Chapter 5

5. Manipulation of Protein Degradation in the Rumen

The microorganisms in the rumen degrade a proportion of dietary protein, and utilise the N derived from the rumen-degradable protein (RDP) for the synthesis of microbial protein and other nitrogenous materials during cell growth. Microbial protein and rumen-undegraded dietary protein flowing out of the rumen into the small intestine are the only net source of amino acids available to ruminant tissues. Therefore, protein degradability in the rumen is an important index of the nutritive value of protein feeds for ruminants, and it is also essential for formulation of ruminant diets in order to predict metabolisable protein supply.

5.1 Factors affecting protein degradation

5.1.1 Dietary factors

5.1.1.1 Nature of protein

The solubility of protein, which is largely determined by the ratio of soluble fractions (albumins and globulins) to insoluble fractions (prolamins and glutelins) in the protein (Tamminga 1982), is related to protein degradability. In addition, peptides with a high proline content are more resistant to ruminal degradation (Yang and Russell 1992). Processing methods and pH during processing of forages and concentrates affect the protein solubility of these feeds (Tamminga 1979). Nevertheless, protein solubility is not always a good indicator of protein degradability since protein structure also determines the rate and extent of protein degradation in the rumen (Nolan 1993).

Protein degradability is related to the three-dimensional structure of protein molecules. The distribution of hydrophobic and hydrophylic amino acids on the periphery of protein molecules determines the accessibility of polypeptide chains to hydrolytic enzymes, and thus affects the rate and extent of protein degradation. Feed protein with many hydrophylic residues on its surface but many hydrophobic residues inside its tertiary structure is susceptible to ruminal degradation (Russell and Hespell 1981). Feed protein with extensive crosslinking (e.g., the disulphide bonding) among its peptide chains is less accessible to proteases and therefore more resistant to ruminal degradation (Tamminga 1979; Leng and Nolan 1984). The crosslinking via disulphide

bonds among peptide chains, rather than the protein solubility, is considered to be mainly responsible for the resistance of feed protein to ruminal degradation (Mahadevan *et al.* 1980).

5.1.1.2 Acid detergent fibre nitrogen (ADFN)

The N combined with acid detergent fibre (ADF) in forages, often called acid detergent fibre N (ADFN), is highly resistant to ruminal degradation. The proportion of ADFN in the total N of forages increases with forage maturity, and at the same stage of maturity, legumes have a much higher content of ADFN than grasses (Sanderson and Wedin 1989). As a result, both rumen degradability and intestinal digestibility of forage protein decrease as forages mature (Van Straalen and Tamminga 1990; Hoffman *et al.* 1993).

5.1.1.3 Feed composition

Dietary fats and other water-insoluble materials may protect dietary protein from being degraded in the rumen by coating protein particles and thus reducing the exposed surface area accessible to microbial proteolytic enzymes (Nolan 1993).

Roughage-based diets usually stimulate the cellulolytic activity of rumen microorganisms (Orskov 1982). The increased cellulolytic activity of rumen microbes increases ruminal degradation of plant cell wall materials bound with plant protein, and consequently increases ruminal degradation of plant protein. Therefore, plant protein given with high roughage diets is thus more completely degraded in the rumen than when given with concentrate diets (Schoeman *et al.* 1972).

5.1.2 Microbial and animal factors

5.1.2.1 Concentration of proteolytic enzymes in the rumen

The rate of proteolysis in the rumen is directly related to the ruminal concentration of proteolytic enzymes which is further related to the species and composition of rumen microorganisms (Baldwin and Allison 1983). The diet composition has a major affect on the population and composition of rumen microorganisms, and consequently influences microbial proteolytic activity in the rumen. For example, high quality fresh forages are more capable of promoting the growth rate of proteolytic bacteria in the rumen by supplying more fermentable substrates than low quality forages (Wallace and Cotta 1988) or hay-based diets (Nolan 1993).

Defaunation also reduces the ruminal concentration of aminopeptidases, deaminases and trypsin-like proteases (Merchen and Titgemeyer 1992). Furthermore, ruminal pH affects the species and population of predominant bacteria present in the rumen. The optimum growth rate and activity of proteolytic bacteria require a ruminal pH of 6-7 (Tamminga 1979; Ørskov 1982).

In vitro studies show that certain metal ions, such as Zn^{2+}, depress the activity of microbial proteinases by combining with peptides, and inhibit microbial proteolysis in the rumen (Ha 1996). In addition, some ionophores, such as monensin, natural and synthetic compounds which contain mimetic amino acids or other toxic molecules, selectively inhibit proteolytic rumen bacteria (Morrison and Mackie 1996).

5.1.2.2 Dilution rate

The extent to which feed protein is degraded in the rumen is related to both the nature of the protein and its retention time in the rumen (Mehrez and Ørskov 1977; Beever and Cottrill 1994). Protein degradation is largely affected by the dilution rate of rumen digesta (Leng and Nolan 1984). A higher dilution rate is usually associated with a lower rate of proteolysis in the rumen, which probably results from the reduction in the concentration of microbial proteolytic enzymes and in the reduced retention time of feed protein in the rumen.

The dilution rate of rumen digesta is usually proportional to feed intake by animals. A higher level of feed intake brings about a lower ruminal degradation of feed protein due to the decreased retention time of digesta in the rumen. AFRC (1993) estimated that when the levels of intake by ruminants were one, two and three times maintenance, the dilution rates of rumen fluid were 0.02, 0.05 and 0.08/h, respectively. Feed intake effects may alter the rankings of protein degradability of feeds (Miller 1982b), and the protein degradability of concentrate-based diets is more likely to be affected by the level of feed intake than that of forage-based diets (Van Straalen and Tamminga 1990).

Other factors, such as the processing method of feeds (e.g., grinding and pelleting) (Elimam and Ørskov 1984), ambient temperature, mineral salts (such as sodium bicarbonate) and systemically active compounds (e.g., slaframine and pilocarpine) (Hoover and Stokes 1991) also influence the dilution rate of rumen fluid.

5.1.2.3 Physiological status

The degradability of feed protein may vary with age of animals. For instance, Weston and Margan (1979) reported that the proportion of dietary protein degraded in the rumen decreases with age of lambs from 15 to 40 weeks. Furthermore, the dilution rate of rumen digesta increases during pregnancy (probably due to the reduced rumen volume) and lactation (probably because of the increased intake by animals), which results in a decrease in ruminal degradation of feed protein.

5.1.3 Management

Apart from increasing the N content of pasture forages, N fertilisation increases the protein degradability of pasture forages (Van Straalen and Tamminga 1990). This is probably due to the increased proportion of NPN in the total N of forages.

