PRINCIPLES FOR THE USE OF NON-PROTEIN-NITROGEN AND 'BYPASS' PROTEINS IN DIETS OF RUMINANTS

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Bypass proteins are *defined here as those dietary proteins that pass intact from the rumen to the duodenum.*

Digestible bypass protein is then that portion of the bypass protein which is hydrolyzed in and absorbed from the small intestine.

Overprotected proteins are neither fermented in the rumen nor digested in the small intestine.

INTRODUCTION

The apparent inefficiency of ruminants compared with monogastric animals in utilising protein rich feeds has been used as an argument to emphasise the importance of monogastric animals in preference to ruminants for meat production. Recent studies however (see Preston & Willis, 1970 ; Ørskov, Fraser, McDonald & Smart, 1974 ; Kempton & Leng, J-976), have indicated that with correct balancing of digestible nutrients, ruminants given feed of apparently variable quality can grow at rates much greater than those generally reported (Ørskov, 1976). These results have been achieved with approximately 50 - 70% of the usual recommended requirements for protein in a diet. Ruminants are therefore potentially highly efficient users of protein feeds under a variety of agricultural situations, including the utilization of low protein by-products of agro-industries.

Efficient utilisation of protein and non-protein nitrogen (NPN) by ruminants in any production system depends on a knowledge of the underlying basic principles, and these are reviewed here. Emphasis in this review, however, is given to the requirements for dietary proteins that escape from the rumen unchanged and are available for digestion. These are termed 'bypass proteins* to differentiate them from proteins fermented in the rumen, and from total available digestible protein (which is digestible dietary bypass protein plus digestible microbial protein) termed "metabolisable protein" by Burroughs, Trenkle and Vetter (1971).

PROTEIN DIGESTION IN RUMINANTS

General considerations

The rumen evolved as a means of digesting the constituents

of plants for which animals did not have the necessary endogenous digestive enzymes. The fermentation of protein in the rumen is a product of this evolution, which under certain circumstances is detrimental but in the absence of other forms of N ensures a supply of N for the microorganisms.

In different production systems, ruminants feed on many types of carbohydrates, proteins and other plant and animal constituents. Most digestible carbohydrates are fermented by essentially the same pathways (see Leng, 1973), to volatile fatty acids (VFA) plus methane and carbon dioxide. Proteins are fermented to the same end-products and, in addition, to ammonia. However, peptides and amino acids are intermediates and may be used in microbial cell synthesis. Ammonia is either absorbed directly across the rumen wall or passes out of the rumen with the fluid phase of digesta or is incorporated into microbial protein. The dietary protein is, however, not totally degraded and some passes intact into the abomasum and duodenum, where it is digested by enzymic hydrolysis.

Microbial, dietary and endogenous protein leaving the rumen is subjected to digestion and absorption in the small intestine. Any protein leaving the small intestine may be fermented by microorganisms in the caecum and colon or excreted in the faeces, but it is generally believed that the microbial protein produced in this organ is not available as amino acids to the animal.

The factors that influence the supply of amino acids to the tissues of ruminants are therefore complex and not fully understood (see Table 1 for some of the major factors).

Ammonia utilisation in the rumen

Peptides, amino acids and ammonia form the nitrogenous starting material for the synthesis of microbial cells. Ammonia is extensively used by many species of rumen microorganisms as a source of N for synthesis of their nitrogenous constituents and this is exemplified by studies in which ammonium salts apparently provided the sole dietary N source for sheep and cows (Loosli, Williams, Thomas, Ferris and Maynard, 1949; Virtanen, 1966). However, these findings can be misleading if two points are not recognised. Firstly, some species of organism commonly found in the rumen require preformed peptides or amino acids (Wright & Hungate, 1967). If these are not provided in the diet, and are in low concentration in rumen fluid, some microorganisms may disappear from the rumen, changing the balance of species. The total quantity of protein synthesised, or the efficiency of microbial synthesis (g protein/kg of organic matter fermented (FOM)), may thus be altered. There may be a reduction in protein yield if ammonia concentration is low, i.e. less than 80 mg N/R, (see Satter and Slyter, 1972), although Ørskov (1976) has concluded that, in sheep fed grain diets, the rate of fermentation and therefore the rate of protein production is reduced if rumen ammonia concentrations are below 200 mg N/R. Levels below this, however, will only reduce microbial protein availability when residence time of feed materials in the rumen is short. The practical implication of these results is that whenever ammonia concentration falls below about 80 mg N/R (although when fermentation rate is rapid the critical range may be higher), the rumen microorganisms may be ammonia deficient, and might be considered likely

Table 1. Factors influencing amino acid availability from the digestive tract

Rumen	Microbial growth	Availability of fermentable substrate
		Efficiency of bacterial growth
		Species composition of microbial community
		Death or destruction of micro-organisms and subsequent fermentation in the rumen
		Rumen turnover rate
	Dietary protein escaping fermentation	Balance of microorganisms
		Solubility of protein
		Particle size and physical form of feed
		Voluntary feed intake
		Rumen turnover rate
		pH
Small Intestine		Digestibility of microorganisms
		Digestibility of dietary protein
		Digestibility of endogenous secretions
		Rate of flow of digesta (influencing efficiency of digestion)
		Presence of parasites and microorganisms in the small intestine.

to respond to dietary non-protein-nitrogen (NPN) supplements. In the grazing ruminant, this situation occurs less frequently than might be expected because sheep, and to a lesser extent cattle, show a marked ability to select the material of high N content from poor quality pastures (see Loosli & McDonald, 1968).

The second point is that even when nutrients are non-limiting in the rumen, the rumen system may not supply sufficient microbial protein to meet the needs for maximum production. Under these conditions, high production depends on an additional exogenous amino acid supply to the duodenum (as for example by feeding proteins that, because of their physical state, escape rumen fermentation and are digested in the duodenum). Although lactating cows could be maintained on protein free diets (Virtanen, 1966), for maximum milk production 20% of the dietary nitrogen had to be supplied as protein (Virtanen, 1967).

