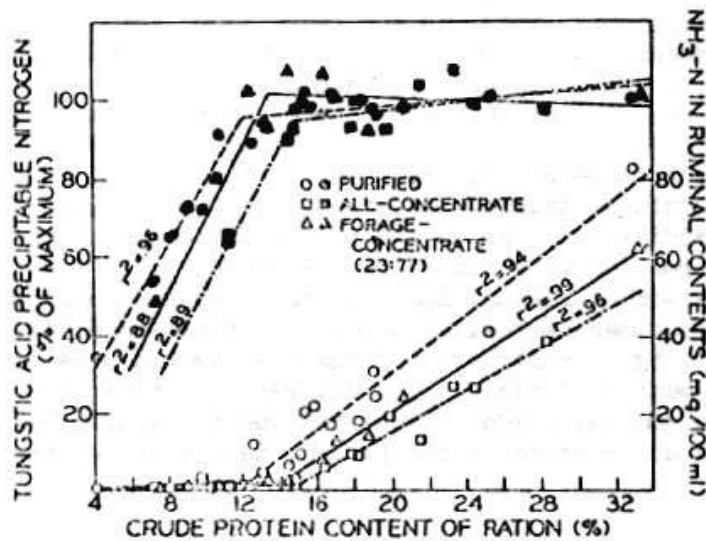
Figure 2 shows the relationship between crude protein content (DM basis) of the mixture added to the fermenter protein output of the fermenter measured as tungstic acid precipitable nitrogen (TAPN), and ammonia concentration of the incubation mixture. Each point is an average of composite samples obtained from one fermenter during the two last 4 days of each 9-day experiment. Protein output by the fermenters increased as the level of urea supplementation was increased, and then levelled off. without effect then Further increases in the amount of supplemental urea were on protein output. The levelling off of protein output coincided with the point where ammonia began to accumulate. Ammonia nitrogen in excess of 5 mg/100 ml of rumen fluid had no effect on the protein content of the fermenter effluent.

The data points in figure 2 were submitted to a least-squares regression analysis. r^2 values for each set of data points ranged between .88 and .99, indicating a very good fit between the computed regression lines and the individual data points. The

intercepts for the TAPN and ammonia plots were at 12.0 and 11.9, 13.5 and 14.2, and 14.7 and 15.0% crude protein content of ration DM for the purified, forage-concentrate, and concentrate rations. This good agreement underscores the close relationship between achievement of maximum TAPN production and the beginning of accumulation of ruminal ammonia.

Figure 2:

Relationship between ammonia concentration (\circ , Δ , \square) and tungstic acid precipitable nitrogen (\bullet , \blacktriangle , \blacksquare) produced *in vitro*, when a purified substrate mixture or a forage/concentrate mixture was added to the fermenter



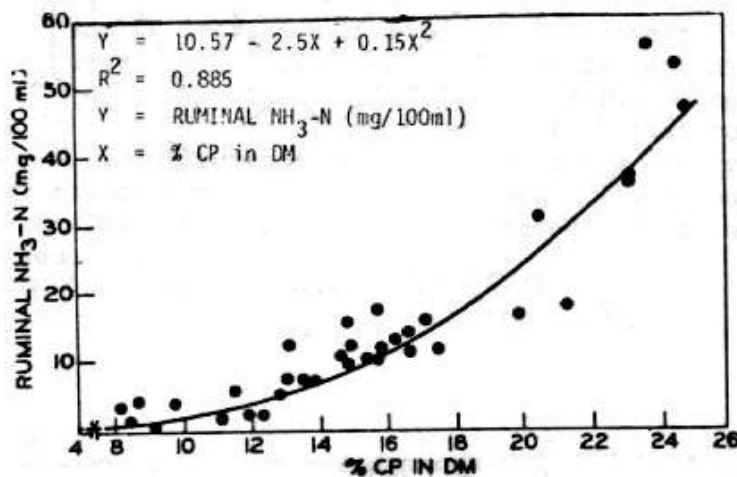
EFFECT OF RATION COMPOSITION ON RUMINAL AMMONIA CONCENTRATION

In order to study the influence of ration composition on mean ruminal ammonia concentrations, a total of 1,038 ruminal ingesta samples were collected via stomach tube from 207 cows in different stages of lactation and maintained under a variety of feeding programs. Animals were sampled prior to feeding and at least three times after feeding to obtain an average ruminal ammonia concentration. All rations were formulated using only natural protein sources. Simple and multiple regressions relating dietary crude protein (CP), total digestible nutrients (TDN), and mean ruminal ammonia concentration were computed. Crude protein was determined in the laboratory and ration TDN was calculated using NRC values.

The relationship between mean ruminal ammonia concentration and dietary crude protein is shown in figure 3. Each point in figure 3 represents a different trial mean. Since the 35 trial means are based on different sample sizes, a weighted regression analysis was performed. The weighting factor used was: Number of observations/variance of the mean.

Mean ruminal ammonia concentration was found to be positively related to percent CP in the ration dry matter. As dietary CP increased above 13% (DM basis), ruminal ammonia increased rapidly and was in excess of 5 mg NH₃-N/100 ml rumen fluid. Thus, when ration CP content is greater than 13%, more ammonia is present in the rumen than can be converted to microbial protein. Addition of NPN to such rations is superfluous. It should be explained that this curve applies only to cattle. Preliminary observations suggest that, as dietary protein increases, ruminal ammonia accumulates more rapidly in sheep than cattle

Figure 3:
Relationship between ruminal ammonia concentration and dietary crude protein



