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MEASURES OF PROTEIN QUALITY FOR RUMINANTS

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SUMMARY

Nitrogen is the main nutrient required for synthesis of protein in animals tissues, secretions, products and maintaining its active and effective biomass.

These nitrogen requirements should be covered efficiently in proper quantities and forms in order to achieve efficient biological transformation of nitrogenous inputs to the end products. According to our recent knowledge in nitrogen metabolism these requirements should be criticized and expressed carefully. Also, the nitrogenous feed resources should be evaluated depending upon the availability of its nitrogen content to be absorbed, utilized, assimilated and transformed to animal own body nitrogen pool.

The major characteristics in protein assimilation, unique to ruminants, that must be considered are:

1- The massive proteolysis and protein synthesis that occurs cranial to the major digestive and absorptive sites.

2- The extensive utilization of nonprotein nitrogen for amino acid synthesis by microorganisms and the subsequent digestion and absorption of these compounds.

3- The recycling of metabolically produced nitrogen through this protein synthesis mechanism.

In this keynote paper, the logistic pathways of nitrogen flow, amount and form of dietary N, extent of proteolysis, microbial growth cycle, digestibility, nitrogen balance and related measurements are briefly investigated.

INTRODUCTION

Protein nutrition for ruminants is of great importance for it's effect on production efficiency system. Our knowledge in ruminant nutrition as a whole and protein nutrition in particular are increasing extensively since, nineteen seventies. These new ideas and concepts should be applied in order to reduce resources use and cost of production in each unique production setting.

Animal performance will be restricted to the nutrient allowed by the lowest level and the energetic efficiency will be influenced by amino acid balance.

The accurate prediction of requirements for a particular production level depends on the ability to predict, both requirements and supply of ruminal degraded carbohydrate, protein, microbial growth, metabolisable energy, net energy, metabolisable protein and limiting amino acids.

Feed resources should be evaluated and ranked according to it's ability and efficiency by which it supply the animal with the different feeding nutrients in the

proper form and balances.

EVALUATION OF PROTEIN

In ruminant animals the dietary protein undergoes considerable degradation and synthetic processes, in the reticulo-rumen vat, so that the material which finally becomes available for digestion and assimilation by the animal tissues is actually different from that originally was found in the diet. It is a fact that the different nitrogenous compounds are both incorporated and produced in a variety of reactions during digestive processes. Therefore it is more accurate and rational to use nitrogen as the basis for evaluation of feed resources than using true protein.

Nitrogen metabolism in ruminants is more than just amino acid supply and response to amino acid absorbed. So that all forms of N must be considered and the interconversions of these forms are necessary for the whole picture of nitrogen evaluation in ruminants.

1- Protein (N) content

In such situation the first stage of evaluation is the determination of crude protein (total N) concentration in the feeding stuff. It is the easiest way to rank the feeds according to it's N content, but it doesn't give any idea about the efficiency by which it is absorbed, utilized and converted to meet the animal requirements.

2- Digestible protein (N)

The term dietary digestible nitrogen calculated as the difference between N intake and that excreted in feces is misnomer, since it is not the digestion of the material that is being measured. There are shunts and alternate routes of excretion. Difference between levels of N in the feces and in feed hasn't anything to do with the supply of amino acids to the tissues of the body because of:

a- Endogenous fecal losses that vary with the total intake of dry matter, or in other way of expression, with the total amount of dried feces.

b- The shunting of nitrogen through the fermentative processes to pathway of excretion that doesn't correspond to the expected route.

c- The microbial activity in the digestive tract, results in a considerably greater source of refractory nitrogen product that come from microbial sources.

d- The apparent digestibility of N has been shown to decrease as the N content of the feed decreases and to increase as the NPN content in the diet increase.

It is to stress that N leaving the rumen via omasum has little relationship to the dietary amino acid intake in the food. In fact, the determination of N loss during digesta passage through rumen my be the only way for estimating the unmetabolised urinary N (Asplund, 1994).

3- Metabolisable protein

It is based on rumen degradable, undegradable and microbial proteins. Metabolisable protein is that part of the dietary N which is absorbed by the host animal and is available for use at tissue level. It consists partly of dietary true protein which resist degradation in the rumen but is broken down to amino acids and subsequently absorbed from the small intestine. Microbial protein synthesized in the rumen, similarly contributes to metabolisable protein.

