Modifying Proteins for Maximum Utilization in the Ruminant[1]

A. DARE McGILLIARD, Department of Animal Science, Iowa State University, Ames, Iowa 50010

ABSTRACT

Much of the early work on nitrogen metabolism in ruminants has clearly established that the rumen microflora have a considerable modifying effect on the utilization of dietary nitrogen. This modifying effect may be advantageous to the animal under certain conditions, i.e., when the diet contains mainly poor quality protein or a nonprotein nitrogenous material such as urea. On the other hand, this modifying effect may not be advantageous when high quality protein is fed. From a practical standpoint it appears that limiting the degradation of dietary protein in the rumen would be advantageous under most conditions. Chemical modification of good quality proteins or coating of proteins and individual amino acids to render them resistant to microbial attack, without greatly reducing their nutritive value in the small intestine, appears to offer the most promising approach.

The digestion of nitrogenous compounds by ruminants is a complex process and until about 1950 many of the pathways were not well understood. Since then considerable work has been done in this field so that, at least qualitatively, the pathways involved are reasonably clear (1-5). Quantitative information, however, is still rather limited.

It seems most convenient for the purpose of this discussion to consider ruminant digestion as two interrelated processes, one being microbial digestion in the reticulum, rumen and omasum, and the other being hydrolytic or enzymatic digestion in the abomasum and intestines.

DIGESTION OF NITROGENOUS MATERIALS IN THE LOWER GUT

Few experiments have been conducted to study digestion in the lower gut (abomasum and intestines) of the ruminant. It has been provisionally assumed that digestion in this part of the tract is similar to that in nonruminants. This assumption is not necessarily valid because a number of conditions are quite different (6). For example, (a) diverse proteins in the diet are converted by microbial activity to a protein of more uniform composition, and most of the carbohydrate is removed from the digesta so that little sugar is presented to the intestine; (b) flow of digesta to the abomasum and to the intestine is nearly continuous and relatively constant in consistency and composition; (c) abomasal secretion of acid is continuous, and the intestinal contents remain acid through the upper part of the intestine; and (d) large amounts of water and electrolytes secreted into the gut in the saliva, gastric juice, bile and pancreatic juice must be efficiently reabsorbed in the small and large intestines. It is possible that these factors could have peculiar effects on the absorption of metabolites from the intestines. However, because there is no real evidence to the contrary, it is probably valid to assume that the digestion of protein in the abomasum and intestines is similar to that in nonruminants.

DEGRADATION OF NITROGENOUS MATERIALS IN THE UPPER GUT

Most of the nitrogenous material ingested by the ruminant fed natural feed consists of proteins. Although it is unusual for dietary protein to be completely degraded upon entering the rumen, these proteins are extensively hydrolyzed by rumen bacteria to their constituent amino acids, which are then rapidly deaminated with the formation of ammonia. The rate of proteolysis in the rumen is closely related to the solubility of the protein in rumen fluid. The rate of deamination is related to the level of protein in the diet. Most species of rumen bacteria prefer ammonia to amino acids or other more elaborate compounds as a nitrogen source for body protein synthesis, and some species have an absolute requirement for it. Mixed rumen bacterial populations digest starch more efficiently in the presence of ammonia than in the presence of amino acids, so it seems probable that ammonia is the main nitrogenous nutrient for bacterial growth. On the other hand, the protozoa in the rumen are primarily ciliates, and available evidence indicates that ciliates in general do not use ammonia as the major source of nitrogen. It is most likely that these organisms obtain their nitrogen by engulfing and digesting bacteria or particles derived from the food, such as chloroplasts, and by actively taking up free amino acids, purine and pyrimidine bases.

Urea and other nonprotein nitrogen compounds are widely used in ruminant diets. These additives may provide all of the dietary nitrogen (7) but in practice not more than 40% is normally used without deterioration of animal performance. The rapid conversion of urea to ammonia in the rumen has been repeatedly demonstrated with the urease activity restricted to the rumen bacteria.

Ammonia, therefore, is a prime intermediate in the conversion of dietary nitrogen to microbial nitrogen. Excessive rates of ammonia production can occur if large amounts of urea or readily soluble protein are ingested. If the rate of production exceeds the rate at which the bacteria can utilize ammonia, concentration of ammonia in the rumen increases. This is particularly evident if the diet is deficient in readily available carbohydrate such as starch. Ammonia in excess of that utilized by the bacteria may be absorbed from the rumen, converted to urea and excreted in the urine, thus representing a substantial loss to the animal. Although this loss clearly depends on composition of the diet, it may be as great as 50% of the nitrogen intake. Urea can also move from the blood to the rumen both in the saliva and by direct transfer across the rumen wall, the latter process being of greater quantitative importance (8). This process would seem to be of greatest benefit to animals on low nitrogen intake.