5.2 Measurements of protein degradability

5.2.1 *In vivo* measurements

Degradability (dg) of feed protein measured *in vivo* is expressed as:

$$dg = 1 - \frac{\text{Non-ammonia duodenal N - (Microbial N + endogenous N)}}{\text{Dietary N intake}}$$

This method requires an accurate measurement of digesta flow through the duodenum and microbial N in duodenal digesta. The former can be measured by a dual marker system, the latter by using internal or external isotope markers, with the assumption that microorganisms isolated from the rumen or duodenal digesta represent those flowing to the duodenum.

The *in vivo* method is a standard measurement of protein degradability, although it is expensive, labor intensive and time-consuming, and its accuracy can be affected by markers used and by animal variation (McDonald *et al.* 1988; Stern *et al.* 1994).

5.2.2 *In vitro* measurements

5.2.2.1 Incubation methods

As reviewed by Miller (1982a) and Stern *et al.* (1994), protein degradability can also be estimated *in vitro* from protein solubility in rumen fluid, enzyme solutions, water, buffer solutions or artificial saliva.

Various microbial proteases have been used in incubation systems simulating ruminal proteolysis for estimating protein degradability, and the estimates are correlated to varying degrees with those determined *in vivo* or *in situ*.

The protein solubility in rumen fluid and in mineral solutions has been used to estimate protein degradability. However, the solubility of protein varies with solvent and is not synonymous with the degradability of protein, so it bears a low relationship with degradability of protein measured *in vivo* or *in situ* over a wide range of feeds However, this technique is useful to compare the degradability of protein within the same class of feeds.

The amount of ammonia and other gases (carbon dioxide and methane) released from an *in vitro* incubation with purified proteolytic enzymes or with rumen fluid has been used as an indicator of protein degradation in the rumen. However, ammonia may be produced from degradation of feed protein and catabolism of microbial protein as well, and its concentration is determined by its rate of assimilation by microbes for growth which can be largely affected by carbohydrates present at the same time.

5.2.2.2 Nylon bag (*in situ*) technique

The nylon bag (*in situ*) technique is most commonly used for estimating protein degradability. With this method, dacron polyester bags containing feed samples are suspended within the rumen of cannulated animals, and N disappearance at various time intervals is measured. The protein degradability (dg) is then calculated as following:

$$dg = \frac{\text{Initial N - N after incubation}}{\text{Initial N}}$$

Although the *in situ* technique is regarded as a relatively reliable and convenient method for measuring protein degradability provided that a standardised procedure is used (Van Straalen and Tamminga 1990), its accuracy can be affected by several factors (SCA 1990; Kandylis and Nikokyris 1991). Firstly, microbial contamination of the feed residues in the nylon bag results in underestimation of the protein degradability of feeds, especially those with a low protein content. As discussed by SCA (1990), the protein degradability of low protein forages and feeds high in starch (e.g., barley and maize grains), or other feeds with a low, but potentially highly degradable protein content, is underestimated due to the extensive bacterial colonisation of feeds during incubation. However, the microbial contamination has a relatively small effect for feeds with a high protein content but a low protein degradability, such as fishmeal and rape seeds.

Secondly, the *in situ* technique is based on the assumption that disappearance of N from the nylon bag is synonymous with degradability. However, a proportion of protein may escape from the bag only partially degraded or even totally undegraded, and solubility and degradability are not highly correlated (McDonald *et al.* 1988; Kandylis and Nikokyris 1991; Stern *et al.* 1994).

Some other factors, such as the porosity and size of bags, ratio of sample weight to bag surface area, particle size of samples, method of suspending bags in the rumen and diet composition also affect *in situ* estimations (McDonald *et al.* 1988; Kandylis and Nikokyris 1991; Stern *et al.* 1994).

Therefore, the procedures for measuring protein degradability by the *in situ* technique should be standardised, in particular for the passage rate of rumen digesta. For example, Elimam and

Ørskov (1984) reported that the degradability of a soybean meal *in situ* increased from 0.47 when the dilution rate of rumen fluid was 0.088 to 0.56 when the dilution rate was 0.064 and further to 0.64 when the dilution rate was 0.047. A standard procedure for the *in situ* technique has been proposed by SCA (1990).

5.2.3 Prediction from feed composition

The degradability of plant protein can be estimated from the chemical composition of plants. For example, the protein degradability (dg) of forages can be estimated by their crude protein (CP) contents and modified acid detergent fibre (MADF) or crude fibre (CF) contents (SCA 1990):

$$dg = \frac{0.90\ (CP - 0.10\ MADF)}{CP}$$

or

$$dg = \frac{0.90\ (CP - 0.125\ CF)}{CP}$$

where MADF, CF and CP are measured as g/kg DM.

However, prediction of protein degradability from feed composition is unlikely to be reliable since protein degradability is affected not only by feed nature, but also by other factors, such as feeding levels.

5.2.4 Prediction from wool production

Wool growth is mainly responsive to the quantity of amino acids absorbed from the small intestine, in particular sulphur-containing amino acids (Reis 1979; Reis and Sahlu 1994). Leng *et al.* (1984) reported that clean wool production in sheep fed a low quality diet was linearly related to intake of supplemental formaldehyde-treated casein, which was almost completely undegradable in the rumen, and the percentage of wool production in sheep given protein supplements relative to that in those given an isonitrogenous amount of formaldehyde-treated casein, was used as an index of the proportion of supplemental protein escaping the rumen. The authors above also suggested that estimating the availability of rumen-undegraded protein from wool production was a relatively easy bioassay for comparing the amount of protein digested and absorbed from protein sources.

However, after comparing the predicted degradability of 10 protein supplements from the wool growth index with their published *in vivo* values, Neutze (1990) concluded that prediction of protein degradability from the wool growth index was insensitive and unreliable. He argued that wool growth varied with not only the amount of the rumen-undegraded protein supplied by a

protein supplement but also the sulphur-containing amino acid content in the rumen-undegraded protein. In addition, estimation of the wool growth index was based on an assumption that the formaldehyde-treated casein was completely undegradable in the rumen but totally digestible in the small intestine. In fact, only 90-98% of the treated casein escaped the ruminal degradation.

5.3 Manipulation of protein degradation in the rumen

The quantity and quality of rumen-undegraded dietary protein can be increased if dietary protein is properly protected from microbial degradation by physical or chemical treatments. The principle of these treatments is to reduce the degradation rate of dietary protein in the rumen but not to reduce the intestinal digestion and absorption of the protected dietary protein (Nolan 1993). Therefore, dietary protein should not be over-protected by these treatments otherwise the intestinal digestibility of the protein will be depressed. The intestinal digestibility of rumen-undegraded protein can be measured by the mobile bag technique (Beever and Cottrill 1994).

5.3.1 Physical treatments

Coating protein sources with whole blood, fish hydrolysate (Mir *et al.* 1984) or calcium soaps of long-chain fatty acids (Sklan 1989) protects feed protein from being degraded in the rumen. However, Ha (1996) noted that blood coating was only effective when heat treatment was applied at the same time.