Satter and Roffler (1977) proposed a diagram for metabolisable protein that could

be obtained from dietary protein, considering some assumptions, that;

a- The NPN proportion in the diet is 15%.

b- The degradability of dietary true protein is 60%.

c- The digestibility of undegradable protein is in the order of 88.23%, while the digestibility of true microbial protein is in the order of 80.36%.

d- The fermentable metabolisable energy is available for full efficiency of microbial protein synthesis.

e- The non amino N in crude microbial protein (MCP) is 20%.

The metabolisable protein measurement require accurate determination of duodenal flow and microbial N. The endogenous fraction represents about 50 - 200 g/kg of duodenal N, but it is difficult to quantify. In addition measures of degradability is a subject of dietary considerations such as level of feeding, size and frequency of meals. It has been calculated that estimates of degradability may vary over a range of ±0.3 to 0.35 owing to errors in it's determination. However it is the only method currently available.

The metabolisable protein is calculated as the sum of digestible microbial true protein plus the digestible undegraded protein.

The digestible microbial true protein = MCP x 0.75 x 0.85 = 0.638MCP.

The digestible undegradable protein = 0.9(UDP-6.25 ADIN).

The microbial crude protein ranges between 9 - 11 g/MJ fermentable metabolisable energy according to the feeding level as times of maintenance level 1-3.

4- Net protein

It is the net value of metabolised protein that is used by the host animal tissues with different efficiencies to cover it's needs for maintenance, protein deposition, pregnancy, lactation, wool production and other functions. It is reviewed that the efficiency by which the ideal amino acid mixture is used equals 1 for maintenance and 0.85 for other synthetic functions (AFRC, 1993).

In practical feeding the absorbed amino acid mixture is used by relative efficiency value (RV) in comparison with the ideal amino acid mixture. These RV values are 1.0 when the amino acid mixture is used to meet basal endogenous losses (Maintenance) and pregnancy requirements, while the RV is 0.8 for lactation, 0.7 for protein deposition and 0.3 for wool production. Accordingly the efficiency (Kn) by which the metabolisable protein (MP) is used for these functions is calculated by multiplying the efficiency of ideal amino acid mixture x RV values. The findings are;

a- For Maintenance (Knm) = 1 x 1 = 1

b- For pregnancy (Knc) = 0.85 x 1 = 0.85

c- For Lactation (Knl) = 0.85 x 0.8 = 0.68

d- For Growth (Kng) = 0.85 x 0.7 = 0.59

e- For Wool production (Knw) = 0.85 x 0.3 = 0.26

Then the net protein value is the product of metabolisable protein times the efficiency of transformation (Kn).

The logistic Pathways of nitrogen flow in the ruminants

It was proposed by Schoenheimer (1942) that protein metabolism in the living cells is in a constant state of flux with synthesis, degradation and other transformations occuring at rapid rates "Dynamic state of N". It deals with the supply and form of N as delivered to and removed from the actively metabolising tissues.

Amount and form of dietary N

Not only the absolute amount of dietary N is important, but the amount relative to other nutrients is also critical. A minimum level of N in the form of NH_3 is necessary for the maximum growth and activity of rumen microbes.

Satter and Slyter (1974) suggested that this level is 5 mg/100 ml rumen liquor for maximum microbial growth. Mehrez *et al.* (1977) reported values ranged from 13.5 - 23.5 mg NH_3-N / 100 ml rumen liquor for maximal rate of fermentation in the rumen for diets varies in concentrate : roughage ratios. However, that level almost varies between diets as well as with the diurnal eating and drinking paterns of the animal. In addition to NH_3-N, individual amino acids and peptides are required by rumen microbes for maximum fermentation capacity (Argyl and Baldwin, 1989).

Recycling N

The recycling of N to the reticulo-rumen (RR) is a very active and well controlled process. Recycling appears to act as a buffer process to keep the N level in the RR adequate for microbial activity.