The following multiple regression equation, which considers both ration CP and TDN, was found to improve the precision of predicting mean ruminal ammonia concentration:

$$Y = 38.73 - 3.04X_1 - 0.490X_2 + 0.171X_1^2 + 0.0024X_2^2 \quad r^2 = 0.92$$

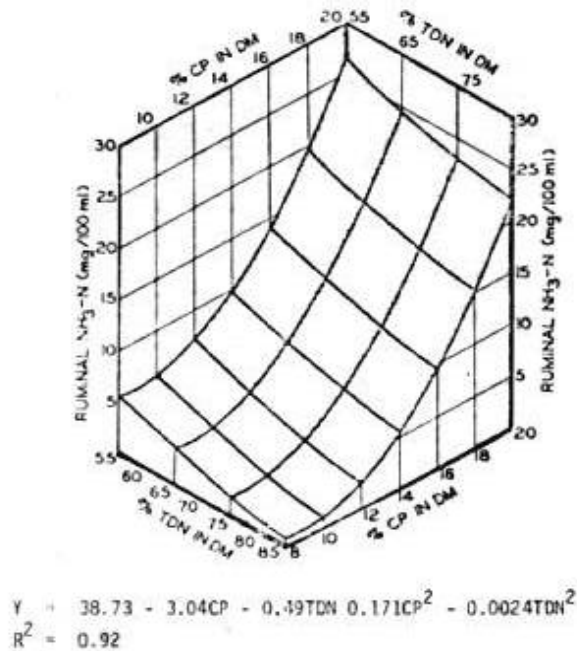
Where Y = Ruminal NH₃-N/100 ml rumen fluid

X₁ = %CP in ration DM

X₂ = % TDN in ration DM

This equation describes the surface shown in figure 4. It can be seen that, with increasing dietary protein, ruminal ammonia reaches 5 mg NH₃-N/100 ml rumen fluid sooner with low energy rations than with high energy rations. It should be emphasized that this equation was derived from experiments where only plant protein was fed, and therefore does not apply directly to situations where NPN is in the diet. It will tend to underestimate ruminal ammonia if NPN is in the ration.

Figure 4:
Relationship between ruminal ammonia concentration and various combinations of dietary protein and energy



Numerical values from figure 4 are presented in table 1. Dietary protein and energy combinations have been divided into three groups: (1) those CP and TDN combinations where NPN would be utilized efficiently, (2) those combinations where NPN would be partially utilized; and (3) those where NPN would not be utilized.

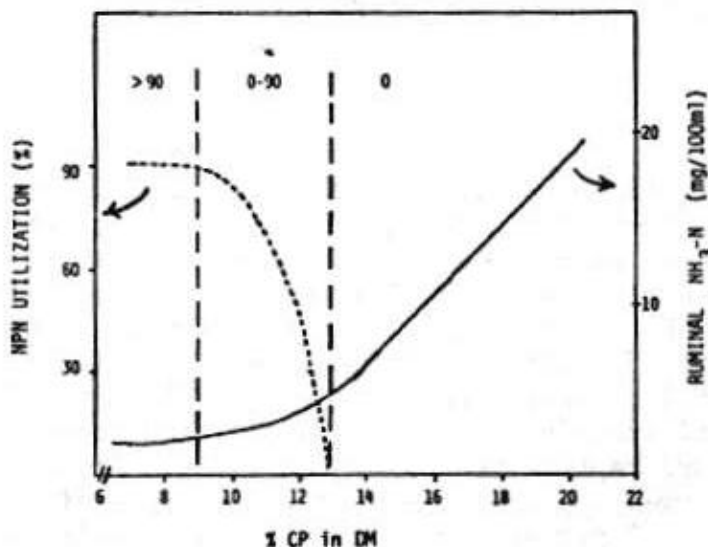
Table 1:
Influence of concentrate composition on mean ruminal ammonia concentration and NPN utilization

% CP in DM	% TDN in DM							NPN utilization %
	55	60	65	70	75	80	85	
	----- (mg/100 ml) -----							
8	6	5	4	3	2	2	1	
9	6	5	4	3	2	2	1	
10	6	5	4	3	2	2	1	>90
11	6	5	4	3	3	2	2	
12	7	6	5	4	4	3	3	0-90
13	8	7	6	6	5	4	4	
14	10	9	8	7	6	6	5	
15	12	11	10	9	8	8	7	
16	14	13	12	11	10	10	10	
17	17	16	15	14	13	13	12	0
18	20	19	18	17	16	16	15	
19	23	22	21	20	19	19	18	
20	27	26	25	24	23	23	22	

EFFICIENCY OF CONVERTING NPN INTO MICROBIAL PROTEIN

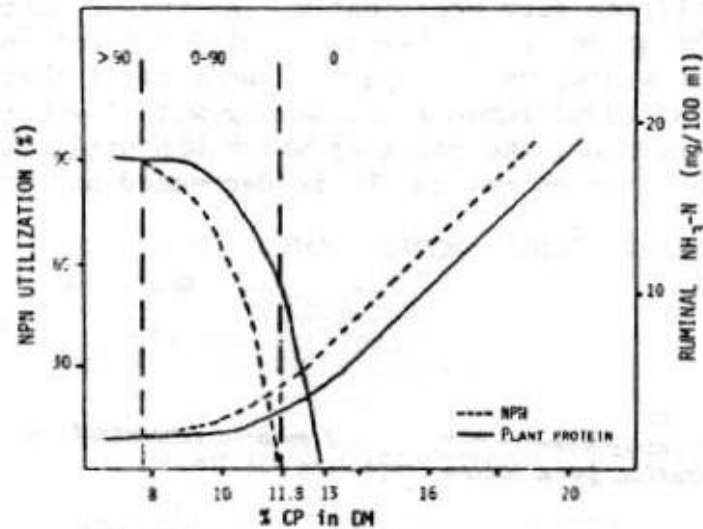
The relationship between mean ruminal ammonia concentration and NPN utilization by the rumen bacteria is presented in figure 5. The solid line represents the ruminal ammonia accumulation curve for rations containing 75% TDN. This curve was taken directly from figure 4. The dotted line is the inverse of the solid line (plotted on a different scale) and represents NPN utilization. When ruminal ammonia concentration is very low (less than 1 to 2 mg NH_3 -N/100 ml), nearly all the ammonia becomes incorporated into the bacterial cells. The efficiency of NPN utilization is therefore high as indicated by the dotted line in figure 5. As ammonia begins to accumulate (the area between the two vertical dashed lines in figure 5), the efficiency with which NPN is converted to microbial protein is reduced. The rate at which the efficiency is decreasing is inversely proportional to the rate at which ammonia concentration is increasing. When the CP content of the ration is low, NPN is utilized very efficiently. As the CP content increases, utilization of NPN decreases to zero, at 13% for a ration containing 75% TDN. Since it was shown in figure 4 and table 1 that ruminal ammonia reaches 5 mg NH_3 -N/100 ml rumen fluid sooner with low energy rations than with high energy rations, the point at which NPN utilization reaches zero would shift to the left as ration TDN is decreased and to the right as it is increased.