PROTEIN REQUIREMENTS OF RUMINANTS

In the past, the protein requirements of ruminants and

evaluation of the protein value of foods for ruminants have been based on digestible crude protein (N x 6.25), although this has been discredited to some extent recently (see Miller, 1973). The use of the concept of digestible crude protein has arisen largely because it was considered that the animal could obtain its essential amino acids from microbial protein produced in the rumen from ammonia, and this removed the necessity for a specific requirement for dietary protein. This in turn led to suggestions that extensive use could be made of non-protein nitrogen materials (such as urea) by ruminants producing meat and milk from low protein - high carbohydrate feeds. These concepts however must be modified in the light of recent research findings which indicate that when amino acid requirements of ruminants are high, insufficient protein is available from microbes. This indicates that amino acid requirements should be expressed in terms of amino acids absorbed by the animal (i.e. digestible bypass protein plus digestible microbial protein).

The protein or amino acid requirements are, however, influenced by a number of factors, i.e.

a) the physiological state of the animal, that is the potential rates of growth and milk production, wool growth rate and stage of pregnancy (see Ørskov, 1970);

b) the rate of growth and production as influenced by metabolizable energy intake (see Preston, 1976);

c) the body composition as influenced by previous nutritional history (Andrews & Ørskov, 1970 a & h);

d) the proportions of different amino acids absorbed (see later);

e) the efficiency of microbial protein production and its net availability (see Thomas, 1973);

f) patterns of ruminal fermentation as these affect production and availability of volatile fatty acids that are glucogenic (propionic, valeric and isobutyric acids) (see Leng, 1976);

g) the requirements for glucose (Leng, 1976).

The protein requirements of ruminants are not constant but vary in relation to changing productive or physiological state (Fig. 1). The dotted line indicates the extent of incorporation of microbial protein into tissue protein. Provided metabolizable energy is non-limiting then the rumen microorganisms appear to be able to provide sufficient protein for maintenance, slow growth' and early pregnancy but not for fast growth, late pregnancy or early lactation.

For the above reasons protein requirements of ruminants cannot simply be stated as digestible crude protein (N x 6.25) in a given diet. It is therefore necessary to assess requirements for N in terms of the amount of NPN and amino acid-N needed by the rumen microbes and the amount of extra digestible protein needed by the animal. However, the many factors that affect such requirements must be understood in order to apply such requirement data.

Protection of proteins from ruminal degradation

Chalmers and Synge (1954) and Annison (1956) established that protein solubility is the major factor that governs the rate of break-

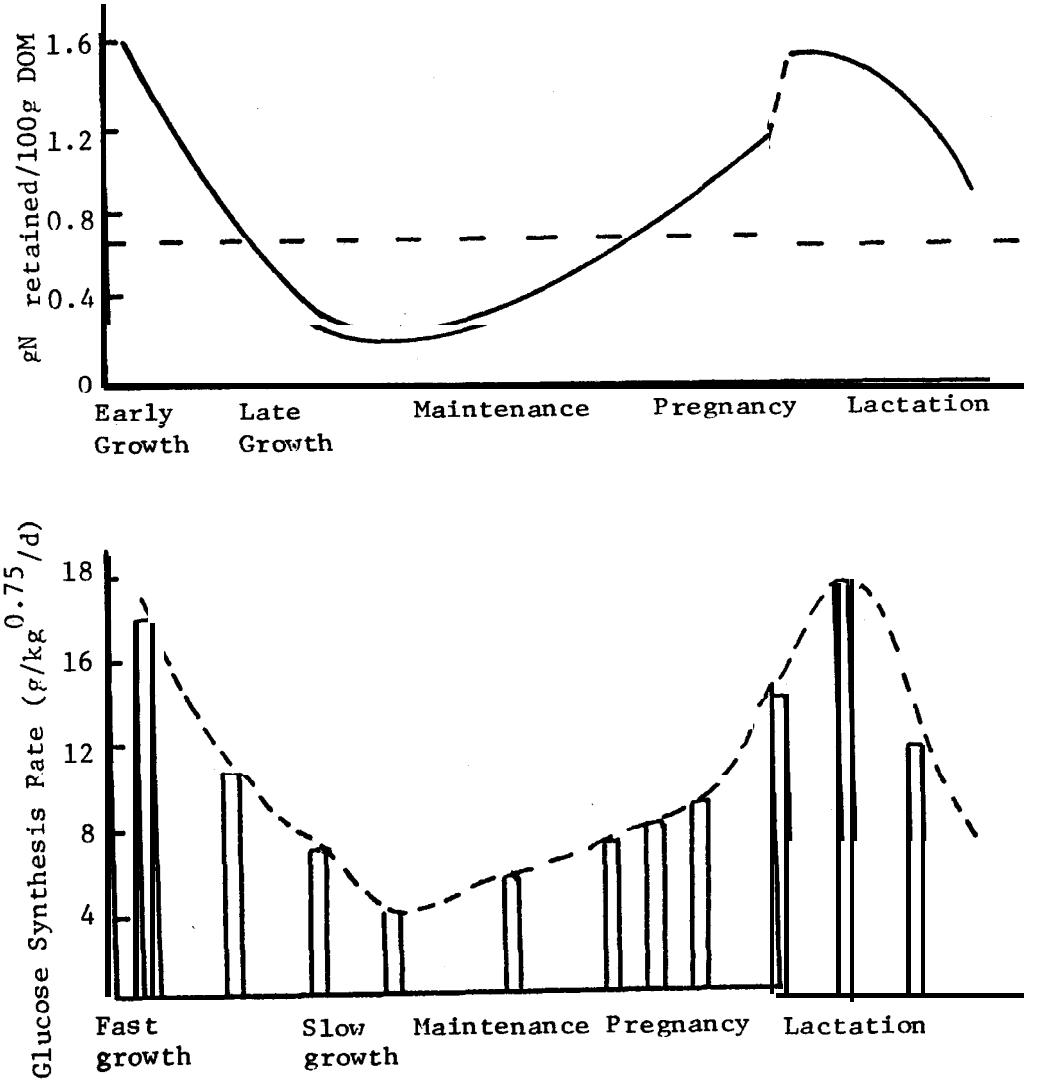

Figure 1. *Amino acid and* glucose requirements in *ruminants in relation to physiological state (from Ørskov,* 1970 *and Leng, 1976).*

down of dietary protein in the rumen. Rate of rumen fluid turnover and other factors are also involved (Table 1). If flow rate from the rumen is rapid some highly soluble dietary proteins may leave the rumen intact. Conversely, relatively insoluble proteins will be degraded if they are retained for long periods in the rumen, and therefore, as discussed by Sutherland (1976), flow rate from the rumen has considerable influence on the quantity of bypass protein (as defined here) in a diet.