The primary mechanism is ammonia or urea diffusion through the RR wall. This is a two way diffusion. The direction and extent of which is governed by the concentration of ammonia in the rumen contents and urea in blood. This diffusion process acts as to stabilize ammonia concentration in the RR at adequate, but not excessive levels.

Recycling, also serves as a mechanism for conversion of N when low N diets are consumed.

There is some recycling of N through the saliva, however N from the saliva may be an important reintroduction of this nutrient during a critical time in the fermentation process.

Extent of proteolysis

Protein entering the RR has three possible pathways relating to the proteolytic process (AFRC, 1993).

1- It will be degraded quickly or slowly.

2- It will not be degraded in the RR, but it will be digested caudal to the RR.

3- It will be undegraded in the whole digestive tract.

Proteolysis is the first and most decisive determinant of the fate of the dietary protein.

The term undegradable or escape proteins and many other nominations are expressions for the dietary protein resistant to microbial attack, while the term "by-pass" is not entirely accurate and should be reserved for the reticular groove.

The manipulation of the extent and rate of degradation of dietary N sources offers the only practical way of changing either the amount or the quality of protein leaving the rumen. This fraction is not constant even with a specified protein, because rates of degradation vary with many variables such as rate of passage and rumen conditions (Gainsworthy, 1989). It should be expected that, with improved methods of analysis to diffrintiate between microbial and undegraded protein and their rates of flow to the abomasum, it will be possible to understand more fully the influence of amino acid supply to the metabolism of the ruminant.

On the other hand if the protein is degraded, it has the possibility of multiple routes of passage. Peptides and amino acids as well as the end products of their

degradation (mainly NH_3), are all excellent sources of N for the metabolism of rumen microbes. They enter the microbial nitrogen pool. However, this pool is not all amino-N, but it is also incorporated into nucleic acids and other N compounds characteristic of the cell. About 20 - 30 % of the microbial N is sequestered from protein metabolism in this way. More over if the free amino acids are used as energy substrate for microbes, NH_3 may be generated and absorbed readily across the rumen wall if it is not used by microbes and removed as a candidate to enter the microbial pool. The term unmetabolised urinary N has been used to describe this fraction (Asplund, 1994).

The efficiency of N capture depends on the degradability of the dietary N constituents and also upon the synchronisation provision of readily available energy in the form of carbohydrates in the diet.

The speed and extent to which protein can broken down depends upon;

a- The surface area available for microbial attack.

b- The physical consistency and chemical nature of the protein.

c- The protective action of other constituents.

It would appear that the value of protein to ruminants depends as much upon overall dietary considerations, as upon the nature of the protein it self. The value will thus differ for any given dietary situation and any attempts to allocate a single accurate value to any protein source is foredoomed to failure.

Measurements of NH_3 concentration in the rumen reflect the balance achieved between protein breakdown and synthesis under practical dietary circumstances and could be useful for predicting the value of proteins in particular dietary combinations without the considerable commitment involved in balance trials.

Measurements of degradability

a- Measurements of duodenal flow

It could be done either by using a single canula with digesta marker or using the reentrant canula. This method is still being used and will continue to be used as a useful technique. However for routine measurements of protein degradability it is unsuitable for three main reasons:

1- The animal has to be given the diet in question for a considerable time before measurements can be obtained.

2- Because of the analytical work in separating microbial and dietary protein, which is tidious and in some times it should be corrected for endogenous concentration.

3- The single measurement gives no information on the dynamic of out flow rate and therefore it is suitable only for conditions in which the same feeding level and feeding system is used.

b- Use of nylon bags technique incubated in the rumen

The degradability and rate of degradation can be calculated from the formula

$$P = a + b\,(1 - e^{-ct})$$

where. P = degradability during time t. a = The soluble protein of the sample (quickly degradable protein of the diet). b = The degradation potential.
c = The degradation rate.

However the rate of digesta outflow has a great effect on the degraded protein of

the diet, therefore it should be included and the formula is rewritten to be;

$$P = a + \frac{bc}{c + r}$$ where: r = The rate of outflow.

c- Laboratory measurements of degradability

Laboratory methods with no use of animals would be preferable and a great deal of efforts is being directed toward developing reliable method. So far measurement of solubility have failed to determine the fraction that will degrade in the rumen.