Nitrogenous substances presented to the abomasum and intestines of the ruminant thus consist mainly of those present in the bacteria and protozoa, though variously supplemented with undigested food residues and digestive secretions. Free amino acids in the rumen contents are usually in such low concentration that they do not form an important source of nutrients to the ruminant either by absorption from the rumen or by flow to the abomasum.

[1]Journal Paper No. J-6943 of the Iowa Agriculture and Home Economics Experiment Station, Ames, Iowa. Presented at the Porter Williams Symposium on Proteins, AOCS Meeting, Houston, May 1971.

The daily protein output from the rumen and the magnitude of the ratio of the microbial, food residual and endogenous proteins are of primary interest in the protein metabolism of ruminants. Measurement of these parameters has proved difficult and the analytical problems are not fully resolved. Several experiments have demonstrated that the protein output from the rumen may be greater or less than the input (9-12). The output can be reduced below input by excessive ammonia production and absorption, especially when nitrogen content of the diet is high. In contrast, when nitrogen content of the diet is low, the addition of endogenous nitrogen to the rumen can provide an extra source of nitrogen for microbial growth and total protein output is thereby increased above input.

Recent studies with sheep by Pilgrim et al. (13) indicate that synthesis of microbial protein is more dependent on ammonia as a starting point with low nitrogen diets than with high nitrogen diets. The leveling effect of the microbial activity in the rumen is clearly demonstrated by the experiments of Hogan and Weston (12). In their studies, sheep were fed high protein (19.8%) or low protein (7.8%) diets and they found that protein output from the rumen was much the same. One can conclude, in general, that the changes which occur in the forestomach are advantageous to the animal when its diet contains mainly poor quality protein or a nonprotein nitrogenous material such as urea, but may be disadvantageous when good quality protein is eaten. Three factors, then, are clearly important in protein utilization by ruminants: the extent to which dietary protein is or is not degraded in the forestomach, quantity and quality of the microbial protein synthesized from dietary and endogenous nitrogen, and amount of endogenous protein passing to the intestines.

The change in quantity and quality of protein between the diet and the duodenum is perhaps the most important aspect of ruminant nitrogen metabolism. Results from a number of experiments indicate that rumen output of microbial protein is a function of the amount of organic matter fermented in the rumen and thus of the amount of energy available to the microbes for their growth. There is general agreement that, with most diets of hay or concentrates or both, in which ammonia accumulation is quantitatively unimportant, about 50-80% of the dietary N is converted to microbial compounds by the time the digesta enter the duodenum. Hogan and Weston (12), in experiments with sheep receiving either high or low nitrogen diets, calculated that somewhat less than 15 g microbial protein was synthesized for each 100 g organic matter fermented in the rumen. This value is roughly in accord with those calculated by Walker (14), Hungate (2), Pilgrim et al. (13) and Walker and Nader (15). One can conclude from the accumulated evidence that fermentation of protein, when provided in excess of the synthetic capacity of the microorganisms, causes extensive loss of nitrogen to the ruminant.

BIOLOGICAL VALUE OF DIGESTA PROTEIN

It is now accepted as a general concept that biological value (BV) of a protein is determined primarily by its content of essential amino acids (16) and, specifically, by the content of that essential amino acid in greatest deficit relative to the animal's requirements. In the absence of adequate experimental data it is necessary to assume that ruminant tissues require assemblages of amino acids similar to those required by nonruminant tissues. There is evidence (17,18) that sheep and cattle are unable to synthesize the amino acids usually accepted as essential. It is clear from a consideration of digestive processes in the rumen that it is not valid to specify the BV of a given feed protein for a ruminant. It is the BV of the digesta protein presented to the small intestines that is nutritionally significant.

Various experimental approaches have been used in attempts to assess the amino acid requirements of ruminants. Estimates have been made from amino acid composition of the microbial protein by examining (a) rumen contents of animals fed diets in which the sole source of nitrogen was urea, (b) mixed microorganisms separated from rumen contents, or (c) rumen microorganisms grown in pure culture. Although there are some differences between sets of data, the amino acid compositions seem remarkably consistent. Evidently the effect of rumen metabolism is to reduce diverse dietary amino acid patterns to a relatively constant pattern in digesta entering the duodenum. Moreover the results show no obvious consistent deficiency in any one of the essential amino acids. Amino acid composition after acid hydrolysis, however, is not necessarily a good indicator of BV.

Nutritive value of rumen microorganisms has been determined by measuring their digestibilities and BV in experiments with rats. Rumen bacteria had a digestibility of 50-70% and a BV of 65-80%. Protozoa were 80-90% digestible with a BV of 65-80% (19-22). The higher digestibility of protozoa is presumably because the cell wall is more easily broken down. It is not known what relevance results with rats have to the situation in the ruminant. Bergen et al. (23) and Tannenbaum and Miller (24) indicate that BV as well as digestibilities of intact microorganisms may differ from the dried organisms fed to rats. Moreover, amino acid requirements of the rat probably differ from those of ruminants such as sheep growing a lot of wool (high content of sulfur-containing amino acids) and lactating cows producing large quantities of milk (casein contains a high proportion of lysine). Thus, it seems desirable to assess in the ruminant itself the adequacy or inadequacy of amino acid absorption from the gut.