Figure 5:
Relationship between mean ruminal ammonia concentration and NPN utilization for a ration containing 75% TDN



The ammonia accumulation curve shown in figure 5 was established from data obtained using rations containing no added NPN. With most rations about 40% of the protein escapes degradation in the rumen and becomes available to the animal post-ruminally. When part of the ration protein is replaced by NPN, the amount of true dietary protein escaping ruminal degradation is obviously reduced. For example, if 3 kg of true protein are consumed in the ration and 40% of this escapes degradation, then 1.2 kg of protein escape. On the other hand, if 50% of the ration nitrogen is replaced by NPN, only 1.5 kg of true protein would be consumed. Therefore, only 40% of 1.5 kg, or 0.6 kg, would escape. The effect of substituting NPN for protein is shown in figure 6.

Figure 6:
Effect of NPN substitution on ruminal ammonia accumulation for rations containing 75% TDN



The solid lines in figure 6 correspond to the ammonia accumulation and NPN utilization curves in figure 5. For each percentage unit of protein replaced by NPN, the point at which mean ruminal ammonia reaches 5 mg NH₃-N/100 ml rumen fluid would be shifted back to the left by 0.4 percentage units of CP on the X axis. For the example shown in figure 6, 3 percentage units of plant protein have been substituted with NPN. Hence, a shift of 1.2 percentage units (from 13% CP to 11.8% CP) was made. The NPN utilization curve would be shifted accordingly. The adjusted curves are represented by dotted lines. It is emphasized that these adjusted curves apply to the specific example of a ration which contains 75% TDN, 10% percentage units of true protein, and 3 percentage units of crude protein equivalent from NPN. Different combinations of TDN, true protein, and NPN would result in different curves. The point

of excessive ammonia accumulation in the rumen is, therefore, affected by the amount of NPN added to the ration as well as the TDN and total protein content of the unsupplemented ration. As the amount of NPN in the ration increases, the point of zero utilization of NPN is reached at a corresponding lower level of ration protein.

Table 2 is a summary of the foregoing discussion, and suggests an upper limit for NPN supplementation with different rations. Nothing is gained by supplementing NPN beyond the upper limit (point of excessive ammonia accumulation). It must be recognized that with a given TDN and crude protein content, the point at which ammonia accumulates will vary slightly between animals as well as between rations. It has been our experience, however, that when typical dairy or feedlot rations are fed, the variation about the mean values (table 2) is small, and the values in table 2 are reliable upper limits from which general recommendations can be made. A more complete review of the above discussion is available (Roffler and Satter, 1975a) The concept of variable NPN utilization is not new. The qualitative aspects of urea utilization have been understood for quite some time, but the quantitative aspects have until now been obscure. Time and space do not permit a thorough discussion of other evidence that supports the conclusions presented in table 2, but mention is made here of the type of evidence available. The subject is discussed in detail by Roffer and Satter (1975b).

Table 2:
Upper limit for NPN utilization

% CP in DM before NPN	% TDN in DM					
	55-60	60-65	65-70	70-75	75-80	80-85
	----- (% CP after NPN addition) -----					
8	No	10.0	10.5	10.9	11.2	11.4
9	No	10.4	10.9	11.3	11.6	11.8
10	No	10.8	11.3	11.7	12.0	12.2
11	No	11.2	11.7	12.1	12.4	12.6
12	No	No	12.1	12.5	12.8	13.0

A review was made of published experiments with sheep where the flow of non ammonia or total nitrogen through the abomasum or small intestine was measured. There was a total of 24 rations where NPN was the source of supplemental nitrogen, and 36 rations where protein was the source of nitrogen. Using regression analysis, the flow of nonammonia nitrogen through the abomasum when NPN was the source of supplemental nitrogen was equal for all rations ranging between approximately 10 and 24% CP (see figure 7). This suggests that the high nitrogen rations containing NPN were no better than the low nitrogen rations containing NPN in terms of protein flow through the abomasum. With rations supplemented with true protein, however,

there was a continued increase in the flow of nonammonia nitrogen through the abomasum as the dietary level of protein was increased to as high as 26% of the ration (figure 8). This incremental increase in flow of protein through the abomasum presumably represents that portion of the dietary protein which escaped ruminal degradation.

Figure 7:
Influence of NPN supplementation on the quantity of non-ammonia nitrogen (NAN) flowing through the abomasum of sheep fitted with omasal, abomasal or duodenal cannulae

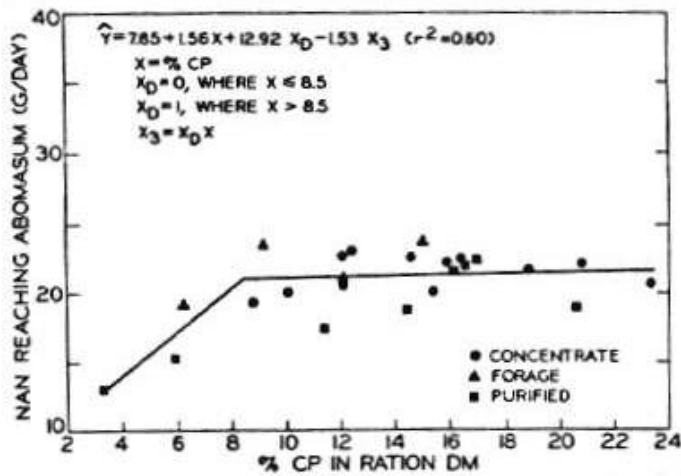
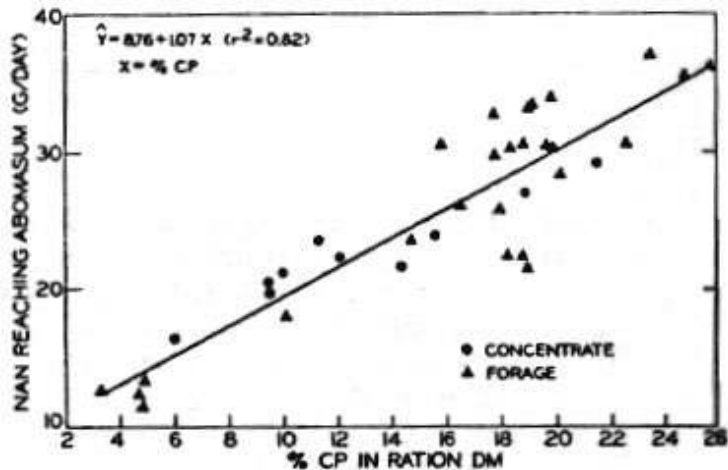
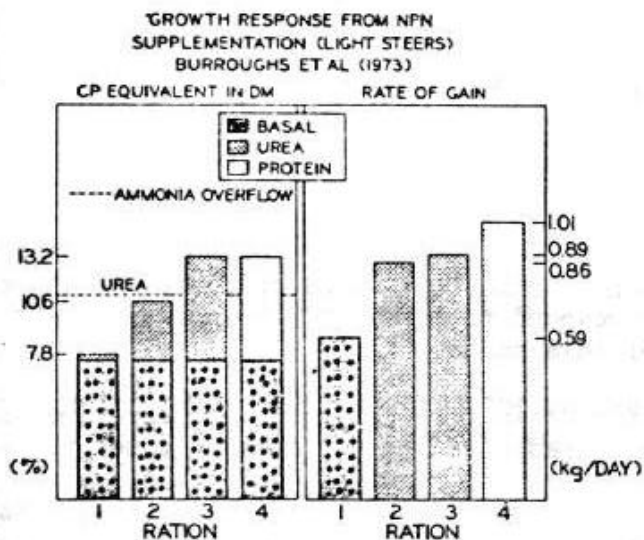