In vitro measurements with rumen fluid appears most suitable. The rumen fluid must be obtained from animals given similar diets to those which the results need to be applied and the rate of degradation must be described.

Protein from various feed stuffs may be classified into three ruminal undegradable categories as follows:

1- Low undegradable protein (less than 40%) Soybean meal.

2- Medium undegradable protein (40 - 60%) Cotton seed meal.

3- High undegradable protein (over 60%) Meat meal, Corn gluten, Blood meal, Feather meal and Fish meal.

It should be cleared that increasing the undegradable protein does not ensure increased animal production, since;

a- The undegradable protein may be poorly digested in the small intestine.

b- The balance of amino acid of post ruminal-N may be poor relative to the tissues requirements.

c- Energy suppornutrients other than amino acids may limit the animal production.

Microbial protein

The average value of microbigrowth yield reported by AFRC was 32 N/kg organic matter digested in the rumen, while Demeyer and Van nevel mentioned a value of 35g N/kg (1979).

It is stated that mixed rumen bacteria grew faster and with peptides, rather than with free amino acids, but only at concentration of 10 mg/l (Argyl and Baldwin, 1989). Amino acid breakdown may be nutritionaly expensive not only because amino acid lost, but also because of the high energetic coast of this breakdown.

The value of microbial protein synthesised depends on how much dietary protein escapes breakdown in the rumen and how much microbial protein is formed, as well as the amino acid composition of both. Lysine, methionine and histidine in forages and cereals are generally lower than in animal products. Supplementation with fish meal and to a lesser extent soybean can help correct these imbalance. Different proportions of these limiting amino acids are released from different bacterial cells by the action of pancreatic enzymes (Bergen *et al.* 1967).

Gram positive bacteria are more resistant to digestion than Gram negative cells, which results in a trend toward a lower digestibility of microbial protein as the proportion of Gram positive bacteria increase (Wallace, 1994).

The amino acid content of microbial protein of different microbes is fairly constant, however it was reported recently that yeast culture stimulated protein flow in dairy cattle and that protein contained higher proportions of limiting amino acids (Lysine and Methionine) than the control animals (Erasmus *et al*, 1992).

Feed additives

1- Ionophores

Several ionophores including, ,monensin, salinomycin, lasalocid and tetronasin improve feed efficiency in ruminants via their effects on microbial population. Ionophores tend to decrease NH_3 production and increase protein flow out of the rumen.

Although it was originally reported that protein breakdown was inhibited by monensin, it now appears that it is the later steps in the degradation process that are affected, namely peptide and amino acid breakdown. The ionophores are most toxic to Gram Positive and the effects of ionophores are usually interpreted in terms of the elimination of these bacteria from the rumen.

Peptides and amino acids accumulation would expected to be beneficial. Firstly, because rumen microbes would be able to use preformed amino acids saving the energy required for their biosynthesis. Secondly, because some of these compounds may escape degradation in the RR and utilised by the host animal tissues.

2- Microbial feed additives

Two main additives are used for adult ruminant. One based on the yeast (*Saccaromyces Cervisiae*) and the other is a fermentation extract of the filamentous fungus (*Asparagillus Oryza*). In some studies microbial yield is improved, while in others the amino acid composition of protein flowing from the rumen is improved.

Post ruminal supply and requirements

The host animal tissues requirements can be subdivided into specific metabolic costs. These include metabolic fecal N, endogenous urinary N, scurf loss, and synthesised products containing N that include tissues growth, foetal growth, milk production and wool production.

Each of these would be covered by a specific efficiency (kn) from the metabolised N according to the balance of amino acids mixture available and required for these several functions.

CONCLUSION

1- It is useful to use the updated concepts of metabolisable and net protein in the future work for protein evaluation and nutrition in ruminants.

2- If these concepts based on scientific principals are accepted, then it is to encourage research work to generate the data required to apply it.

3- The wise use of undegradable and protected protein sources would help to improve production.

4- Applying degradability informations on forage proteins to improve it's nutritive properties for ruminants and using it as a breeding index in forage cultivations is of great value.

5- Our knowledge on fluctuation and dynamics of the rumen microbial population, particularly in those involved in protein metabolism is week and need to be strengthen.

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