Since some protozoa can ingest solid feed particles, these may assist in breaking down relatively insoluble, particulate protein and the extent to which this occurs depends on the total biomass of protozoa in the rumen (see Leng, 1976). There also must be large differences between cattle and sheep since, in general, sheep grind their feed more fully in chewing and therefore make a greater surface area of protein available for colonisation by microorganisms.

The oesophageal groove reflex also enables dietary proteins to become directly available to the animal. This has been used by Ørskov and Benzie (1969) and Lawlor, Kealy & Hopkins (1971) to supplement growing lambs with proteins.

Naturally occurring bypass proteins

Bypass proteins have been defined as being dietary proteins which escape ruminal fermentation and arrive at the site of enzymic digestion. Bypass proteins occur naturally in feedstuffs or can be produced by various chemical or physical manipulations (see Table 2).

Table 2. The solubility (stated as % fermented) of a number of protein meals. The values may vary considerably between samples depending on a large number of variables and should be taken only as a guide.

Meal	% Fermented	Meal	% Fermented
Meat	30	Cottonseed (heated)	20
Fish	20-80	Dehydrated lucerne	40
Soyabean	20-70	Lucerne hay	60
Lupin	65	Fresh clover	75
Casein	100	Dry clover	45
Formaldehyde-casein	0-10	Rye Grass	65-100
Peanut	65	Silage	70-80
Formaldehyde-peanut	20	Wilted silage	50-70
Heated peanut	20	Grains	30-50
Linseed (heated)	20		

The solubility of proteins in most herbage species varies considerably with both stage of vegetative growth and environmental conditions. Hume and Purser (1974) have found that ruminal degradation of clover proteins in sheep declined from 74% in green material to

45% in mature material. In freshly cut grass fed to sheep there was little bypass protein present (see MacRae, 1976). Up to 60% of pasture protein goes into solution in chewing (see Reid, Lyttelton and Mangan, 1962; Bryant, 1964; Hogan, 1965) indicating its highly soluble nature.

Protection of dietary proteins during processing

Many of the processes of preserving herbage such as sun-drying, force-air drying or freezing significantly decrease the solubility of the protein. Ensiling, (unless preceded by wilting), generally results in a decrease in bypass protein content of the final material (Goering and Waldo, 1974).

Heat treatment protects dietary proteins for ruminants but it is important that apparopriate temperatures and heating times are employed for particular feeds. The optimal conditions however, are often not known. The effects of temperature on soluble N content, N digestibility and nitrogen retention in lambs fed dried lucerne are shown in Table 3 (Goering and Waldo, 1974). Heating above 160° depressed

Table 3. The effects of drying temperature on the solubility and digestibility of nitrogen in lucerne fed to lambs.

Temperature of drying (°C)	Soluble N (%)	N digestibility (%)	N retention (g/d)
65	43	69	6.0
130	40	68	7.4
160	40	66	6.9
180	34	52	3.4

(from Goering and Waldo, 1974).

N retention in lambs indicating overprotection of the dietary protein (see later). The extent to which overprotection occurred however, may have been influenced by the composition of the lucerne plants at the time of harvest. The content of sugars (see later) influences the extent of heat 'damage' brought about by the so-called Browning reaction. For instance, heating of meat meals with molasses has resulted in considerable reduction in biological value of the protein as indicated by chicken growth assay (Edwards, 1976) due to the Browning reaction (Miller, 1976).

Techniques including grinding, rolling, cracking, micronisation and wafering are often used in feed compounding and these must afford some protection to dietary proteins through changes in both physical and chemical characteristics and subsequent changes in digesta flow patterns (Thomson, 1972). Pelleting of diets also appears to protect the proteins owing to the heat generated in the dye. Heat treatment during solvent or pressure extraction of oil-seeds results in a variable degree of protection of the proteins in the resulting meals.

Chemical protection of proteins

Proteins may also be protected chemically using substances such as tannins, formaldehyde, glutaraldehyde, glyoxal and hexa-methylenetetramine (e.g. formaldehyde treated casein Ferguson, Helmsley and Reis, 1967; Schmidt, Jorfensen, Bemevenga and Breinghardt, 1973). Because of the availability of low cost naturally-occurring bypass proteins, chemical treatment of dietary proteins is probably uneconomical. Chemical treatment, however, may find application in some developing countries where oil-seed meals are often prepared without heat and fish meals are prepared from sun-dried fish, since the proteins of these meals are highly soluble. However, heat treatment will in general also protect these protein meals.

In the past, because of the lack of recognition of the occurrence of naturally bypass proteins, many attempts have been made to use chemical treatments to protect proteins that were already protected (see later).

Overprotection

Various treatments can cause overprotection of proteins in meals, i.e. the proteins are rendered wholly or partially indigestible in the small intestine. For instance Kempton, Nolan and Leng (1976) found that 100% of formaldehyde treated casein escaped from the rumen of lambs and of this only 70% was digested in the small intestine.

As has already been mentioned, heating or pelleting of meals high in sugar may result in considerable loss of protein quality because of the Browning reaction (see Miller, 1976).

RESPONSES TO BYPASS PROTEINS BY RUMINANTS

The first reported responses to additional amino acids given in the duodenum of sheep were those by Egan & Moir (1965). Voluntary intake of a low protein roughage by sheep was stimulated by infusion of amino acids into the duodenum (Egan, 1965). Responses in wool growth have also been obtained with intraduodenal infusion of protein and by feeding bypass proteins (see review, Ferguson, 1975).