5.3.2 Chemical treatments

Introducing peptide bonds by heat treatments between lysine residuals and the β- or γ-carboxamide group of asparagine and glutamine residuals, which is referred to the Maillard reaction (Khorasani *et al.* 1993), depresses ruminal degradation of protein. Beever and Cottrill (1994) indicated that the Maillard reaction increased the content of ADFN of feeds. ADFN is highly resistant to ruminal degradation. However, Windschitl (1988) and Stanford *et al.* (1995) reported that controlled Maillard reactions between protein meals and reducing sugars, such as xylose and lignosulfonate, decreased the rumen degradability of protein without affecting the intestinal protein digestibility.

Heat treatments occurring during feed processings, such as roasting, pelleting, extrusion, stream rolling and flaking, effectively reduce protein degradability in the rumen (Clark *et al.* 1992; Ha 1996). The protein of oilseed meals manufactured with treatments of heat, pressure or solvent extraction is less degradable in the rumen than the protein of forages and cereal grains (Nolan 1993). In general, increasing heat input (temperature × time) decreases both the rumen degradability of protein and intestinal digestibility of rumen-undegraded protein (Satter 1986; Van Straalen and Tamminga 1990).

Treating feeds with condensed tannins decreases ruminal degradation of feed protein (Barry and Manley 1984) and sulphur-containing amino acids (McNabb *et al.* 1993), and increases the intestinal flow and absorption of essential amino acids, such as methionine (Waghorn *et al.* 1987; McNabb *et al.* 1993). Tannin treatment creates a stable tannin-protein complex which cannot be broken down above pH 3.5 (Nolan 1993). Tannin treatment protects feed protein from being degraded in the rumen, but does not affect the digestibility of the protected protein at a lower pH in the small intestine (Barry and Manley 1984; Nunez-Hernandez *et al.* 1991). As a result, tannin treatment may shift N digestion from the rumen to the small intestine without affecting the N balance (Nunez-Hernandez *et al.* 1991). Condensed tannins applied at 100 to 150 g/kg DM to a soybean meal give the best protection of its protein from ruminal degradation (Zhou *et al.* 1996). However, tannin treatment may depress ruminal digestion of cellulose and hemicellulose (Beever and Cottrill 1994).

Formaldehyde treatment also increases the proportion of protein escaping the rumen in oilseed meals (Rooke *et al.* 1983; Crooker *et al.* 1986; Faichney *et al.* 1994; Wang and Feng 1995), fresh forages (Beever *et al.* 1987) and silages (Van Straallen and Tamminga 1990) by creating methylene crosslinkages (McDonald 1982; Satter 1986). For example, treating silages or protein meals with formaldehyde reduced protein degradability from about 0.8 to 0.2-0.5 (SCA 1990; Ha 1996). However, the intestinal digestibility of protein overprotected by formaldehyde treatment is also low (Rooke *et al.* 1983). Furthermore, treating plant protein with formaldehyde may reduce the rumen degradability of some specific amino acids creating an imbalance in the array of amino acids absorbed (Miller 1982b).

Alcohol treatment also reduces ruminal degradation of protein by altering the structure of protein (Van Der Aar *et al.* 1984; Lynch *et al.* 1987).

Introducing disulphide bonds into protein by the oxidation of sulphydryl bonds with hydrogen peroxide or treatments with sodium tetrathionate effectively decreases ruminal degradation of dietary protein (Mahadevan *et al.* 1980). In addition, Mir *et al.* (1984) reported that introducing crosslinkages into protein by treating feeds with sodium hydroxide at 20 g/kg DM successfully decreased the degradability of feed protein. More recently, N-terminally modified peptides have been found to be resistant to ruminal degradation (Wallace *et al.* 1993).

Chapter 6

6. Conclusion

Performance of ruminants is difficult to predict from a knowledge of dietary composition because rumen fermentation modifies feeds before gastric and intestinal digestion. Degradation of dietary carbohydrates, lipids and protein, the synthesis of microbial protein, hydrogenation of fatty acids and modification of plant toxins are major functions of rumen microorganisms. The rate and extent of ruminal degradation of feed constituents depend on the solubility, chemical composition, physical structure of the constituents and intake levels. VFA and microbial protein derived from fermentation of dietary carbohydrates, protein and lipids in the rumen are the major sources of energy and amino acids to ruminants. As a result, rumen functions determine feed intake and the quantity and balance of nutrients available to the host animal and thus largely determine animal productivity.

It is logical to partition the N requirements of ruminants into two categories: fermentable dietary N for rumen microorganisms, and dietary protein which escapes rumen fermentation and serves as a direct source of amino acids for the host. This is the core concept of evaluating the N requirement of ruminants in the UK, France, Scandinavia and the USA. More recently, metabolisable protein being composed of digestible microbial true protein and digestible undegraded dietary protein has been proposed by AFRC (1993) to quantify the digestible amino acids available to ruminants for metabolism.

As a result of the subdivision of metabolism into two parts, the primary evaluation of feed protein for ruminants is also considered at two stages: one is microbial protein yield from rumen-degradable dietary protein, the other is the degradability of dietary protein which determines the contribution of rumen-undegraded dietary protein to the total intestinal amino acid flow.

Three strategies are required to improve the quantity and quality of the duodenal amino acid supply to ruminants: maximising microbial yield from the rumen, manipulating the degradability of dietary true protein and the amino acid composition of undegradable protein, and supplementing the animal with ruminally protected protein or amino acids.

Microbial protein is the least expensive amino acid source, and thus its output from the rumen should be firstly optimised. The higher the microbial yield, the higher is the efficiency of microbial protein synthesis and thus the ratio of protein to energy in the nutrients flowing to the duodenum.

Ruminal degradation of dietary protein should be inhibited rather than eliminated since optimum digestion of both non-structural and structural carbohydrates and efficient microbial growth in the rumen require sufficient supplies of ammonia, amino acids or peptides originated from rumen-degradable dietary protein. Further studies that are aimed at developing highly effective and selective means of inhibiting microbial hydrolysis of dietary protein are warranted if in situations where excess rumen-degradable protein is available.

If, however, microbial protein supply to the intestine is insufficient to meet the animal's requirements for a desired level of production, protein supplements which are properly protected by physical or chemical treatments to increase their resistance to ruminal degradation, can be used to augment the intestinal supply of amino acids to animals. A successful manipulation of protein supplementation depends on two factors: one is that ruminal ammonia, peptides and amino acids required by microbial protein synthesis should be satisfied by sufficient supplies of urea and rumen-degradable protein. The other is that protein supplements should be highly resistant to ruminal degradation but readily available to intestinal absorption.