An alternative to examining only the microbial nitrogen fraction of digesta entering the duodenum is to determine to what extent dietary protein influences the amino acid composition of the complete digesta. This approach was taken in sheep by relating dietary composition of the digesta to composition of digesta in the duodenum and ileum (10) and the rumen (25). Results of these two studies were similar and demonstrated, for the diet used, that rumen fermentation decreased proline, arginine and glutamic acid and increased lysine, tyrosine, threonine and isoleucine. The marked decrease in proline and increase in lysine were probably the most dramatic. The net effect in general was to make the amino acid compositions of the different digesta samples similar to each other even though the dietary compositions were markedly divergent. Although diet had some influence, that the digesta composition tended to approach that of microbial protein is consistent with the large (70-80%) conversion of dietary nitrogen to microbial nitrogen.

There have been a number of attempts to relate composition of the diet to concentrations of amino acids in the blood plasma of ruminants. Techniques devised for this purpose in nonruminants are not readily applicable to ruminants because they depend upon comparison with plasma amino acid concentrations under fasting conditions, a state not readily achieved in ruminants. Results obtained by this method therefore are difficult to interpret. Thus far, the values for plasma amino acid concentration have given no clear-cut evidence regarding adequacy of the amino acid mixtures absorbed from the ruminant small intestine.

An approach that has attained considerable emphasis in recent years is the supplementation of the digesta with specific amino acids or proteins by administering these materials into the abomasum or duodenum. In most studies nitrogen retention (as measured by balance procedures) has been used as the criterion of efficiency of nitrogen utilization, although in some, wool growth, tissue growth or milk production have been used. The early studies of

Cuthbertson and Chalmers (26) and Chalmers et al. (27) clearly showed that casein administered into the abomasum or duodenum of sheep was utilized more efficiently than when given in the diet. Similar results for lambs administered casein or soybean protein were obtained by Little and Mitchell (28). However, these investigators found that abomasally infused gelatin or zein failed to increase nitrogen retention over that for the orally administered controls. Colebrook and Reis (29) gave supplements (ca. 100 g protein per day) of casein, whole egg protein, egg albumin, maize gluten and gelatin to sheep via the abomasum and measured their effects on wool growth, body weight gain and nitrogen retention. Casein increased wool growth substantially. Wool growth with whole egg protein and egg albumin was about equal to that with casein, whereas maize gluten gave less than half the response, and gelatin gave little response at all. All protein supplements except gelatin effectively stimulated body weight gain and enhanced nitrogen retention. However, egg albumin was inferior to whole egg protein and maize gluten. The results strongly indicate that relative values of the protein for wool growth and for body weight gain and nitrogen retention are not the same.

Impressive evidence that specific amino acids may limit productive performance of a ruminant is seen in experiments of Reis and Schinckel (30,31) and Reis (32-34). These studies show that casein consumed in the diet of mature sheep had very little effect on wool growth, whereas casein infused into the abomasum increased the rate of wool growth up to threefold. Small amounts of cystine or methionine administered directly into the abomasum substantially increased wool growth and nitrogen retention. Abomasal infusion of casein gave much greater wool growth than did abomasally infused gelatin, and addition of cystine or methionine to either of the infused proteins significantly improved wool growth. This suggests that the sulfur-containing amino acids per se are important in promoting wool growth. Additional evidence concerning importance of the sulfur-containing amino acids for wool growth in sheep have been presented by Dreiden et al. (35) and Downes et al. (36).

Schelling and Hatfield (37) obtained a substantial positive nitrogen balance in lambs by infusing casein into the abomasum. In a subsequent study (38) they reported that abomasal infusion of a mixture of the 10 essential amino acids at the same levels as supplied by casein resulted in increased nitrogen retention. Additional evidence has been obtained (39) that indicates the pattern of amino acids offered post-ruminally affects the animals' response to the nitrogen source. More recently, Nimrick et al. (40) have presented data suggesting that the order of limiting essential amino acids for growing lambs fed urea as the sole nitrogen source was (a) methionine, (b) lysine and (c) threonine. Subsequent studies indicated the supplemental levels needed in the diet for maximum nitrogen retention were 0.10% methionine, 0.16% lysine and 0.10% threonine (41).

Devlin and Woods (42) infused lysine into the abomasum of steers and increased the nitrogen retention above that of control animals. Sibbald et al. (43) stated that they obtained marked increases in weight gain and nitrogen retention in steers receiving abomasal infusions of methionine or lysine or both, but they gave no data concerning level of the infusions or magnitude of the responses.

Our present knowledge of amino acid requirements of ruminants and the extent to which these requirements are met is rather limited. Microbial protein, which provides much of the amino acid supply, seems of good quality for ruminant growth, but there is good evidence that methionine and cystine may be deficient for maximum wool growth in sheep, and some evidence that methionine, lysine and threonine may be limiting for tissue growth. Although there is some speculation from existing data that lysine may be limiting for milk production in high-producing cows, no confirmatory evidence for this is available.