Under practical conditions Preston and his colleagues (see Preston & Willis, 1970) were the first to demonstrate that feed intake and growth could be stimulated by inclusion of bypass proteins in a low protein diet (see Fig. 2). Relatively insoluble proteins, such as fish meal, added to a low protein diet, stimulated the intake and growth of cattle much more than soluble proteins such as rape seed meal. Similar results were obtained with grain based diets by Ørskov and his colleagues (see Table 4). Growth rates in lambs on a diet of pelleted barley plus 1% urea and minerals were stimulated by supplementing with fish meal. Faichney & Davies (1973) compared diets with soluble and formaldehyde-treated peanut meal (insoluble) and obtained increased growth where proteins were treated.

Studies with low protein-cellulose diets in these laboratories also show that feed intake is often restricted by dietary protein availability. Young lambs on diets of 70% oat-hulls, 30% Solka-Floe (a pure wood cellulose) plus minerals, were used. Additions of 2-4% urea

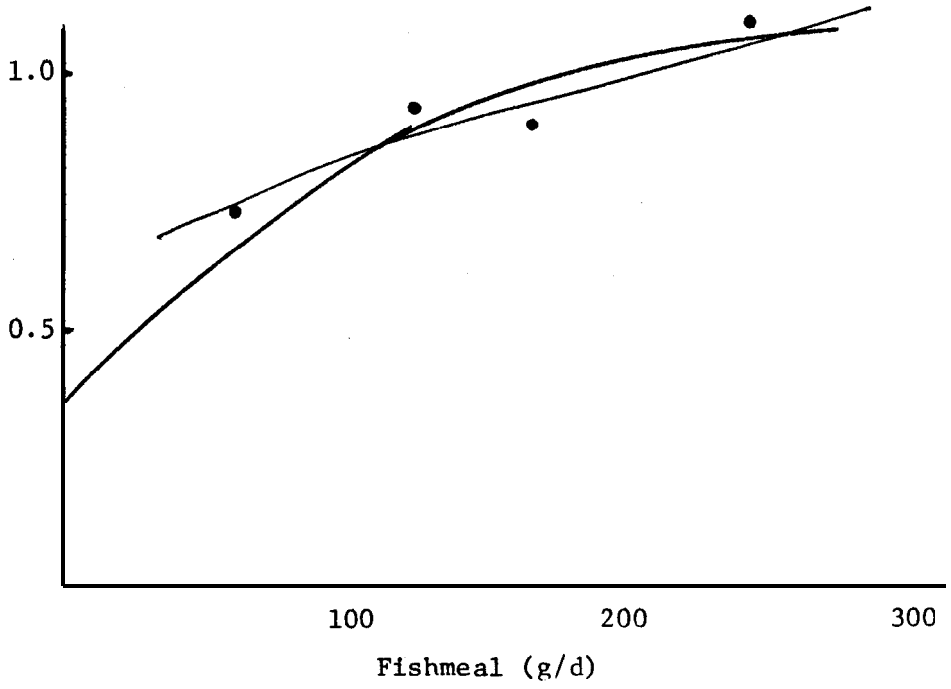

Figure 2. *Effect of fishmeal supplementation on liveweight pin in cattle (Preston & Willis, 1970).*

Table 4. Growth rate and feed conversion efficiency (FCE) of lambs on a pelleted diet of barley + 1% urea and minerals supplemented with fish meal (from Ørskov, Fraser and Pirie, 1973).

Fish meal supplement (g/d)	Growth rate (g/d)	F.C.E.* (kg/kg)
0	230	4.3
0 + 10 g urea	224	4.3
17	300	3.5
34	326	3.2
51	332	3.0

* kg of feed required for 1 kg increase in body weight

(sufficient to supply adequate N for microbial fermentation) and various combinations of casein, which McDonald and Hall (1957) found was completely hydrolysed in the rumen, and formaldehyde-treated casein (bypass casein) were made. The results are shown in Fig. 3. There was a much greater response in total feed intake and growth rate from

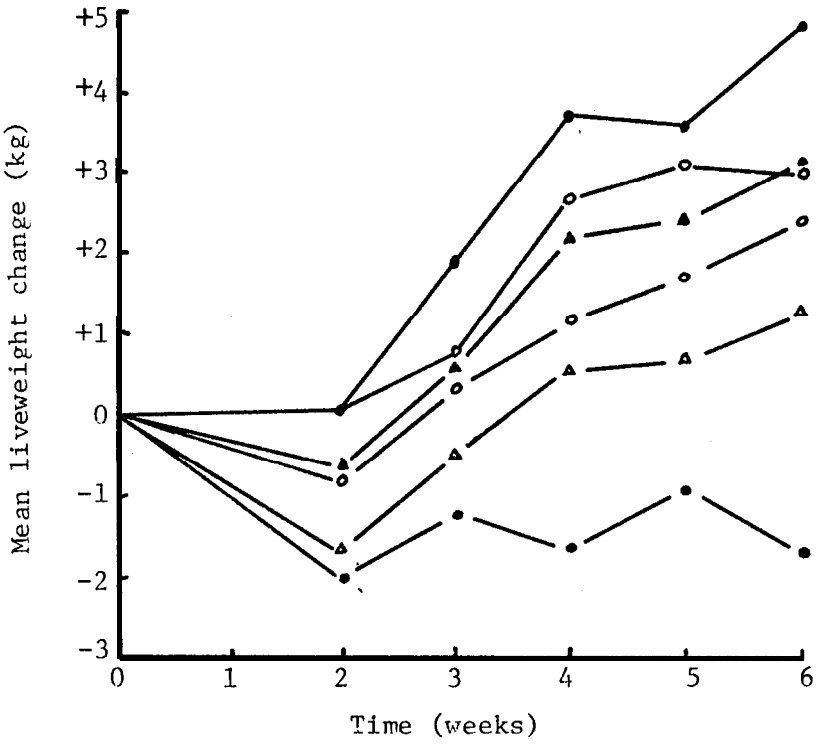

Figure 3. *Growth of lambs on a roughage based diet (30% Solka Floc, 70% oat-hulls) supplemented with:*

1. *control;*
2. *2.5% urea;*
3. *7.5% formaldehyde treated casein;*
4. *7.5% soluble casein;*
5. *2.5% urea + 7.5% formaldehyde treated casein;*
6. *2.5% urea + 7.5% soluble casein.*

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protected proteins in conjunction with urea, as compared with soluble proteins or urea alone. In other experiments lambs were given the same basal diet plus 2% urea with graded quantities of casein and protected casein. As the bypass protein content of the diet was increased, the intake of feed increased but was at a maximum at 10% bypass casein in the diet (Fig. 4). It was subsequently shown that about one third of the protein in the bypass casein was undigested suggesting that the actual requirement for protein was only 7% of this diet.