Figure 8:
Influence of protein supplementation on the quantity of non-ammonia nitrogen (NAN) flowing through the abomasum of sheep fitted with omasal, abomasal or duodenal cannulae



A second line of evidence, using growth of feedlot cattle as the measured response, is available from the studies of Burroughs et al (1973) (figure 9). Experiments in this group of studies were well designed, having four treatment groups consisting of: (a) a low protein basal ration, (b) a basal ration plus low NPN, (c) a basal ration plus high NPN, and (d) a basal ration plus plant protein isonitrogenous with treatment (c). Eighteen animals were assigned to each treatment, thus assuring a sound statistical evaluation of the results. The point of ammonia accumulation, as predicted from our regression equation (from figure 3), is illustrated by the dotted line in figure 9. The conclusion reached from this experimentation was that addition of NPN to appropriate low protein rations could result in an increase in growth rate, but that further additions could be without benefit, even though the animal needed additional protein as evidenced by superior performance of the plant protein-supplemented group. The point of predicted ruminal ammonia accumulation agrees with the observed growth performance.

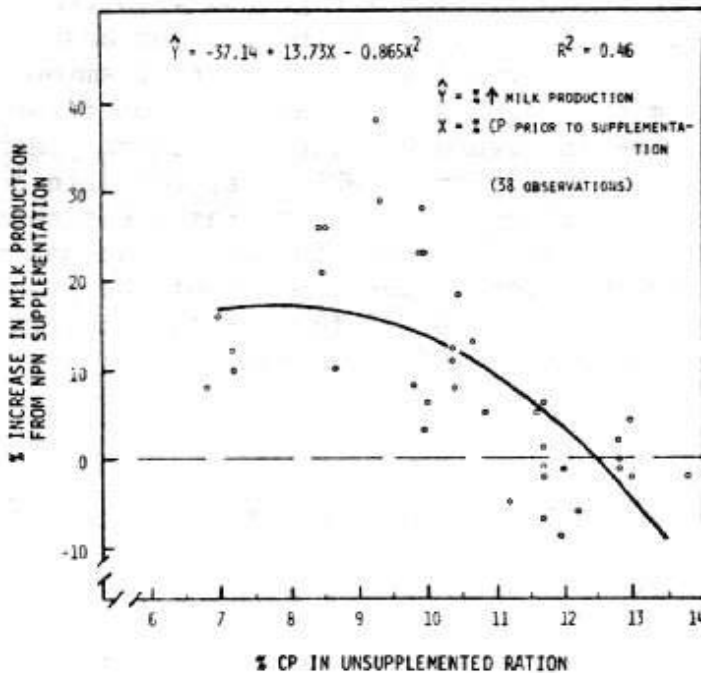
Figure 9:
Growth from plant protein or NPN supplementation of feedlot rations



A third line of evidence regarding ineffectiveness of NPN in rations containing more than 12 to 13% crude protein was obtained using data from published lactation trials involving 38 comparisons of NPN-supplemented rations to unsupplemented negative control rations. These comparisons involved 406 cows. A simple regression equation was computed relating percent improvement in milk production following NPN supplementation (figure 10). Addition of NPN to low protein rations caused a substantial increase in milk production. The milk production response diminished progressively as the protein content of the ration prior to NPN supplementation was increased. The point of zero response in milk production to NPN supplementation occurred when the rations contained, prior to supplementation, about 12.5% crude

protein. This line of evidence is somewhat equivocal, for many of the cows used in these experiments probably had a dietary protein requirement of 12.5% or less, and therefore would not have been able to respond to either NPN or plant protein supplementation in excess of this amount.

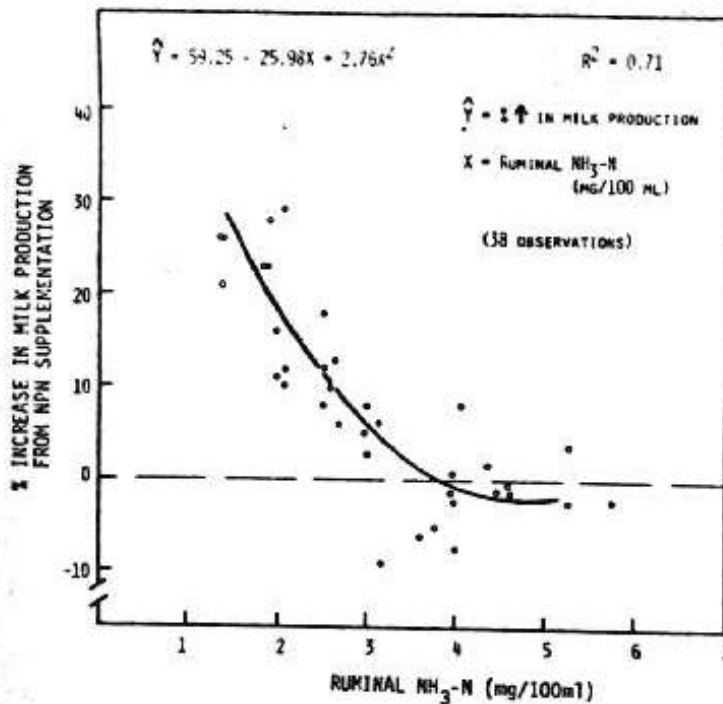
Figure 10:
Summary of published studies where milk production response to NPN supplementation was measured