CONTROL OF PROTEIN DEGRADATION IN THE RUMEN

From the foregoing discussion it seems that a practical advantage might be gained by limiting or preventing the degradation of dietary protein in the rumen. It should be borne in mind, however, that this might lead to reduced microbial population and possibility of secondary effects such as reduced cellulolytic activity and volatile fatty acid production. The advantage of preventing degradation of protein in the rumen is clearly illustrated by Black (44). In this study he calculated the theoretical utilization of dietary protein for a 20 kg lamb whose entire diet was fermented in the rumen (ruminant lamb) and for another lamb whose diet was digested by host enzymes in the lower gut (nonruminant lamb). Based upon a daily gross energy intake of 1780 kcal and protein intake of 61 g, the net daily dietary protein value was 24.3 for the ruminant lamb and 45.4 g for the nonruminant lamb. Thus, in this example, fermentation of dietary protein in the rumen reduced the protein available for utilization by about 50%.

There seem to be several alternatives for reducing or preventing the degradation of proteins in the rumen so that they would pass to the abomasum and intestines for subsequent digestion. Feeding a natural protein such as zein, which is very insoluble in rumen fluid, reduces its degradation in the rumen (45) but it also is poorly digested in the small intestine (28). Thus the use of a protein of this type probably offers no particular advantage.

Procedures that increase the rate of passage of feed particles through the rumen may effectively reduce the extent of their fermentation in the rumen. Hemsley (46) indicates that a high salt intake might induce such a train of events. However, the long-term effects of high salt intake on animal performance and health are not known. McGilliard (47) obtained evidence that a low ratio of hay to concentrate in the diet may result in a preferential increase in rate of passage of the concentrate part of the diet from the rumen. In this study, it seemed that passage of the concentrate was considerably more rapid when the hay to concentrate ratio was 30:70 than when the ratio was 0:100 or 70:30. As a consequence, digestion in the lower gut was highest with the 30:70 ratio.

Dietary protein in liquid solution or suspension can be induced to bypass the rumen in young and adult sheep by encouraging closure of the reticular groove (48-52). These studies have demonstrated that utilization of a number of different proteins is greater when they bypass the rumen. Although this procedure would seem to have certain limitations from a practical standpoint, it seems to have considerable advantage for intensive rearing of young animals during the period when amino acid requirements are high and for research purposes in establishing amino acid requirements.

Perhaps the most promising approach proposed thus far is the modification of good quality protein to markedly reduce its susceptibility to microbial attack in the rumen without reducing its nutritive value in the lower gut. It has been demonstrated on a number of occasions that heating proteins (particularly severe heating that causes denaturation) reduces their nutritive value for nonruminants. On the other hand, Chalmers et al. (27) observed that heat treatment of casein decreased its rate of breakdown in the rumen, reduced ammonia formation and increased nitrogen utilization in sheep. Similar improvements of protein by heating were obtained by Chalmers et al. (53) with peanut meal for nitrogen retention by lactating goats, by Whitelaw et al. (54) with peanut meal for nitrogen retention and growth in calves and by Tagari et al. (55) with soybean

meal for nitrogen retention by rams. The main effect of heating was to reduce proteolysis and ammonia formation in the rumen, the advantage of which probably outweighs any reduction in nutritive value in the lower gut.

Recent studies by Ferguson et al. (56) have shown that treatment of casein with formaldehyde (4%) renders it resistant to microbial attack. The cross-linking of protein chains by formaldehyde is firmly bound under alkaline or neutral condition and becomes less tightly bound as conditions become more acid. The change of pH from about 6 in the rumen to about 3 or less in the abomasum and proximal duodenum can be exploited in this way. Ferguson et al. (56) observed that formalin treatment of casein, which markedly reduced its solubility at pH 6, almost completely prevented microbial degradation in the rumen contents in vivo and in vitro and increased wool growth about 70%. Digestion of the treated casein in the lower gut seemed unimpaired. Reis and Tunks (57) studied the effects of three types of casein supplements to an all-roughage diet on wool growth, body weight gain and nitrogen retention. The supplements were untreated casein in the diet, untreated casein by abomasum and formaldehyde treated (4%) casein in the diet. Formaldehyde treated casein and casein per abomasum were similar in nutritional value, and both were superior to untreated casein in the diet for all parameters studied. The treated casein was 90% digestible. In a subsequent study Reis and Tunks (58) found that treated casein in the diet and casein per abomasum increased the concentration of plasma amino acids and caused proportional increases in essential amino acids. In contrast, untreated casein in the diet had little effect upon either the concentration or proportions of amino acids in the plasma. Hughes and Williams (59) supplemented hay and grain diets of sheep at three levels of intake with 50 g of untreated casein or formaldehyde treated (1%) casein per day. The treated casein significantly increased liveweight gains at the low and medium levels of intake and increased wool growth at all levels of intake.