The diets used above had a low degree of lignification and hence rumen fill may not have been a primary limitation to feed intake (Balch and Campling, 1962). The first experiment was therefore repeated using oaten chaff as the basal diet. Similar increases in feed intake and growth were obtained when lambs were given bypass proteins and urea (Kempton & Leng, 1976), suggesting again that protein status and not rumen fill was the first limitation to intake.

Responses to protected protein on green pasture

There is some evidence that the proteins in young fast-growing pastures may be so soluble that little dietary protein passes out of the rumen (see MacRae & Ulyatt, 1974); at times therefore productive ruminants at pasture may be protein deficient (see Leng, 1975; MacRae, 1976) resulting in low feed intake and production (see Leng, 1976).

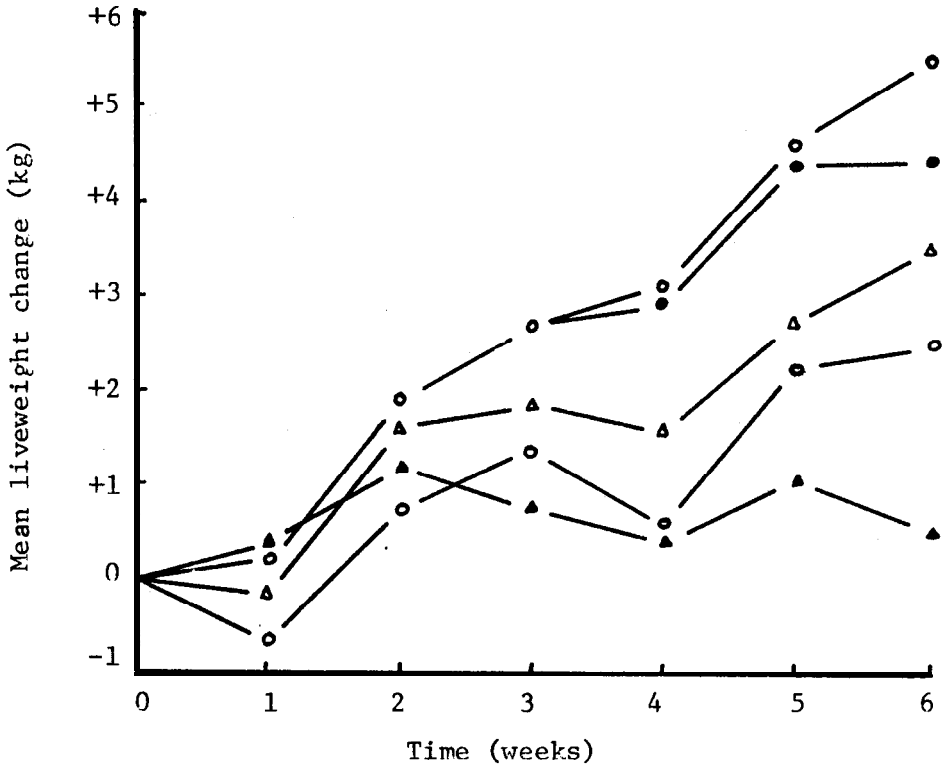

Figure 4. *Growth of lambs on a roughage based diet (30% Solka Floc, 70% oat-hulls) supplemented with:*

A. control;
B. 15% soluble casein;
C. 10% soluble casein + 5% formaldehyde treated casein;
D. 5% soluble casein + 10% formaldehyde treated casein;
E. 15% formaldehyde treated casein.

Preliminary studies in these laboratories have indicated that lamb growth may be stimulated at pasture by drenching the animals with a slurry containing fish meal (Archer, Bar-wick, Kempton & Leng, 1976).

Bypass protein in the diet and feed intake

The'effect of bypass proteins in all diets used in these laboratories is mediated largely through stimulation of feed intake (see Fig. 5) as indicated by the linear relationship between feed intake and growth rate on all diets in both studies (see also Preston, 1976).

RUMEN AND METABOLIC FACTORS INFLUENCING THE REQUIREMENTS FOR BYPASS PROTEINS

Efficiency of microbial protein synthesis

The efficiency of microbial protein synthesis, expressed as the quantity of microbial amino acids available for absorption in the small intestine per unit of organic matter fermented in the rumen (FOM) must influence markedly the requirements for dietary amino acids.

Many factors influence this efficiency (see Table 1) including feed intake, feeding patterns, age of animal and species used, (or