Another noteworthy observation drawn from this group of experiments deals with the relationship between predicted ruminal ammonia concentration and milk production response. Estimates of ruminal ammonia concentration for cows in each of the published studies were obtained by using the equation discussed earlier to predict ammonia concentration from ration protein and TDN content. A regression of milk production response on predicted ruminal ammonia ($r^2 = .71$) indicated zero response with ruminal ammonia nitrogen concentration in excess of 4 mg/100 ml (figure 11). This agrees closely with the value of 5mg $\text{NH}_3\text{-N}/100$ ml previously demonstrated to be adequate for maximal microbial growth rates.

A common mistake in the design of most studies carried out with NPN is that isonitrogenous comparisons of NPN and plant protein are made under conditions where the animals have an excess supply of protein (amino acids). Under these conditions the inability of NPN to provide an equivalent amount of amino (compared to plant protein) for absorption is not detected, and the investigator erroneously concludes that NPN is equivalent to plant protein.

Figure 11:
Summary of where milk production response to NPN supplementation is plotted against predicted ruminal ammonia concentration prior to NPN supplementation



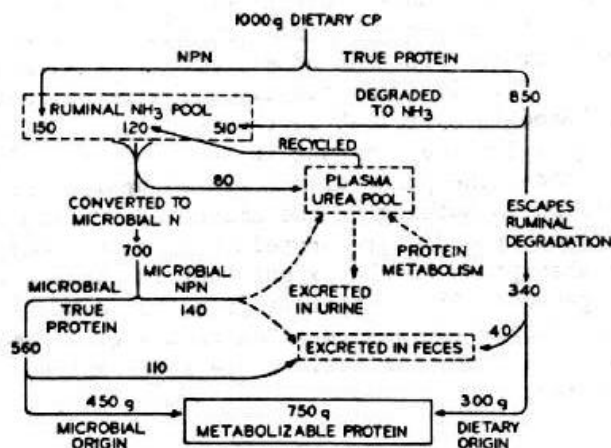
CALCULATION OF METABOLIZABLE PROTEIN

Crude protein (N x 6.25) has been the favored terminology for expressing protein composition of feeds and animal protein requirements. This is particularly true for ruminants, since NPN may have nearly the same nutritive value as amino nitrogen under some conditions. The amount of metabolizable or absorbable protein is not linearly related to dietary CP intake when a wide range of protein intakes are considered. In other words, the amount of metabolizable protein per unit of CP intake is much higher under conditions where ruminal ammonia is totally utilized for microbial protein production than under conditions where ruminal ammonia is in excess. This is schematically illustrated in figure 1. Below the point of "ammonia overflow" in the rumen, all nitrogen sources are approximately equal for the ruminant in terms of providing metabolizable protein. Above the point of ammonia accumulation, however, additional NPN contributes nothing to the amount of protein available for absorption. Additional true dietary protein adds to the amount of protein available for absorption to the extent that it escapes ruminal degradation and is digested. It is apparent from this discussion that the amount of metabolizable protein obtained from a given amount of CP is somewhat less when high protein rations are fed compared to when low protein rations are fed.

The discussion that follows outlines an approach for calculating metabolizable protein. It is simple and logically structured, and may be readily modified to accommodate more accurate information as it becomes available. The assumptions made and values derived therefrom must be considered tentative. It is hoped that this approach may provide a framework within which more definitive experimentation can be planned, and that it may ultimately be useful as a vehicle for expressing National Research Council recommendations regarding protein requirements for dairy and feedlot beef cattle. This subject is discussed in more detail by Satter and Roffler(1975).

Assumptions Upon which The Metabolizable Protein Scheme is Based: The following assumptions form the basis for estimating metabolizable protein. Some can be well documented with evidence, others must be considered tentative. All values must be considered as average values, recognizing that each mean value is associated with some variance. Assumptions: (a) the amount of nitrogen recycled into the reticulorumen is equal to 12% of the dietary nitrogen intake, (b) 85% of the dietary nitrogen intake with typical ruminant rations unsupplemented with protein is in true protein form, and 15% in NPN form, (c) 40% of the true dietary protein escapes degradation in the rumen, and goes to the intestine, while all of the dietary NPN and recycled nitrogen passes through the ruminal ammonia pool, (d) 90% of all ruminal ammonia produced is incorporated into microbial nitrogen when the ration fed does not exceed the "upper limit" value for CP given in table 2, (e) 0% of all ruminal ammonia derived from dietary CP fed in excess of the "upper limit" value for dietary CP (table 2) is incorporated into microbial nitrogen, (f) 80% of microbial nitrogen is in true protein form, and 20% is in a nonutilizable NPN form, (g) 80% of the microbial true protein will be absorbed (metabolizable protein), (h) 87% of the dietary true protein that escapes degradation in the rumen will be absorbed (metabolizable protein).

Figure 12:
Schematic summary of metabolisable protein calculations



Calculation of Metabolizable Protein: Application of the assumptions the calculation of metabolizable protein is illustrated in figure 12. Under conditions where ruminal ammonia production is not in excess of microbial need, 1 kg of dietary protein results in 750 g of metabolizable protein. Of this amount, 450 g is of microbial origin and 300 g of dietary origin. In other words, whenever the CP content of a ration is equal to or less than the "upper limit" value of table 2, the amount of metabolizable protein will equal 75% of the CP consumed. For the sake of simplicity, all forms of dietary nitrogen are considered equal when the dietary protein level does not exceed the "upper limit" values. This is not true to the extent that microbial nitrogen derived from dietary NPN does not provide quite the amount of absorbable amino acids as would be supplied by the equivalent amount of dietary true protein. The difference is very small, however, and can be ignored.