Rattray and Joyce (60) fed rations containing either meat meal or linseed meal, untreated or treated with formaldehyde (2-1/2%), to wethers in a nitrogen retention-growth study. Sheep fed treated linseed meal retained more nitrogen than did those fed untreated linseed meal, but the reverse was observed for meat meal. Digestibility of the treated meat meal was markedly reduced. Although differences were obtained in nitrogen retention between rations, no differences were observed in wool growth or liveweight gains. The reduction in digestibility of formalin treated meat meal seen by these authors has also been observed in calves.

Peter (61) has screened a number of aldehydes to determine which may be used effectively as treatments to depress protein degradation of soybean meal. All aldehydes were tested over a wide range of concentrations with a 5 or 6 hr incubation time. Formaldehyde, glyoxal and glutaraldehyde at concentrations of 0.6, 1.5 and 1.5%, respectively, of the soybean meal seemed most effective both in vitro and in vivo in reducing solubility of soybean protein and ammonia production by rumen microorganisms. Zelter et al. (62) have indicated that these aldehydes, at concentrations from ca. 0.15% to 3%, were effective agents for reducing bacterial breakdown of protein in peanut, soybean, linseed, rapeseed, sunflower and alfalfa meals, dried skimmilk and casein.

Nitrogen balance experiments (61) in which either formaldehyde, glyoxal or glutaraldehyde treated soybean meal was fed to lambs showed that these treatments significantly increased nitrogen retention. In a subsequent growth study with lambs, both formaldehyde and glyoxal treated soybean meal supported more rapid and efficient gains than did the control soybean meal. However, wool growth was not affected by treatment.

Amino acid analysis of soybean meal treated with either formaldehyde, glyoxal or glutaraldehyde revealed no significant differences for any amino acid except lysine. All these aldehydes reduced the lysine content of the soybean meal, measured after acid hydrolysis, below that of the untreated soybean meal. This finding was not expected because of the involvement of lysine in the protein-aldehyde complex; the binding of lysine seemingly was not completely reversed by acid hydrolysis.

In feedlot trials conducted with steers, an all-concentrate diet of high moisture corn was supplemented with soybean meal treated with either 1% formaldehyde, 2% glyoxal or 2% glutaraldehyde. Although rate of gain and feed efficiency with the treatments was improved over that of the controls, the improvement was not significant. In a second trial, in which formaldehyde or glyoxal treated soybean meal was used, no differences between treatments were observed. Clark et al. (63) in a study with lactating cows found that feeding formaldehyde treated (0.9%) soybean meal had no beneficial effect on milk or milk protein production.

Recently Downes et al. (64) evaluated a system for testing the effectiveness of various methods of protecting amino acids and proteins from degradation in the rumen. In this study doses of L-[^{35}S] methionine (2 g) and [^{35}S] casein (20 g) were given to sheep in the diet or via the abomasum and the patterns of ^{35}S-labeling in blood, wool and excreta were examined for the 7 days after administration of the dose. [^{35}S] casein treated with an 8% aqueous solution of formaldehyde gave results showing that the casein had been protected from degradation in the rumen without reducing its subsequent digestibility. On the other hand, treatment of [^{35}S] casein with 40% aqueous solution of formaldehyde gave a product that was completely indigestible.

It seems relatively clear from the evidence presented that formaldehyde, glyoxal and glutaraldehyde are effective agents for treating proteins to reduce their solubility and susceptibility to degradation in the rumen and to allow their subsequent digestion in the lower gut. It is equally clear that the optimal conditions for processing protein for maximum response are not well defined and that different proteins may respond quite differently to a given treatment.

Tannins are known to form hydrogen bonds between the hydroxyl groups of the tannin and the carboxyl groups of the protein peptide bonds, thus rendering the protein considerably less soluble. Zelter and LeRoy (65) proposed that dietary proteins could be treated with tannin to decrease their degradation in the rumen. They treated peanut and soybean meal samples with aqueous solutions of chestnut tannin (13%) and dried them. The treated meals were not degraded by rumen microorganisms in vitro whereas the untreated meals were rapidly degraded. Zelter et al. (62) extended these studies to include linseed, rapeseed and sunflower oil meals, dried skimmilk, casein and alfalfa meal and observed similar results. Pepsin and trypsin digestion of the peanut and soybean meals was not affected by treatment (65). Driedger (66) used tannins (aqueous solutions, 0-14%) from four sources (Allepo, Sumac, Tara, Quebraco) to treat soybean meal and also observed that pepsin digestion of the treated meals was not affected. However, meals treated with the different tannins differed in their susceptibility to pancreatin digestion, and increasing levels of a given tannin markedly reduced digestion.