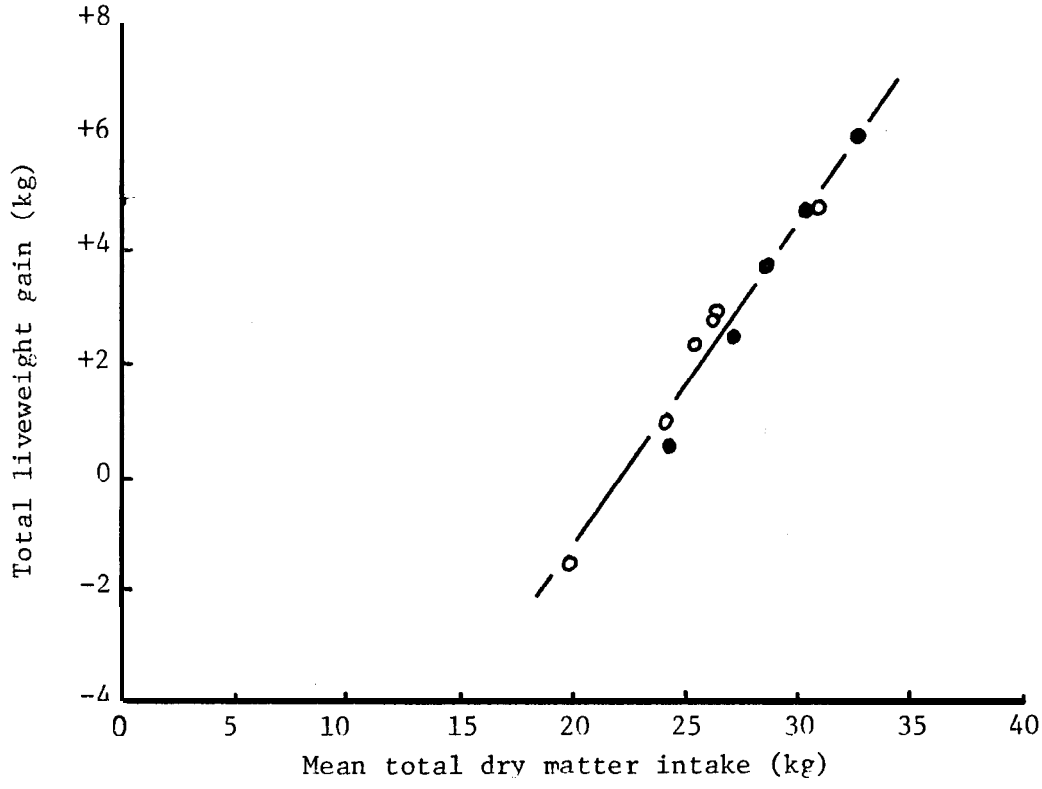

Figure 5. *Relation between total dry matter intake and liveweight gain of lambs given the diets summarised in Figs. 5 and 6.*

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experimental technique). For each kg of FOM, between 15 - 53 g N as microbial protein have been estimated to leave the rumen of sheep (see Thomas, 1973). It is difficult to relate much of this work to the practical feeding situation since much of this data was obtained with animals consuming 85 - *95%* of *ad libitum* feed intake. On some diets restriction of feed intake markedly changes the species composition of the microbial communities. This occurs for example, on grain diets where a restriction of feed intake results in the appearance of a large protozoa1 population (Eadie & Mann, 1970). It seems that even with *ad libitum* feeding regimes, the availability of microbial protein per kg of FOM is variable and it is clear that this is a factor that must be considered when formulating diets.

Turnover of microorganisms in the rumen. The amount of microbial protein available for intestinal digestion depends upon the efficiency of microbial growth which is affected by the rate of degradation of microbial cells in the rumen. The longer a microorganism remains in the rumen, the more likely it is to become damaged and digested in the rumen with a consequent decrease in the outflow of microorganisms. Damage and degradation of microorganisms result from predation by protozoa which actively ingest bacteria (Coleman, 1964) and infection by bacteriophages and mycoplasmas (Hoogenraad, Hird, Holmes and Millis, 1967). Marked changes in environmental conditions in the rumen may precede the death of protozoa (see Leng, 1976) and bacteria (Raigent, pers. corn.). Dead microorganisms are substrate for other microorganisms (see Hoogenraad, Hird, White & Leng, 1970) and are fermented to VFA, ammonia and methane. An internal cycle in the rumen has been demonstrated (viz. NH_3-N → microbial N → NH_3-N) suggesting

that at least 30% of the microbial biomass is continually degraded in the rumen (Abe & Kandatsu, 1969; Nolan & Leng, 1973).

Retention of protozoa in the rumen. Protozoa appear not to leave the rumen in any quantity relative to their concentration in the rumen fluid (Weller & Pilgrim, 1974; Leng & Preston, 1976: Baigent, Bird, Dixon & Leng, 1976). If these organisms do not leave the rumen, they are most certainly turned over in the rumen since their numbers vary from day to day (see Clarke, 1965; Leng & Preston, 1976); this turnover in the rumen will reduce the availability of microbial protein to the animal.

Digestibility of rumen microorganism

The digestibility of rumen microorganisms has often been considered to be constant. However, recent results have suggested that the digestibility of rumen microbes in the small intestine may vary from 30 - 70% (see Smith, 1975). This variability will have a marked effect on the requirements of animals for dietary bypass proteins for optimal production.

Availability of branched chain and higher fatty acids

There are indications that the branched chain and higher VFA are essential growth factors for some ruminal microorganisms (Bryant & Doetsch, 1955), and in animals given low protein diets, feed intake and fermentation rates have been stimulated by dietary supplementation with these materials (Hemsley & Moir, 1963; Hume, 1970). Valerie and isobutyric acids are also glucogenic and some of the increased feed intake could be attributed to their amino acid sparing effect (see later).

Fermentation pattern

The efficiency of microbial growth in the rumen may change with the pattern of fermentation as indicated by the molar proportions of VFA. Microbial yields have been reported to be highest on diets in which propionate proportions are high (Jackson, Rook & Towers, 1971) but there is some controversy on this point (Thompson, Beever, Mundell, Elderfield & Harrison, 1975; Latham & Sharpe, 1975). The presence of entodiniomorph protozoa in the rumen has been associated with a high butyrate, low propionate type of fermentation (Schwartz & Gilchrist, 1975).

Where protozoa occur there are possibly two constraints to animal production: (1) a reduced quantity of available microbial protein and (2) an increased requirement for gluconeogenesis since less propionate is absorbed (see later). The overall effect may be an increased requirement for dietary protein. This will only become a limiting factor where the availability of dietary protein is low and the animal's requirements are high.

GLUCOSE REQUIREMENTS AND METABOLISM OF RUMINANTS

Interaction between requirements for glucose and amino acids. Responses to bypass proteins may not be due entirely to an increased supply of essential amino acids to the animal. Considerable evidence

from these laboratories indicates that at least part of the response may be attributed to the supply of glucogenic amino acids which can' assist in meeting glucose 'req.uirements' (Kempton & Leng, 1976; see Leng, 1976). This is an extremely important point since it means that responses to high quality or low quality proteins, as defined in terms of amino acid composition, may be similar and also that responses may be obtained to other glucogenic materials, such as propionate, and carbohydrates that escape ruminal fermentation.