Compared with other internal or external marker techniques, estimation of microbial protein yield from the rumen from urinary excretion of purine derivatives has some distinct advantages: it does not require cannulated animals, collection of total urine and analysis of purine derivatives by the colorimetric method are easily carried out. Therefore, this technique is non-invasive and practical, and can be considered as an alternative to existing methods which require cannulation of the gut of animals. Furthermore, this technique is a useful tool for monitoring the nutritional status of animals, evaluating diets and rumen manipulations, and selecting for animals with a high efficiency of microbial protein synthesis.

Chapter 7

7. Growth Rate and Wool Production in Xinjiang Merino Sheep in Response to Cottonseed Meal and Sunflower Meal Supplementation with or without Maize

7.1 Introduction

Around 22.8 million sheep are raised in Xinjiang Province, China. In 1988, total wool production was 46,400 tonnes, which accounted for more than one-fifth of the annual wool production in China (Zhang and Peng 1994). Nevertheless, productivity of sheep is generally low in Xinjiang, which is largely due to the poor quality of feeds available. In 1988, the annual greasy wool production in finewool sheep was 3.3 kg per head with a clean wool yield of around 42% and the annual meat output of sheep was 4.7 kg per head (Zhang and Peng 1994) Therefore, supplementation of sheep, especially during the winter period, is one of the most important feeding strategies for improving sheep production in Xinjiang Province.

There are two feeding systems for sheep in Xinjiang Province. One is the grazing system in which sheep obtain their major nutrients from seasonal, natural pastures except during the winter when they are given supplements. The other system is found in agricultural areas and lot-feeding situations where sheep are usually kept in sheds throughout the year and fed on straw, stalk or hay sometimes with concentrate supplements.

Production in grazing sheep in Xinjiang is usually limited by the availability and quality of pasture herbages. The investigation carried out by the nutrition group of the China-Australia Sheep Research Project (CASRP 1996) showed that herbage production dropped from 1,446 in summer pastures to 663 kg DM/ha in spring pastures. *In vitro* DM digestibility of pasture herbages ranged from about 57% in summer to 43% in winter and the protein content of herbages decreased from around 11% in spring and summer pastures to 6% in autumn pastures. As a result, liveweight of ewes varied between a maximum of around 56 kg in summer and a minimum of around 45 kg in winter. Clean wool growth rate reached the highest level of 16.6 g/d with the largest fibre diameter of 24.6 μm in summer and dropped to the lowest of 2.9 g/d with the smallest fibre diameter of 16.7 μm in the winter.

Xinjiang is one of major oilseeds-producing areas in China. In 1994, the total oilseed production was 0.5 million tonnes (Xinjiang Statistics Bureau 1995). Oilseed meals, such as cottonseed, sunflower and rapeseed meals, which are potential feed sources for sheep, are abundant in Xinjiang. Therefore, use of oilseed meals as feed supplements for sheep in Xinjiang deserves further study.

Fish meal, soybean meal and cottonseed meal (CSM) are usually regarded as excellent protein sources for ruminants due to their high concentrations of protein which has a relatively low rumen degradability. However, the most appropriate protein supplement for any particular situations depends on its N content, rumen degradability and market price.

The crude protein content and rumen degradability of N supplements readily available in Xinjiang together with their local prices are shown in Table 7.1. Fish meal and soybean meal are not produced locally, and have higher prices. It would be difficult to persuade sheep herders to purchase these supplements even if they were cost effective, and in practice the choice of protein supplements for sheep therefore is restricted to the locally produced oilseed meals.

Table 7.1. *Crude protein (CP), protein degradability (dg) and prices of N supplements readily available in Xinjiang Province, China.*

Supplements	CP *	dg‡	Price (yuan)	
	(%)	(%)	/kg	/kg CP
Cottonseed meal	34.2	60	1.10	3.21
Fish meal	43.8	40	4.70	10.73
Rapeseed meal	36.0	75	1.20	3.33
Soybean meal	40.7	60	1.95	4.79
Sunflower meal	27.6	80	0.75	2.72
Urea	287.5	100	4.20	1.46

*: Xinjiang Animal Husbandry Institute (1984 unpublished).

‡: McDonald *et al.* (1988).

As shown in Table 7.1, sunflower meal (SFM) is the cheapest among the three locally available oilseed meals both in absolute terms and on the basis of cost per unit crude protein. It can be selected in preference to rapeseed meal on this basis without further consideration because there is little difference between them in rumen degradability. The choice between SFM and CSM cannot however be made on the basis of cost per unit of crude protein alone because of the lower rumen degradability and potentially more effective utilisation of the protein in CSM for animal

production. Differences in live-weight gain and wool growth in response to SFM and CSM supplementation must be determined.

Comparison of supplementation of sheep with SFM and CSM was therefore made in this experiment at two energy levels which simulated the two planes of energy intake by sheep in the following two situations in Xinjiang Province: one is for the maintenance of grazing sheep during periods of low pasture availability or quality, and the other is for production (e.g., live-weight gain in young sheep for meat production, or pregnancy and lactation in breeding ewes) either grazing or lot-fed.

7.2 Materials and methods

The experiments reported in this thesis were carried out at Nanshan Stud Farm, Xinjiang Province, China. The farm is located at 43°31′N, 87°00′E with an altitude of about 1,500 m on the northern slopes of the Tianshan mountain range. Annual rainfall at the farm varies between 300 to 600 mm, and mean monthly temperatures range between a minimum/maximum of 10/25 °C in mid summer and a minimum/maximum of -20/-5°C in mid winter (CASRP 1996).

7.2.1 Animals, treatments and management

Twenty-four Xinjiang Finewool ewe weaners approximately seven months old and with a liveweight of around 19.8 (s.d. 2.4) kg were taken from Nanshan Stud Farm, Xinjiang Province.

Owing to the limited availability of metabolism pens, the experiment was carried out in two periods. According to liveweight of the animals, the heaviest 12 animals were used in the first period, and the lightest 12 for the second period.

At the start of the first period, the heaviest 12 were dosed with anthelmintic (Avemectin B1 1%, Beijing Agricultural University) against internal and external parasites, and kept in a naturally lit animal house with temperature ranging from -5 to 5°C.

These 12 sheep were then maintained over two successive feeding regimes: a six-week pre-experimental regime and a six-week experimental regime. Under the pre-experimental regime, the sheep were kept in a group pen and given *ad libitum* milled pasture hay with a daily supply of 10 g mineral mix to each sheep, and live-weight gain, clean wool production and fibre diameter of each sheep were measured, which were used as covariates later for statistical analysis of cereal and oilseed supplementation effects.

The experimental regime consisted of a one-week introductory part and a five-week feeding part. At the beginning of the introductory part, the 12 animals with an initial liveweight of 20.9 (s.d. 1.7) kg were moved into individual metabolism pens. They were then stratified by liveweight into

six groups, and animals within each group were randomly allocated to the following two energy levels:

-No maize grain supplement
-200 g cracked maize grain/h.d

Maize grain was offered to increase total ME intake to the recommended requirement of 20 kg sheep with a growth rate of 150 g/d (6.9 MJ/d, ARC 1984).