When dietary CP is fed in excess of the "upper limit" value of table 2 a twostep calculation is needed. Above the point of ammonia accumulation in the rumen, only escaped dietary true protein will contribute to metabolizable protein. One kg of protein supplemented under these conditions results in only about 0.3 kg of metabolizable protein. Therefore, the total amount of metabolizable protein available from a high protein ration ("upper limit" value) will equal the sum of 75% of dietary CP fed up to the "upper limit" value, and 30% of the dietary CP of that portion fed above the "upper limit" value (assuming only plant protein is fed). If a cow were consuming 20 kg of a ration containing 72% TDN and 17% CP (DM basis and no added NPN), the "upper limit" value would be 12.8% CP (table 2). The amount of metabolizable protein obtainable below the point of ruminal ammonia accumulation would, therefore, be $20 \text{ kg} \times .128 \times .75$, or 1.92 kg. The amount of metabolizable protein obtainable above the point of ammonia accumulation would be $20 \text{ kg} \times (.17 - .128) \times .30$, or .252 kg. The total amount of metabolizable protein available from this ration equals $1.92 + .252$, or 2.172 kg. If the ration in the above example contained 11% CP rather than 17%, calculation of metabolizable protein would simply be $20 \text{ kg} \times .11 \times .75$, or 1.65 kg. Thus, metabolizable protein is 64% and 75% of CP for the 17% and 11% CP rations, respectively.

In future research suggests that there are important differences between the major protein supplements with regard to the amount of protein escaping ruminal degradation, different constants may be assigned to different supplements. For example, fishmeal might have a value of .55, soybean meal .30, peanut meal .15, etc. This is of practical significance only if the protein is fed over and above the point of ruminal ammonia accumulation. Protein degradation below the point of ammonia accumulation makes little difference except as it may shift the point of ammonia overflow. As mentioned earlier, protein from the basal ration (protein supplied by the "home-grown feeds") makes up the major portion of the protein fed below the point of ammonia accumulation, and the common mixtures of these proteins do not seem to affect the point of ruminal ammonia accumulation significantly.

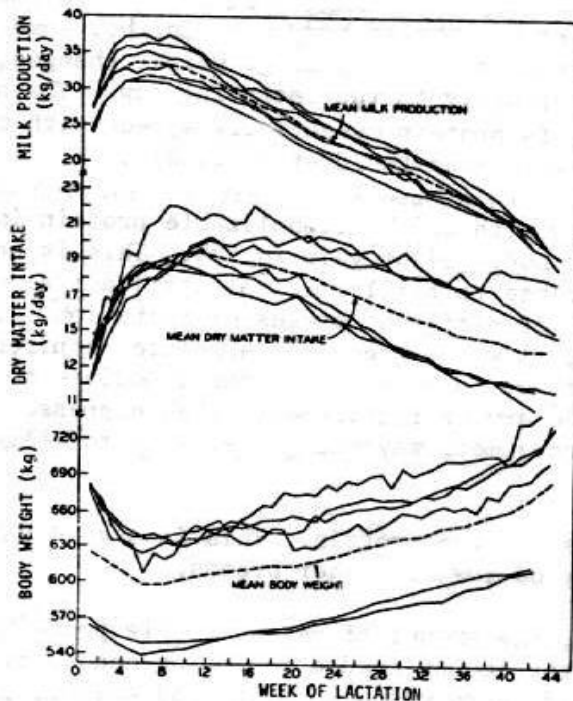
A Feeding Standard Based Upon Consumption of Metabolizable Protein: The relationship between milk production, dry matter intake, body weight, and stage of lactation for six groups of cows (Everson 1973; Swanson et al 1967; Trimberger et al 1972) was plotted and is shown in figure 13. The dotted line is the mean of all groups, weighted according to the square root of the number of cows in each group. Each group contained between 10 and 20 cows. Crude protein content of the total ration fed to each group during early lactation ranged between 14.8 and 18.8%, averaging 16.9%. The protein content averaged slightly lower during later lactation. If the six groups of cows may be considered typical, the mean response curves might serve as reference curves, suitable for use in developing feeding standards. One shortcoming of this approach is that the "standard" group of cows may have been deficient in protein in early lactation, resulting in milk production short of potential production. As will be pointed out later, predicted protein requirement in early lactation approached the level being fed in these experiments. If the "standard" group of cows were deficient in protein, calculated estimates of protein requirement in early lactation will be low.

As shown in figure 13, peak milk production occurred at about 5 to 7 weeks but dry matter consumption did not reach its maximum until 9 to 11 weeks. body weight losses occurred during the first 6 weeks, followed by a stabilization period of about 2 weeks, after which there were gradual weight gains posted. The crucial period, from a protein nutrition point of view, is during the first 9 to 11 weeks of lactation. Unless a cow can mobilize protein and fat (calories) proportionate to need, the ration will have to be enriched with protein. Available evidence suggests that the ability of cows to mobilize tissue protein for the synthesis of milk is quite small relative to the ability to mobilize energy (Coppock et al 1968). Therefore, protein must be "packed" into a smaller total quantity of feed during early lactation if protein requirements of the cow are to be met.

At 9 to 11 weeks, energy intake has reached a maximum, and milk production is on the decline. In contrast to the first few weeks of lactation when low protein, high energy body reserves were contributing to milk synthesis, the situation beyond 11 weeks is reversed, with deposition of body tissue low in protein and high in energy taking place. A cow producing 30 kg milk on the ascending portion of the lactation curve has quite different dietary requirements for protein than the same cow producing 30 kg milk on the descending portion of the lactation curve. Our present feeding standards do not recognize this point.

The important effect that level of feed intake may have on the required percentage of dietary protein, combined with the effect of inefficient protein utilization in high protein rations (12-13% CP), results in potentially high dietary CP requirements for cows in early lactation. This may be illustrated with some tentative calculations based upon metabolizable protein intake and metabolizable protein requirements in early lactation.