Zelter and LeRoy (65) observed that when tannin treated peanut and soybean meals were fed to fistulated sheep, ammonia production in the rumen and plasma urea nitrogen levels were reduced. Delort-Laval and Zelter (67) found that tannin treated linseed meal and peanut meal increased nitrogen retention in lambs. Similar results with lambs fed tannin treated soybean meal were reported by

Driedger (66). However, results obtained from infusion of the treated meal into the abomasum indicated that the tannic acid-protein complex was not completely digested in the lower gut.

Zelter et al., in a personal communication cited by Driedger (66), reported that tannin treated linseed meal and peanut meal increased nitrogen retention in sheep and goats and were used more efficiently for milk production in goats. In feeding trials with steers Driedger (66) observed consistently greater average daily gains and feed efficiency for steers receiving tannin treated soybean meal than for those receiving untreated meal but the differences were not large enough to be significant.

From the availabile evidence there seem to be a variety of tannins that can be used to treat dietary protein to reduce its bacterial degradation in the rumen. The level of treatment required to obtain optimal protection is not well established and probably will vary with the source of the tannin and the protein being treated. There is some evidence that proteins treated with different tannins vary considerably in their susceptibility to hydrolytic breakdown in the small intestine.

Attempts to improve performance of the ruminant with dietary amino acid supplementation, either singly or in combination, have been inconsistent and for the most part unsuccessful. The inconsistencies seem related to the extent to which the amino acid escapes degradation in the rumen and to the basic composition of the diet. Available evidence suggests that positive responses to amino acid supplementation can be obtained provided that the amount of amino acid added as a supplement is large in relation to any likely deficiency. In recent studies with sheep in which the diet was supplemented with 5 g of methionine or lysine Lewis (68) found that 90% or more of the added amino acid was degraded in the rumen. Thus a small but perhaps significant proportion in terms of subsequent response reached the lower gut. Results with three methionine analogs indicated that they too were degraded in the rumen at a rate similar to that of methionine and that similar quantities reached the lower gut. These results suggest that the amino acid status of a ruminant may be improved by supplementing amino acids or amino acid analogs to its diet but it is evident that this is likely to be unprofitable.

An alternative procedure for supplementing the diet with amino acids is the encapsulation of protein or amino acid with a polymer insoluble in rumen contents but soluble in the acid conditions of the abomasum. Sibbald et al. (43) reported that a dietary product consisting of a core of methionine, kaolin and triglycerides, enveloped in a continuous film of triglycerides, reduced rumen degradation and increased methionine concentration in the blood plasma of steers. Steers fed encapsulated methionine gained more rapidly than did control steers. Broderick et al. (69), using lactating dairy cows, supplemented a 15% protein diet with encapsulated methionine. No effects on production due to methionine feeding were noted. Plasma methionine and methionine-valine ratios increased in all cows fed 15 g per day of methionine. Similar results for milk production of lactating cows were reported by Williams et al. (70). Although the results of encapsulation of amino acids are not very encouraging so far, many of the problems involved seem mainly technical and should not prove insurmountable. Future studies may demonstrate efficient methods of encapsulation and thus may provide an effective method for supplementing ruminant diets.

PROTECTION OF FATS BY PROTEIN MODIFICATION

Development of methods for protecting proteins from degradation in the rumen has lent impetus to development of a procedure for altering the degree of saturation of milk and body fat in the ruminant. That the rumen provides an excellent reducing medium, wherein unsaturated fatty acids of dietary origin are saturated, is well established (1-3). Moreover the usual diet of the ruminant is relatively low in fat. Thus much of the body and milk fat is derived from nonlipid components of the diet. A method for greatly increasing the polyunsaturated fatty acid content of milk fat has been proposed by Scott et al. (74). The technique involves the production of a dietary supplement consisting of small particles of polyunsaturated oil droplets encapsulated by a layer of protein. The particles are obtained by spray-drying a homogenate containing equal parts of linseed oil and sodium caseinate. After spray-drying, the particles are treated with formalin to protect the protein coating from microbial degradation in the rumen (56). In experiments with goats Scott et al. (74) fed a diet of 1.0 kg alfalfa hay, 1.0 kg oats and 0.5 kg untreated linseed oil-protein particles. The proportion of linolenic acid in the milk fat increased from 1% to 4%. The proportion of linolenic acid increased to 21-25% when formaldehyde treated linseed oil-protein particles were substituted for the untreated particles. Scott et al. (74) also studied the effect of these particles on milk fat composition in the cow. Control animals were fed a diet containing 5.5 kg chopped alfalfa hay and 3.5 kg oats. Experimental animals received the same diet plus 1.5 kg of the treated linseed oil-protein particles. The polyunsaturated fatty acid content of the milk fat of cows fed the treated particles was markedly increased. Treated safflower oil-protein particles caused a great increase in linoleic acid content of the milk, but no significant change in linolenic acid (safflower oil contains ca. 75% linoleic acid). Treated safflower oil-casein supplements fed to lambs caused significant increases in the proportion of linoleic acid in perinephric, mesenteric and subcutaneous fat. Similar effects were observed in cattle.