Animals with or without maize grain supplement were further stratified by liveweight into two groups, and animals within each group were randomly allocated to the following three oilseed supplement treatments:

-No oilseed supplement
-Cottonseed meal (CSM)
-Sunflower meal (SFM)

Protein meals were offered to increase total N intake to the recommended requirement of 20 kg sheep with a growth rate of 150 g/day (12.8 g N/d, ARC 1984).

As a result, there were six treatments:

Treatment 1: No cereal or oilseed supplement
Treatment 2: 120 g SFM/h.d
Treatment 3: 80 g CSM/h.d
Treatment 4: 200 g maize grain/h.d
Treatment 5: 200 g maize grain + 75 g SFM/h.d
Treatment 6: 200 g maize grain + 45 g CSM/h.d

Each of the supplemented sheep received the same amount of supplemental N daily, i.e., 5.0 g N/d. Supplements were given to sheep at 10 00 h. Then 200 g of milled pasture hay mixed with 10 g mineral mix and 20 ml water containing 10 g urea was fed to each sheep. At 18:00 h milled pasture hay was offered *ad libitum* to each sheep. The amount of hay offered each day was adjusted to 110% of the previous day's consumption to ensure *ad libitum* intake. Water was available all the time.

During the first six weeks of the first period, the lightest 12 sheep were kept in a group pen and fed milled pasture hay *ad libitum* with a daily supply of 10 g mineral mix to each animal. Then the same experimental procedures as described above were applied to these 12 sheep. When the experimental regime started, these sheep had an initial liveweight of 20.5 (s.d. 1.4) kg.

7.2.2 Measurements

7.2.2.1 Feed analysis

Samples of pasture hay, cracked maize grain, CSM and SFM were analysed for dry matter (DM), gross energy (GE) by a bomb calorimeter (Model 1261, Parr, USA), Kjeldahl nitrogen (N) by a nitrogen measurement system (Model Vapodest-5, Gerhardt, Germany), crude fibre (CF) by a fibre measurement system (Model Fibertec M6, Tecator, Sweden), ether extract (EE) and crude ash (Ash) according to AOAC (1980).

In vitro DM digestibility of pasture hay was determined according to Tilley and Terry (1963). In addition, 24 h degradability of crude protein in the rumen of hay, maize grain, CSM and SFM was measured by the nylon bag technique in rumen-cannulated sheep fed hay at the Inner Mongolia College of Animal Science, Inner Mongolia Province, China.

7.2.2.2 Feed intake

All feed offered and feed refusals were weighed and recorded daily.

7.2.2.3 Live-weight gain

The sheep were weighed weekly, and average daily gain in the pre-experimental and experimental regime was calculated.

7.2.2.4 Wool production

At the start of the pre-experimental regime, a 10×10 cm area of mid-side skin on the left side of each sheep was delineated by the tattooing method described by Short and Chapman (1965). Wool grown on the delineated area during each designated experimental period was separately harvested (Oster No. 40 blades). The wool samples were cleaned and weighed, then fibre diameter was measured by an optical fibre diameter analyser (OFDA) at the Wool Technology Laboratory of the China-Australia Sheep Research Centre, Xinjiang Province, China.

7.2.2.5 Apparent digestibility of DM and N

Total collection of faeces was carried out daily for each sheep over the last 5 days of the experimental regime. The faeces were weighed daily and a sub-sample of 10% of the daily output was taken for each sheep, then the sub-samples were pooled for each animal and kept frozen (-18°C) for DM and N measurements. Apparent digestibility of DM and N was then calculated from total intake and faecal output of DM and N over the 5-day period, respectively.

7.2.2.6 Plasma urea-N concentration

On the last day of the experimental regime, a 20 ml blood sample was collected into a glass tube containing 0.5 ml of 6% EDTA from each sheep by jugular venipuncture 4 h after the morning feeding. The blood samples were centrifuged immediately at 3000 r/min for 10 min, and plasma samples obtained were stored at -18°C. The plasma urea-N (PUN) concentration was determined by an enzymatic method at the Xinjiang Medical College, Xinjiang Province, China.

7.2.2.7 Nitrogen balance

Total collection of urine was also carried out daily for each sheep over the last 5 days of the experimental regime. The urine was collected in a bucket containing 100 ml 1M HCl solution, and the volume of urine was recorded daily and diluted to 4 l, then a 20 ml sub-sample of diluted urine was taken and pooled for each anima , and stored at -18°C for Kjeldahl N and allantoin measurements. N retention was calculated from total N intake, urinary and faecal N output over the 5-day period.

7.2.2.8 Microbial protein yield from the rumen

Urinary allantoin concentrations were determined by a colorimetric method described by Chen and Gomes (1992) with a Spectrophotometer (Model Cary 2, Varian, Australia). Microbial protein yield from the rumen was then estimated from urinary excretion of allantoin according to Chen and Gomes (1992).

7.2.3 Statistical analysis

With the aid of the Minitab program (version 9.1, 1992, Minitab Inc., USA), all measurements were subjected to an analysis of variance for a completely random experimental design. The analysis of covariance was conducted to examine live-weight gain, clean wool weight and fibre diameter. Multiple comparisons were carried out by the least significant difference (LSD) test as described by Steel and Torrie (1980).

7.3 Results

The results for live-weight gain, clean wool weight and fibre diameter are present as the least square means obtained from the analysis of covariance. No interaction between protein and maize feeding was detected for any measurements in this study, therefore only main effects of protein and maize supplementation are reported. The mean values of measurements for the six treatments are present in the Appendix.

7.3.1 Feed analysis

The composition of feeds is shown in Table 7.2. *In vitro* dry matter digestibility (IVDMD) and protein degradability (dg) of feeds are shown in Table 7.3.

Table 7.2. *The content of gross energy (GE), dry matter (DM), crude protein (CP), crude fibre (CF), ether extract (EE) and crude ash (Ash) of feeds.*

	GE	DM	g/kg DM			
	(MJ/kg)	(g/kg)	CP	CF	EE	Ash
Pasture hay	16.9	940	88	378	17	81
Maize grain	16.4	885	92	24	37	11
Cottonseed meal	17.7	912	447	117	7	77
Sunflower meal	19.0	954	277	269	90	79

Table 7.3. *In vitro DM digestibility (IVDMD, %) and protein degradability (dg, %) of feeds.*

	IVDMD	dg*
Pasture hay	47.1	68.7
Maize grain	ND	41.0
Cottonseed meal	ND	56.4
Sunflower meal	ND	67.4

ND: Not determined.

*: Net disappearance of protein from sample when placed in the rumen of sheep for 24 h.