Figure 13:
Effect of stage of lactation on milk production, dry matter intake and body weight



In making these calculations, the following assumptions were made:

(a) The maintenance requirement for metabolizable protein is 2.4 g/kg. This was calculated from the digestible protein requirement for maintenance of 1.6 g/kg⁷⁵ for cattle as suggested by Preston (1972) in a recent review of protein requirements of cattle. The value suggested by Preston is approximately 60% as much as recommended by NRC. Under most maintenance feeding conditions, 1 g of apparently digestible protein would be equivalent to about 1.5 g of metabolizable protein. This follows if one uses as an example a ration that meets the NRC maintenance requirements for energy and digestible protein (1.6 g/kg⁷⁵). Converting digestible protein to CP with the regression (Knight and Harris 1966) Digestible protein = .877 (% CP) - 2.64, and by knowing that metabolizable protein is equal to about 75% of CP when low protein rations are fed, then it can be shown that metabolizable protein is approximately 1.5 times the amount of digestible protein under maintenance feeding conditions, or 2.4 g/kg⁷⁵.

(b) The protein content of a kg of body weight loss or gain is .15 kg. During early lactation each kg of body weight loss would contribute .15 kg metabolizable protein to the overall supply, and during later lactation when body reserves are being replenished and fetal growth is occurring, each kg of body weight gain would result in

deposition of .15 kg protein. Using these values together with typical weight changes (see figure 13) results in a larger amount of metabolizable protein than suggested by Coppock et al (1968) but somewhat less than suggested by Paquay et al (1972). Paquay et al suggested the mature cow is able to store and then lose 15 kg or more of body protein, but some of their observations were made with atypically large weight changes, and it is questionable whether they apply to the lactating cow. The assumption that 15% of body weight is protein essentially agrees with body composition data from studies with cows (Reid et al 1955).

(c) The efficiency with which metabolizable protein is utilized for milk production and body weight gain is 60%. This is an estimate. It is used in this instance to facilitate calculation of a metabolizable protein requirement during different stages of lactation. If this assumption is an error, it will affect the absolute requirement that is calculated, but it will not interfere with our immediate objective of illustrating how the dietary CP requirement, when expressed as a percentage of dry matter consumed, may change relative to stage of lactation.

(d) Milk contains 3% true protein. This is a good value for milk from Holsteins, but may be low for other breeds.

With these assumptions, the amount of metabolizable protein required was calculated by summing the amount needed for maintenance, milk production, and body deposition. If there was a loss in body weight, protein mobilized was credited to the supply of metabolizable protein. The "standard curves" of figure 13 were the source of information for milk production, feed intake, and body weight values.

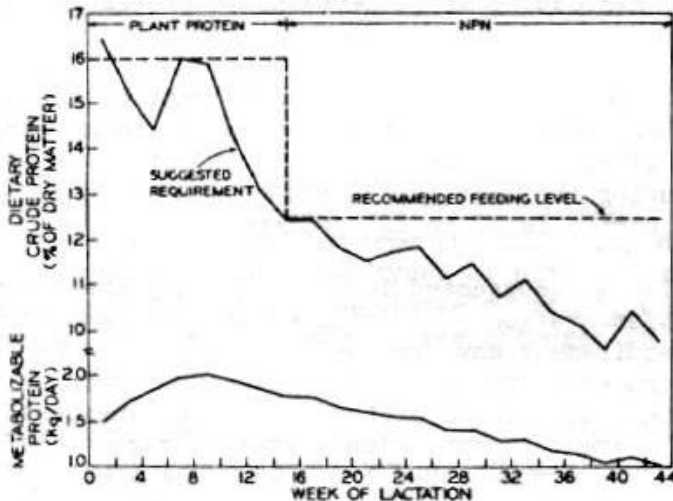
Figure 14 shows the proposed requirements of metabolizable protein and the amount of CP (% of dry matter consumed) needed to supply the metabolizable protein during different stages of lactation. The proposed requirements are very tentative, and subject to modification as information becomes available.

It appears from calculations based upon metabolizable protein that the minimum amount of protein needed, when expressed as percent dietary CP averages 15 to 16% (DM basis) in early lactation, and drops to as low as 10 to 11% in later lactation. There is an anomalous reduction in percent CP required around the 5th week, with an apparent rebound to a higher value on the 7th week. This is result of the abrupt change from a large daily weight loss during the 5th week to a small daily weight gain during the 7th week (figure 13). Since the amount of mobilized protein is calculated from weight change, the supply of mobilizable protein changes with equal abruptness. Though mobilization of tissue protein bears a general relationship to weight loss, it is unlikely that the weight loss curve reflects exactly the availability of mobilizable protein. The apparent reduction in percent CP required during weeks 3 to 5 is therefore probably distorted.

An error in estimating mobilizable tissue protein can have an important effect on protein requirements in early lactation. Coppock et al (1968) have estimated that cows

of the average size referred to in this study (figure 13) would have about 5 kg mobilizable protein. The estimate used in these calculations was approximately 7 kg. If 5 kg is used in the calculations instead of 7 kg, then the amount of CP in the ration (figure 14) would have to be increased by 1.2 percentage units during the first 5 weeks to supply the needed metabolizable protein.

Figure 14:
Effect of stage of lactation on dietary protein requirements



Recent studies designed to examine the effect of dietary protein level in early lactation indicate (Gardner and Park 1973; Grieve et al 1974; Sparrow et al 1973) that milk production is increased 10 to 20% when dietary CP is elevated from about 13 to 15% CP to approximately 16 to 19%. It is important to note that these experiments were initiated at or within 2 weeks of parturition, thus ensuring that cows were on experiment during the peak requirement period. There is little information available with cows of above average productivity and in early lactation that can either support or refute these striking observations.

Much more information about protein requirements in early lactation is needed. There are some cows that produce very well when fed low protein rations. Thomas (1971) reported that the two highest individuals in a group fed a 10.5% CP ration (air dry) produced 7,916 and 9,537 kg milk in 305 days. This is as striking as the benefits obtained from high protein supplementation mentioned earlier. It is noteworthy, however, in terms of metabolizable protein, that the actual difference between a ration containing 14.5 and 17.0% CP is about the same as between rations containing 10 and 11% CP. Due to this and the lack of sensitivity in measuring animal response, it will not be surprising to see considerable disparity in response to what appear to be greatly different protein levels when rations containing in excess of 12 to 13% CP are fed.

The protein requirement for cows in later lactation is much lower than in early lactation. As suggested in figure 14, the requirement drops to 12.5% around the 15th week of lactation, and gradually decreases after that. Thomas (1971) concluded that the bulk of evidence from published studies indicated that cows producing less than 20 kg/day performed satisfactorily when rations containing 11.5 to 12% CP.