A further ramification of the use of treated oil-protein particles has been suggested by Black (44) in a discussion of the effect of preventing rumen fermentation on the efficiency of utilization of dietary energy and protein in lambs. He calculated that substitution of fat for carbohydrate would result in a considerable increase in the productive energy available to the ruminant lamb, but pointed out that the level of substitution would be limited to ca. 10% because of the depressing effect of higher levels of fat on digestibility of other dietary components. Feeding treated oil-protein particles (74) would provide a method for increasing the quantity of dietary fat well above 10% without affecting digestibility of other dietary components. Certainly the feeding of high levels of fat (20% of the diet) of specified fatty acid composition to veal calves has been common practice for several years and in these animals the fat is well utilized.

Desirability of feeding high levels of fat to the ruminant by this method and of increasing the relative proportion of unsaturated fatty acids in milk and meat is speculative. A full evaluation cannot be made until more information on the effect of these procedures is obtained regarding animal health and productivity. More information also is needed on the effects of human consumption of such altered products.

REFERENCES

1. Blackburn, T.H., in "Physiology of Digestion in the Ruminant," Edited by R.W. Dougherty, Butterworths, Washington, 1965, p. 322.
2. Hungate, R.E., "The Rumen and Its Microbes," Academic Press, New York and London, 1966.
3. Waldo, D.R., J. Dairy Sci. 51:265 (1968).
4. Smith, R.H., J. Dairy Res. 36:313 (1969).
5. Allison, M.J., in "Physiology of Digestion and Metabolism in the Ruminant," Edited by A.T. Phillipson, Oriel Press, Newcastle upon Tyne, England, 1970, p. 456.