7.3.2 Feed intake

Hay DM intake by sheep was not affected ($P>0.05$) by SFM and CSM supplementation, but it was depressed ($P<0.05$) by maize feeding (Table 7.4). However, total DM intake by sheep was increased ($P<0.05$) by SFM or CSM supplementation, but it was not affected ($P>0.05$) by maize feeding (Table 7.4). Hay DM intake and total DM intake by sheep given SFM were not different ($P>0.05$) from those by sheep given CSM (Table 7.4 and Appendix).

7.3.3 Live-weight gain

As shown in Table 7.4 and Figure 7.1, live-weight gain was increased in sheep offered CSM ($P<0.05$) or maize ($P<0.05$). Effects of SFM and CSM feeding on growth rate of sheep were similar ($P>0.05$) (Table 7.4 and Appendix).

7.3.4 Wool production

Clean wool production was higher in sheep given SFM ($P<0.05$) or maize ($P<0.05$) than that in those in the control group (Table 7.4 and Figure 7.2). SFM and CSM supplementation increased ($P<0.05$) diameter of wool fibres, but maize feeding did not affect fibre diameter ($P>0.05$) (Table 7.4 and Figure 7.3). Effects of SFM and CSM supplementation on clean wool growth and fibre diameter were similar ($P>0.05$) (Table 7.4 and Appendix).

Table 7.4. *Hay and total dry matter intake (DMI, g/kg $W^{0.75}$.d), average daily gain (ADG, g/d), clean wool weight (CWW, g/100 cm^2) and fibre diameter (FD, μm) of wool grown by sheep given a basal diet of pasture hay supplemented with protein meals and maize grain (mean ± se)*

	Protein supplement			Maize supplement	
	0	SFM	CSM	0	200 g
DMI					
Hay	60.9±3.1	60.7±4.3	66.5±4.3	67.9^{a}±2.8	57.5^{b}±2.9
Total	69.6^{a}±3.3	78.1^{b}±4.0	80.1^{b}±2.7	74.0±3.1	77.8±2.8
ADG	13.1^{a}±7.6	35.7^{ab}±8.1	58.8^{b}±6.5	21.0^{a}±6.2	50.7^{b}±6.1
CWW	1.7^{a}±0.4	2.2^{b}±0.5	2.1^{ab}±0.5	1.6^{a}±0.3	2.4^{b}±0.4
FD	15.6^{a}±0.8	17.7^{b}±0.7	16.9^{b}±0.9	16.5±0.6	17.0±0.8

$^{a, b}$: Means bearing different superscripts in the same row within protein supplement or maize supplement differ ($P<0.05$).

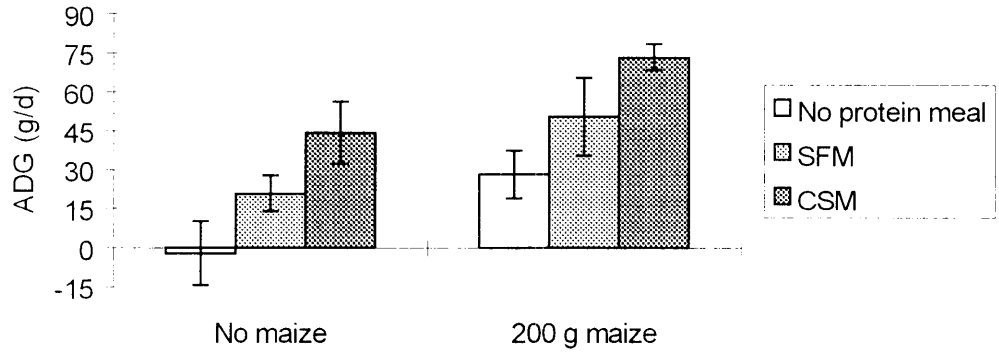

Figure 7.1. *Average daily gain (ADG, g/d) of sheep given a basal diet of pasture hay supplemented with protein meals and maize grain.*

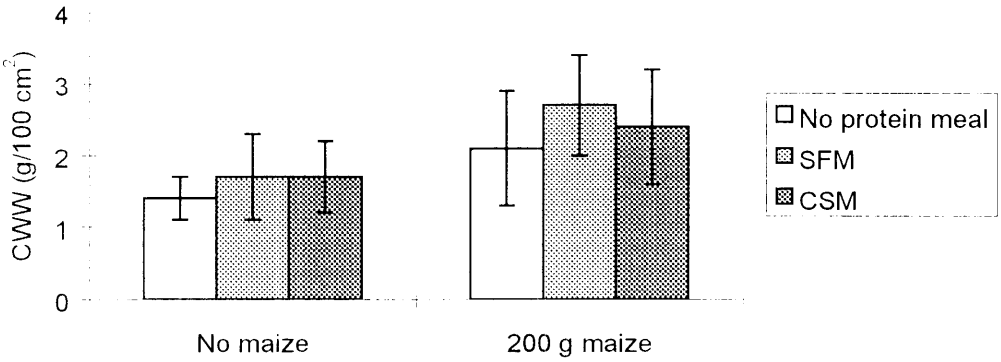

Figure 7.2. *Clean wool weight (CWW, g/100 cm^2) from mid-side patches of sheep given a basal diet of pasture hay supplemented with protein meals and maize grain.*

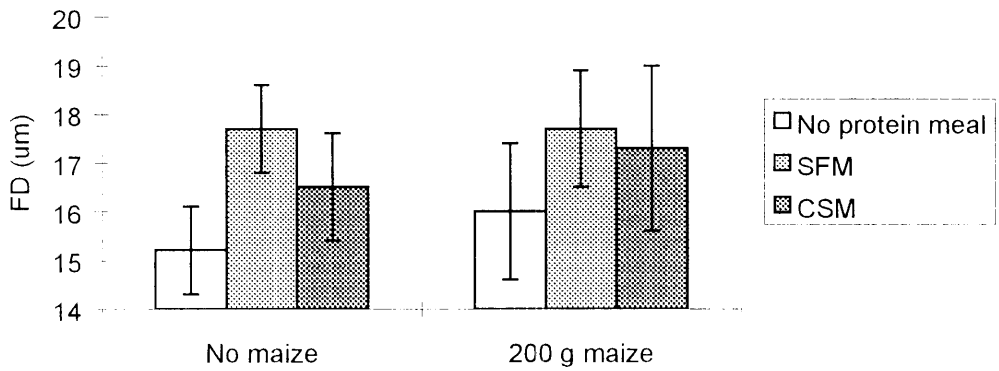

Figure 7.3. *Fibre diameter (FD, μm) of wool grown by sheep given a basal diet of pasture hay supplemented with protein meals and maize grain.*

7.3.5 Apparent digestibility of DM and N

As shown in Table 7.5, apparent DM digestibility (DMD) was higher ($P<0.05$) in sheep given SFM than that in those in the control group or given CSM. Maize feeding also improved DMD ($P<0.05$). Apparent N digestibility was increased ($P<0.05$) by SFM, CSM and maize feeding. Effects of SFM and CSM supplementation on N digestibility were similar ($P>0.05$).