The available evidence suggests that the current NRC recommendations regarding protein supplementation are reasonably accurate early in lactation, but result in over supplementation in later lactation. Table 3 of the NRC publication is particularly misleading as it pertains to protein supplementation recommendations.

As indicated in figure 14, true protein (plant protein) must be used in early lactation if the higher levels of supplementation are to be effectively achieved. Nonprotein nitrogen may be the major, if not sole, source of supplemental nitrogen during the last two-thirds of lactation when the required CP content of the ration is 12 to 13% or less.

The dotted line in figure 14 is a suggested level of CP supplementation. The suggested level of supplementation is maintained at a minimum of 12.5%, even though the proposed requirement may drop well below that. The reason for this is to provide the rumen microorganisms with enough ammonia to maintain maximum growth potential. This can be done rather inexpensively with a NPN source, and may prevent a depression in ration digestibility. More research is needed to determine whether there would be a significant reduction in digestibility by going to as low as 10% dietary CP, but it is conceivable that digestibility of cellulose in high forage rations could be reduced. This point would not be as important with feedlot cattle fed high concentrate rations, for a reduction of starch digestibility in the rumen could be compensated for by increased starch digestion in the intestine.

An attempt was made to get some measure of how better a protein requirement scheme based upon metabolizable protein would be than our present system based upon CP ($N \times 6.25$). A regression analysis was made of milk production and protein intake on data from published studies comparing milk production studies comparing milk production when either a negative control (10% protein) or treatment (added NPN or protein) ration was fed. The results are shown in table 3. When plant protein was fed, use of metabolizable protein instead of CP improved the r^2 value of the regression from .34 to .42. When NPN was supplemented, use of metabolizable protein instead of CP increased the r^2 value from .15 to .34. Quite a number of factors other than percent dietary protein were affecting milk production in this diverse group of studies hence the low r^2 values. The significant improvement in the regression obtained by using metabolizable protein instead of CP, particularly when NPN was the source of supplemental nitrogen, suggests that use of the term is worthy of consideration.

The calculation of protein requirements in terms of metabolizable protein, with final expression in terms of CP, improves accuracy yet retains the simplicity of the CP designation. It is an approach that recognizes the nitrogen needs of the rumen bacteria, as well as the host ruminant, and has the requisite features of being easily understood by people in the livestock and feed industry.

Table 3:
Relationship between percent increase in milk production (Y) and percent increase in dietary crude protein (CP) or metabolizable protein (MP)

Type of supplement	Number of observations	Predictive equation	r ²
NPN	38	Y = -0.76 + 0.36 CP	0.15
	38	Y = 1.24 + 0.52 MP	0.34
Protein	51	Y = -0.18 + 0.39 CP	0.34
	51	Y = 0.85 + 0.44 MP	0.42
Combined	89	Y = -0.32 + 0.37 CP	0.23
	89	Y = 1.10 + 0.47 MP	0.37

Implications for the Dairy and Feed Industry: If dairymen adopt feeding practices that are implied from the foregoing discussion, the feed industry will need to supply two types of nitrogen supplements. One would be all-plant protein, the other largely, if not all, NPN. The demand for supplements where 15 to 40% of the nitrogen is of NPN origin should diminish greatly.

Dairymen with larger herds may need to reconsider the old question of whether to group cows and feed each group more according to need. With historically low prices for feed grains and protein supplements, overfeeding and inefficient utilization of nutrients may not be as tolerable in the future as it has been in the past. Lactating cows would logically be divided into at least two groups one group for cows in the first third of lactation, and a second group for cows in the latter two-thirds of lactation. The first group would be supplemented with plant protein to raise protein content of the ration to approximately 16 to 17% (DM basis), while the second group would be supplemented with largely NPN to raise protein content to 12 to 13% CP.

Grouping of cows may prove advantageous for utilization of other nutrients. The relationship between milk production and feed intake as lactation progresses applies to other nutrients as well. All things considered, grouping of cows on the basis of stage of lactation deserves more attention. Dairymen with small herds have an obvious disadvantage when it comes to grouping cows, but other options exist, such as topdressing or special supplements in the milking shed.

Research nutritionists interested in protein requirements must look more closely at the first few weeks of lactation. Traditionally, cows are placed on experiment following achievement of peak production. By the time enough cows can be assimilated for an experiment, many of the cows have passed the critical period. Statisticians may loathe the variability of lactation between cows in those early weeks, and the need to use relatively insensitive experimental designs, but there may be no alternative to requiring large numbers of animals per treatment if reliable answers are to be obtained.

REFERENCES

- Burroughs W, Trenkle A H and Vetter RL 1973 AS Leaflet R173: Iowa State University, Ames
Coppock C E, Tyrell H F, Merrill W G and Reid J T, 1968 Proc Cornell Nutrition Conference:
Cornell University, Ithaca, N Y
- Everson, R A 1973 Ph D Thesis:University of Wisconsin, Madison
- Gardner R W and Park R L 1973 J Dairy Sci 56:390
- Grieve D G, MacLeod G K and Stone J J B, 1974 J Dairy Sci 57:633
- Knight A D and Harris L E 1966 J Anim Sci 25:593
- Paquay R, DeBaere R and Lousse A 1972 Br J Nutr 27:27
- Preston R I 1972 Proc University of Nottingham Nutrition Conference for Feed Manufacturers:
Vol 6 Swan H and Lewis D, ed
- Reid J T, Wellington G H and Dunn H O 1955 J Dairy Sci 38:1344
- Roffler R E and Satter L D 1975a J Dairy Sci 58:1880
- Roffler R E and Satter L D 1975b J Dairy Sci 58:1889
- Satter L D and Slyter L L 1974 Br J Nutr 32:199
- Satter L D and Roffler RE 1975 J Dairy Sci 58:1219
- Sparrow R C, Hemken R W, Jacobson D R, Button F S and Enlow C M
1973 J Dairy Sci 56:664
- Swanson E W, Hinton S A and Miles J T 1967 J Dairy Sci 50:1147
- Thomas J W 1971 J Dairy Sci 54:1629
- Trimberger G W, Tyrell H F, Morrow D A, Reid T J, Wright M J, Shipe W F,
Merrill W G, Loosli J K, Coppock C E Moore L A and Gordon C H 1972 New York's Food and
Life Sci Bull No 8 Cornell University, Ithaca, N Y

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