Recent reviews of glucose metabolism are available (Leng, 1970; Lindsay, 1970) and this topic will be discussed here only briefly. It is not possible to determine directly the requirements for glucose in ruminants. It is assumed here that requirements and synthesis rates are closely correlated, since any unneeded extra synthesis would be energetically very wasteful since gluconeogenesis is expensive in terms of requirements for energy. Synthesis of glucose in ruminants is related to digestible energy intake (Judson and Leng, 1968; Lindsay, 1970), stage of growth (T.J. Kempton, 1975, unpublished observations), stage of pregnancy (Steel and Leng, 1973) and lactation (Annison and Linzell, 1964; Bergman and Hogue, 1967) (for review, see Leng, 1970) (see Fig. 1).

In general glucose is apparently not absorbed in significant quantities except in animals given some grain diets (e.g. maize) (Armstrong, 1972). Propionic acid and amino acids are the major precursors of glucose in ruminants, however, a number of substrates (e.g. branched and higher fatty acids, etc.) may also contribute to a small but significant extent (Leng, 1970).

Glucose requirements for production

When amino acid requirements are high, glucose synthesis rates are high (see Fig. 1). The pattern of requirements for glucose follows closely that for amino acids suggesting that part of the apparently high requirement for amino acids may be for glucose precursors (100 g of amino acids from a typical protein can give rise to 57 g glucose, Krebs (1964). Therefore, contrary to previously held views (see Leng, 1970) it is possible that under conditions when productivity is potentially high, ruminants find difficulty in synthesising sufficient glucose, particularly on relatively low protein diets. During growth and lactation there may be competing needs for amino acids for glucose synthesis and for protein deposition (see Leng & Preston, 1976). The important point to be stressed here is that in growing, pregnant or lactating ruminants there is a high demand for amino acids for protein deposition, and for both amino acids and propionate for glucose synthesis. The central importance of glucose is indicated by the fact that 20 - 30% of digestible energy available to sheep may pass through the glucose pool (Judson and Leng, 1973).

In a further study in these laboratories, lambs in which the oesophageal groove had been maintained by suckling (see Ørskov & Benzie, 1969) and fed on a basal diet of oaten chaff, sugar and fishmeal, were supplemented with glucose by bottle feeding. The interraction between amino acid and glucose.supply on growth rate and the efficiency of growth is shown in Fig. 6.

In the highly productive ruminant in which the requirement

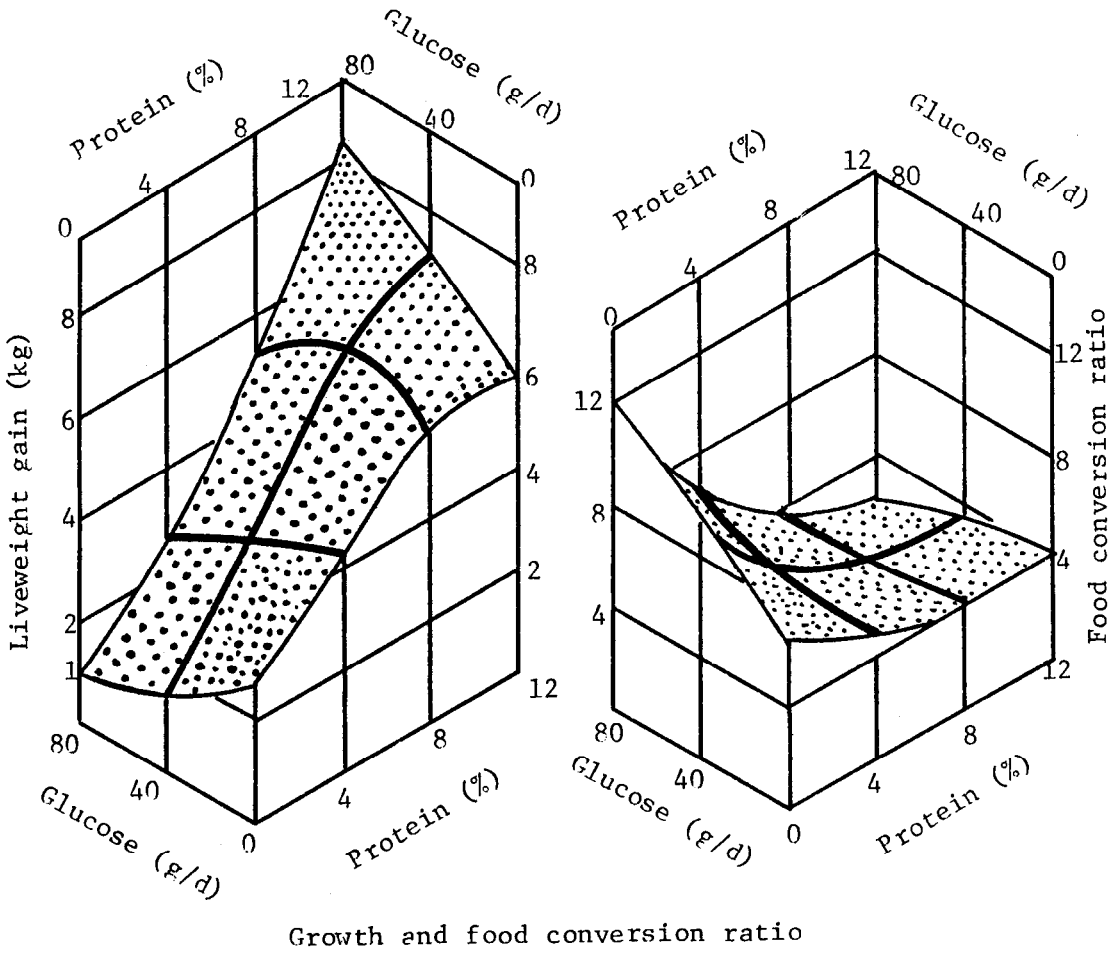

Figure 6. *Interraction between amino acid and glucose supply on growth.*

for bypass amino acids has been met, an additional response in production can be gained by increasing the supply of glucose to the animal, suggesting ruminants have a specific requirement for energy (as glucose). If the requirement for amino acids is not met however, glucose supplementation has a negative effect on production.

Amino acid composition of bypass proteins

The likelihood that part of the responses obtained with supplements of dietary proteins may be attributable to the supply of glucogenic materials implies that the essential amino acid composition of the bypass proteins may not be as critical as previously believed (Leng, 1975). For instance, equal growth rates of lambs on low protein diets supplemented with cotton seed meal or fish meal have been obtained (Djajanegara, Kempton & Leng, 1976).