7.3.6 Plasma urea-N concentration

CSM supplementation tended to increase PUN concentration ($P<0.10$), but maize feeding tended to depress PUN concentration ($P=0.09$) (Table 7.5).

7.3.7 Nitrogen balance

The total N intake was increased by protein supplementation ($P<0.05$), but was not affected by maize feeding ($P>0.05$) (Table 7.5).

The total faecal and urinary N outputs were not affected by protein supplementation ($P>0.05$) but were reduced by maize feeding ($P<0.05$). Overall, N retention by sheep was increased ($P<0.05$) by CSM and maize feeding (Table 7.5).

Table 7.5. *Apparent digestibility of dry matter (DMD, %) and nitrogen (ND, %), plasma urea-N (PUN, mg/100 ml), nitrogen intake (NI, g/d), output of faecal nitrogen (FN, g/d) and urinary nitrogen (UN, g/d), and nitrogen retention (NR, g/d) in sheep given a basal diet of pasture hay supplemented with protein meals and maize grain (mean ± se).*

	Protein supplement			Maize supplement	
	0	SFM	CSM	0	200 g
DMD	50.8^{a}±2.0	54.1^{b}±1.9	51.0^{a}±2.1	48.5^{a}±1.3	55.5^{b}±1.3
ND	59.4^{a}±2.2	66.6^{b}±2.8	65.9^{b}±3.2	61.2^{a}±2.2	66.7^{b}±2.5
PUN	15.1±1.5	15.9±2.1	18.9±0.8	18.1±1.2	15.1±1.3
NI	15.4^{a}±0.5	19.5^{b}±0.9	21.0^{b}±0.9	18.8±1.0	18.4±0.8
FN	6.2±0.2	6.4±0.2	7.2±0.8	7.2^{a}±0.6	6.0^{b}±0.4
UN	8.4±0.6	10.0±0.8	8.7±0.6	9.9^{a}±0.5	8.2^{b}±0.7
NR	0.8^{a}±1.1	3.2^{ab}±0.9	5.1^{b}±0.9	1.8^{a}±1.4	4.3^{b}±1.0

$^{a, b}$: Means bearing different superscripts in the same row within protein supplement or maize supplement differ ($P<0.05$).

7.3.8 Microbial protein yield

The total urinary excretion of allantoin, and the predicted microbial N yield from the rumen were increased (P<0.05) by SFM, CSM or maize feeding. Effects of SFM and CSM supplementation on urinary allantoin excretion and microbial N production were similar (P>0.05) (Table 7.6).

Table 7.6. *Excretion of urinary allantoin (UA, mmol/d) and net microbial nitrogen flow out of the rumen (MN, g N/d) predicted from excretion of UA by sheep given a basal diet of pasture hay supplemented with protein meals and maize grain (mean ± se).*

	Protein supplement			Maize supplement	
	0	SFM	CSM	0	200 g
UA	4.7^{a}±0.5	6.2^{b}±1.0	6.4^{b}±0.6	4.4^{a}±0.4	7.1^{b}±0.5
MN	3.6^{a}±0.5	5.1^{b}±0.9	5.3^{b}±0.5	3.4^{a}±0.4	5.9^{b}±0.5

$^{a, b}$: Means bearing different superscripts in the same row within protein supplement or maize supplement differ (P<0.05).

7.4 Discussion

7.4.1 Protein degradability of supplements

In the present study, 24 h *in situ* protein degradabilities of SFM and CSM were 67.4 and 56.4%, respectively. Coombe (1985) reported that 12 h *in situ* protein degradability of SFM in the rumen of sheep fed lucerne and wheaten chaff was 69.5%. Sibanda *et al.* (1993) reported that 24 h protein degradabilities of SFM and CSM measured *in situ* in sheep fed maize stover were 95.9 and 60.7%, respectively, and these authors attributed the lower degradability of CSM protein to its nature and fibre content. However, compared with 24 h protein degradability of SFM (72.0-83.0%) and CSM (60.5-93.0%) measured by the *in situ* technique in sheep fed various basal diets as reviewed by Kandylis and Nikokyris (1991), the protein degradability SFM and CSM measured in the present study were lower.

The protein degradability of maize grain was 41.0%, which was within the range of 24 h protein degradability of maize, i.e., 25.0-64.2%, measured by the *in situ* method in sheep and cattle as reviewed by Kandylis and Nikokyris (1991). In general, compared with other cereal grains, such as oat, wheat, barley, the degradability of maize protein and starch is lower (Spicer *et al.* 1985; Stern *et al.* 1994; Poppi and McLennan 1995).

In this study, microbial growth in the rumen was unlikely to have been limited by the availability of rumen-degraded protein (RDP) since the ratio of RDP to ME intake (MEI) for all diets, as shown in Table 7.7, was higher than 8.4 g/MJ which is recommended as a minimum value by ARC (1984).

Table 7.7. *Calculated values for rumen-degraded protein (RDP, g/d), metabolisable energy intake (MEI, MJ/d), the ratio of RDP to MEI (RDP/MEI, g/MJ), efficiency of utilisation of RDP for microbial protein synthesis (MCP/RDP) and total duodenal protein flow (DPF, g/d) from sheep given a basal diet of pasture hay supplemented with protein meals and maize grain.*

	No maize			200 g maize		
	0	SFM	CSM	0	SFM	CSM
RDP	68.4	89.3	95.9	71.3	85.6	84.4
MEI*	3.9	5.1	5.1	5.5	6.5	6.3
RDP/MEI	17.7	17.5	18.8	12.9	13.2	13.4
MCP/RDP	0.24	0.22	0.29	0.40	0.51	0.46
DPF‡	41.0	54.5	67.9	56.3	78.8	80.0

*: Predicted from apparent DM digestibility of diets (SCA 1990).

‡: = total CP intake - RDP + microbial protein yield (predicted from urinary excretion of allantoin).

7.4.2 Intake of hay

In the present study, despite being different in protein degradability, supplementation with SFM and CSM both supplying 3.0 g CP/kg $W^{0.75}$.d had no effect on hay DM intake by sheep. Unchanged maize stover intake was also reported by Sibanda *et al.* (1993) when sheep were supplemented with SFM and CSM both supplying 3.4 g CP/kg $W^{0.75}$.d. In addition, Huston *et al.* (1993) reported that, when supplementary energy intake was constant and supplemental protein intake was around 7.3 g/kg $W^{0.75}$.d, the difference in protein degradability (58.0 or 82.5%) had no effect on pasture intake by grazing Angora kids.