6. Kay, R.N.B., Proc. Nutr. Soc. 28:140 (1969).
7. Virtanen, A.I., Science 153:1603 (1966).
8. Houpt, T.R., in "Physiology of Digestion and Metabolism in the Ruminant," Edited by A.T. Phillipson, Oriel Press, Newcastle upon Tyne, England, 1970, p. 119.
9. Gray, F.V., A.F. Pilgrim and R.A. Weller, Brit. J. Nutr. 12:413 (1958).
10. Clarke, E.M.W., G.M. Ellinger and A.T. Phillipson, Proc. Roy. Soc.(B) 166:63 (1966).
11. Hogan, J.P., and R.H. Weston, Aust. J. Agr. Res. 18:803 (1967).
12. Hogan, J.P., and R.H. Weston, Aust. J. Agr. Res. 18:973, (1967).
13. Pilgrim, A.F., F.V. Gray, R.A. Weller and C.B. Belling, Brit. J. Nutr. 24:489 (1970).
14. Walker, D.J., in "Physiology of Digestion in the Ruminant," Edited by R.W. Dougherty, Butterworths, Washington, 1965, p. 296.
15. Walker, D.J., and C.J. Nader, Aust. J. Agr. Res. 21:747 (1970).
16. Block, R.J., and H.H. Mitchell, Nutr. Abstr. and Rev. 16:249 (1946).
17. Black, A.L., M. Kleiber and A.H. Smith, J. Biol. Chem. 197:365 (1952).
18. Downes, A.M., Aust. J. Biol. Sci. 14:254 (1961).
19. Johnson, B.C., T.S. Hamilton, W.B. Robinson and J.C. Garey, J. Anim. Sci. 3:287 (1944).
20. Reed, R.M., R.J. Moir and E.J. Underwood, Aust. J. Sci. Res.(B) 2:304 (1949).
21. McNaught, M.L., J.A.B. Smith, K.J. Henry and S.K. Kon, Biochem. J. 46:32 (1950).
22. McNaught, M.L., E.C. Owen, K.M. Henry and S.K. Kon, Biochem. J. 56:151 (1954).
23. Bergen, W.G., D.B. Purser and J.H. Cline, J. Nutr. 92:257 (1967).
24. Tannenbaum, S.R., and S.A. Miller, Nature 214:1261 (1967).
25. Bigwood, E.J., in "The Role of the Gastrointestinal Tract in Protein Metabolism," Edited by H.N. Munroe, Blackwell, Oxford, England, 1964, p. 155.
26. Cuthbertson, D.P., and M.I. Chalmers, Biochem. J. 46xvii (1950).
27. Chalmers, M.I., D.P. Cuthbertson and R.L.M. Synge, J. Agr. Sci. 44:254 (1954).
28. Little, C.O., and G.E. Mitchell, Jr., J. Anim. Sci. 26:411 (1967).
29. Colebrook, W.F., and P.J. Reis, Aust. J. Biol. Sci. 22:1507 (1969).
30. Reis, P.J., and P.J. Schinckel, Aust. J. Biol. Sci. 16:218 (1963).
31. Reis, P.J., and P.J. Scinckel, Aust. J. Biol. Sci. 17:532 (1964).
32. Reis, P.J., Aust. J. Biol. Sci. 20:809 (1967).
33. Reis, P.J., Aust. J. Biol. Sci. 22:745 (1969).
34. Reis, P.J., Aust. J. Biol. Sci. 23:441 (1970).
35. Dreiden, G.M., G.A. Wickham and F. Cockrum, N.Z.J. Agr. Res. 12:580 (1969).
36. Downes, A.M., P.J. Reis, L.F. Sharry and D.A. Tunks, Aust. J. Biol. Sci. 23:1077 (1970).
37. Schelling, G.T., and E.E. Hatfield, J. Anim. Sci. 26:929 (Abstract) (1967).
38. Schelling, G.T., and E.E. Hatfield, J. Anim. Sci. 26:1484 (1967).
39. Schelling, G.T., and E.E. Hatfield, J. Nutr. 96:319 (1968).
40. Nimrick, K., E.E. Hatfield, J. Kaminski and F.N. Owens, J. Nutr. 100:1293 (1970).
41. Nimrick, K., E.E. Hatfield, J. Kaminski and F.N. Owens, J. Nutr. 100:1301 (1970).
42. Devlin, T.J., and W. Woods, J. Anim. Sci. 23:872 (1964). Abstract.
43. Sibbald, I.R., T.C. Loughheed and J.H. Linton, Presented at the 2nd World Conf. Anim. Prod., College Park, Maryland, 1968.
44. Black, J.L., Brit. J. Nutr. 25:31 (1971).
45. Ely, D.G., C.O. Little, P.G. Woolfolk and G.E. Mitchell, Jr., J. Nutr. 91:314 (1967).
46. Hemsley, J.A., Aust. J. Exp. Biol. Med. Sci. 45:39 (1967).
47. McGilliard, A.D., PhD Thesis, Michigan State University, 1961.
48. Orskov, E.R., and D. Benzie, Proc. Nutr. Soc. 28:30A (1969).
49. Orskov, E.R., and C. Fraser, J. Agr. Sci. 73:469 (1969).
50. Orskov, E.R., D. Benzie and R.N.B. Kay, Brit. J. Nutr. 24:785 (1970).
51. Orskov, E.R., C. Fraser and E.L. Corse, Brit. J. Nutr. 24:803 (1970).
52. Black, J.L., Aust. J. Sci. 32:332 (1970).
53. Chalmers, M.I., J.B. Jayasinge and S.B.M. Marshall, J. Agri. Sci. 63:283 (1964).
54. Whitelaw, F.G., T.R. Preston and G.S. Dawson, Anim. Prod. 3:127 (1961).
55. Tagari, H., I. Ascarelli and A. Bondi, Brit. J. Nutr. 16:237 (1962).
56. Ferguson, K.A., J.A. Hemsley and P.J. Reis, Aust. J. Sci. 30:215 (1967).
57. Reis, P.J., and D.A. Tunks, Aust. J. Agr. Res. 20:775 (1969).
58. Reis, P.J., and D.A. Tunks, Aust. J. Biol. Sci. 23:673 (1970).
59. Hughes, J.G., and G.L. Williams, Anim. Prod. 12:366 (Abstract) (1970).
60. Rattray, P.V., and J.P. Joyce, N.Z.J. Agr. Res. 13:623 (1970).
61. Peter, A.P., PhD Thesis, University of Illinois, 1971.
62. Zelter, S.Z., F. LeRoy and J.P. Tissier, Ann. Biol. Anim. Biochem. Biophys. 10:111 (1970).
63. Clark, J.H., C.L. Daivs and E.E. Hatfield, J. Dairy Sci. 54: (Abstract), in press.
64. Downes, A.M., P.J. Reis, L.F. Sharry and D.A. Tunks, Brit. J. Nutr. 24:1083 (1970).
65. Zelter, S.Z., and F. LeRoy, Z. Tierphysiol. Tierernahr. Futtermittelk. 22:39 (1966).
66. Driedger, A., PhD Thesis, University of Illinois, 1970.
67. Delort-Laval, J., and S.Z. Zelter, Presented at the 2nd World Conf. Anim. Prod., College Park, Maryland, 1968.
68. Lewis, A.J., PhD Thesis, University of Nottingham, England, 1971.
69. Broderick, G.A., T. Kowalczyk and L.D. Satter, J. Dairy Sci. 53:1714 (1970).
70. Williams, L.R., F.A. Martz and E.S. Hilderbrand, J. Dairy Sci. 53:1709 (1970).
71. Reiser, R., Fed. Proc. 10:236 (1951).
72. Shorland, F.B., R.O. Weenink, A.T. Johns and I.R.C. McDonald, Biochem. J. 67:328 (1957).
73. Ward, P.F.V., T.W. Scott and R.M.C. Dawson, Biochem. J. 92:60 (1964).
74. Scott, T.W., L.J. Cook, K.A. Ferguson, I.W. McDonald, R.A. Buchanan and G. Loftus Hills, Aust. J. Sci. 32:291 (1970).

[Received June 30, 1971]