EXPLANATION OF REPORTED LACK OF RESPONSE TO SUPPLEMENTARY BYPASS PROTEIN

There are many studies in the literature which record a lack of response to protection of proteins in a diet for ruminants. Reasons for the lack of response may be found in the type of diet and its preparation, in the levels of feeding, or the productive state of the

animals. In many instances much of the protein is naturally protected, or the level of bypass protein in the so called 'control' diet is already adequate. Many studies have reported the effect of formaldehyde treatment of meals where the proteins were already largely protected. Fish meal proteins for instance are usually protected, yet numerous workers have examined the effects of formaldehyde treatment of these meals when no large effects could be expected. Such treatments may actually decrease protein availability through over-protection. Moreover, where the "protected" and "unprotected" diets are pelleted, the "unprotected" control diets may also become protected by heat, and treatment responses therefore not observed. It is therefore important in the study of the use of bypass proteins that the amount of digestible dietary protein available in the small intestine is measured with and without "protection".

Responses to bypass proteins should be expected only when the requirements for amino acids are not being met. It follows that the lack of responses to protection of dietary protein reported by some workers may have been that the experimental animals were in a low productive state and consequently'had a low protein demand, e.g. non-pregnant,non-lactating, near-mature or mature ruminants where protein requirements are low or where energy intake is restricted.

EVALUATION OF PROTEIN MEALS FOR INCLUSION INTO RUMINANT DIETS

The requirement by ruminants for bypass protein under certain dietary and production conditions necessitates feeding small amounts of a protein meal. The quantity of dietary bypass protein required depends on several factors; the protein requirement of the animal (see Fig. 3a); the supply of digestible amino acids from microbial protein (c. 5 g digestible protein/MJ ME), and the supply of amino acids from the basal diet. Having established the digestible bypass protein requirement of the animal, the quantity of protein meal required in the diet can be calculated provided certain characteristics of the meal (including crude protein content, protein solubility in rumen liquor and the digestibility of the protein in the small intestine) are known. A method of protein evaluation of available plant and animal protein meals has been developed in this laboratory based on some readily measurable parameters.

a) Protein content (N x 6.25) following Kjeldahl oxidation procedures.

b) Protein solubility - the protein meal under consideration is shaken in phosphate buffer and the nitrogen in solution as a proportion of total nitrogen used as an index of solubility.

c) Amino acid composition measured using a T.S.M. autoanalyser.

d) Chick growth assay. The relative biological availability of protein meals is examined by adding the test proteins individually to a basal diet and feeding it to chickens. The response in growth rate and food conversion ratio is compared with that from a standard protein.

e) Lamb growth assay. Lambs are established on a basal diet of sugar and oaten chaff, supplemented with minerals and vitamins. Graded amounts of the test protein meals are added to the basal diet and responses in growth and feed intake monitored over 42 d. These

responses are related to responses to feeding graded amounts of a standard protein.

A simple method of protein evaluation based on these tests, either singly or in combination, will give the feed formulator a valuable new tool, which may considerably improve the economics of supplementary and lot feeding.

CONCLUSIONS

In this review we have attempted to demonstrate the inter-relationships between amino acid and glucose requirements of ruminants. From these considerations it is evident that past recommendations on protein requirements for ruminants have been vastly over-simplified, and now need revision.

Present recommendations for the protein content of diets for growth and milk production in ruminants are based on studies with experimental diets which contained significant amounts of bypass proteins. Concentrate diets may also contain bypass proteins and these tend to support efficient microbial systems in the rumen, minimising the need for bypass proteins.

In particular it is now evident that requirements for nrotein cannot be stated adequately in terms of digestible crude protein. Requirements presented in this way apply only to the particular conditions under which they were determined. They are not widely applicable and are often inappropriate.

Requirements of ruminants for protein need to be stated in terms of:

(a) quantities of absorbed essential amino acids per unit of digestible energy;

(b) amounts of glucogenic precursors (i.e. glucogenic amino acid and propionic acid in particular) per unit of digestible energy;

(c) the minimum amounts of essential amino acids relative to glucogenic precursors.

Recommendations for protein content of a diet for ruminants must consider:

(a) the percentage of the dietary protein that is undegraded in the rumen and is digested in the small intestine;

(b) the availability of N in the rumen (i.e. level of rumen ammonia);

(c) the fermentation pattern;

(d) the influence of species composition of the rumen microbial community on the amount and digestibility of microbial protein reaching the intestines.

Evaluation of foods as protein sources for ruminants should be made in terms of:

(a) the availability of N in the forms of ammonia and amino acids for the rumen microbes;

(b) the availability of dietary proteins in the small intestines and their digestion;

(c) the ability of the protein to supply essential amino acids and glucose precursors.

The suitability of treatments of foods must also be evaluated in terms of these factors.

Practical implications. Under applied conditions these stipulations will be difficult to meet, but any approach to teaching, research or practice which ignores or glosses over these complexities will be grossly inadequate. It seems likely that the practical way to formulate diets which are nutritionally and economically optimal for protein will reauire either research, or trial and error in the production system or a large element of empiricism. However the factors considered above should provide a rational basis for these approaches. We stress that the principles developed should apply to all feeding systems and in particular to systems using low protein agro-industrial byproducts. The primary considerations are: (i) that it is necessary to first ensure that the ruminal microorganisms are not restricted for N (i.e. ammonia), and (ii) that the animal is not restricted for amino acids (glucogenic or essential).

The responses of ruminants given low protein diets to supplementary bypass proteins are in terms of increased feed intake and are relatively easily determined in feeding trials. The adequacy of N for the microorganisms under practical conditions is not easily determined but in general this can be relatively inexpensively assured by routine addition of 2 - 4% urea to the feed, (other inexpensive forms of NPN that are totally available will also suffice for this purpose - e.g. poultry manure). At these levels toxicity problems are unlikely and this strategy can therefore be used whenever soluble N deficiency is suspected.

In all countries there is a great need to evaluate the commercially available protein meals in order to determine their potential value as ruminant feeds. The lamb growth assay developed in these laboratories may be one means of doing this under standard conditions in various centres.

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