That there was no response in hay intake by sheep given SFM might be partly related to the relatively high fat content (90 g EE/kg DM) of SFM. Fats increase energy density of ruminant diets. Garcia *et al.*(1993) noted that high-fat supplements, such as rice polishings (163 g EE/kg DM), reduced grass hay intake by sheep. However, Ludden *et al.* (1995) found that addition of fat to corn- or soybean hull-based diets at 5% of diet DM had no effect on total DM intake by wethers.

Hovell *et al.* (1983) proposed that protein supplementation affected forage intake only when forage intake ranged from 20 to 30 g/kg $W^{0.75}$.d. This conclusion seems to be supported by the results of the current study which showed that voluntary hay DM intake by sheep was as high as 65 g/kg $W^{0.75}$.d (see Appendix), and SFM and CSM supplementation had no effect on hay intake. On the contrary, Osuji *et al.* (1993) noted that maize stover intake by sheep was still increased by CSM supplementation when voluntary DM intake of maize stover was 46 g/kg $W^{0.75}$.d.

The lack of a significant effect of protein supplementation on hay intake in this study was possibly related to the N content and particle size of the mixed pasture hay, and limited number of animals in treatments. Protein supplementation is considered to have no effect on intake of forages with a protein content higher than 5-7% (Allden 1981; McCollum and Galyean 1985). In the present study, the pasture hay contained 8.3% of crude protein and was further augmented by a daily supply of 10 g urea to each sheep. In addition, a higher consumption of pasture hay by animals (65 g/kg $W^{0.75}$.d) in the current study was likely due to the small hay particles after hammer-milling. Milling of forages usually increases their intake by animals due to the increased passage rate (McDonald *et al.* 1988; SCA 1990). As a result, hay intake by sheep in this study may have approached the maximum so that protein supplementation was unable to increase hay intake any further. In addition, the availability of NPN may influence the effect of protein supplementation on forage intake. For example, McLennan *et al.* (1995) found that CSM supplementation increased grass hay intake, but decreased intake of ammoniated straw by steers. In the current study, a daily supply of 10 g urea was offered to each sheep. It is unknown whether urea feeding affected hay intake.

In the current study, maize grain given to sheep at a level of 19.0 g/kg $W^{0.75}$.d depressed hay DM intake from 68 to 58 g/kg $W^{0.75}$.d, but did not significantly affect total DM intake. In contrast, Osuji *et al.* (1993) reported that maize grain supplemented at a level of 6.4 g/kg $W^{0.75}$.d had no effect on maize stover intake by sheep. Feeding other energy supplements to animals may also decrease forage intake. For example, Chen *et al.* (1992a) reported that supplementing sheep with sugar-beet pulp and rolled barley both at a rate of 33 g/kg $W^{0.75}$.d depressed intake of ammonia-treated barley straw. Osuji *et al.* (1995) noted that molasses fed to dairy cows at 27.0 g DM/kg $W^{0.75}$.d depressed straw intake, which was attributed by the authors to the possibly increased rumen fill due to the depressed fibre digestion.

In the current study, the depression of hay intake by maize offered in an amount equivalent to 29% of voluntary hay intake by sheep supported the view that forage intake will be depressed if energy supplements account for more than 25% of voluntary forage intake (Weston 1988). Sanson *et al.* (1990) reported that intake of 4.3% CP meadow hay by steers decreased quadratically with increasing maize intake: increasing maize grain supply from 0 to 12.6 g/kg

$W^{0.75}$.d depressed hay intake by 5%, and a further increase from 12.6 to 25.2 g/kg $W^{0.75}$.d depressed hay intake by 17%.

Energy supplementation usually reduces rumen volume, depresses ruminal digestion of fibre and intake of forages (Doyle 1987; Weston 1988, Carey *et al.* 1993; McLennan *et al.* 1995; Osuji *et al.* 1995). Furthermore, the detrimental effect of energy supplementation on ruminal fibre digestion is more evident for low quality forages (Weston 1988). Over 90% of starch in cereal grains may be completely digested in the rumen, which results in a rapid decline in ruminal pH. This depresses the metabolic activity of cellulolytic microbes, consequently the retention time of fibrous feeds in the rumen is extended and forage intake is depressed (Jarrige *et al.* 1986; Van Soest 1994). In addition, Sanson *et al.* (1990) reported that the apparent digestibility of NDF and hemicellulose decreased quadratically with increasing maize intake from 0 to 25.2 g/kg $W^{0.75}$.d by steers fed 4.3% CP meadow hay. Sibanda *et al.* (1993) also found that maize grain, which was slowly degraded in the rumen, depressed ruminal degradation of dietary fibre.

Fibre digestion in the rumen is usually depressed when energy supplements account for 15-20% of the diet (Doyle 1987). In the present study, maize grain was equivalent to around 22% of total feed intake, ruminal fibre digestion was therefore likely to be depressed, and an increase in DM digestibility due to maize feeding likely resulted from the ingestion of more digestible maize supplement.

7.4.3 Plasma urea-N concentration

CSM feeding tended to increase PUN concentration by 25% ($P<0.10$) in this study. Increased PUN concentrations were observed in sheep given CSM (Gaskins *et al.* 1991) and in cattle offered urea plus protein supplements (Hennessy *et al.* 1995). In contrast, Krysl *et al.* (1987) reported that the PUN level in sheep receiving CSM was not different from that in the control animals.

In the present study, at each level of maize feeding, the PUN concentration was approximately proportional to total N intake by sheep (see Appendix). In general, PUN originates from urea synthesised in the liver from ammonia arising from deamination of amino acids and from the surplus ammonia absorbed from the gut. Part of the PUN is recycled to the gut via the saliva or by direct movement through the gut epithelium and may contribute to the N available for microbial protein synthesis, the rest of the PUN is excreted in the urine. PUN concentrations are generally proportional to dietary N intake and ruminal NH_3 concentrations in sheep and cattle (Topps and Thompson 1984; Siddons *et al.* 1985; Gaskins *et al.* 1991; Hoaglund *et al.* 1992; Hennessy *et al.* 1995). For example, Preston *et al.* (1965) reported that in growing lambs

receiving an adequate energy supply, the PUN concentration increased from 2.5 to 32.4 mg/100 ml as dietary protein content increased from 6.2 to 22.0%.

The results from the present study that the total N intake was increased ($P<0.05$), but the PUN concentration was unchanged in the sheep receiving SFM possibly indicated an increase in utilisation of ruminal ammonia, or a reduction in ruminal protein degradation due to the high fat content of SFM. Sheep supplemented with SFM received an average daily supply of 8.2 g crude fat, i.e., 1% of total DM intake. Addition of fats to ruminant diets more than 2-3% of diet DM depresses the population of protozoa and cellulolytic bacteria, thus depressing fibre digestion, methane production and ratio of acetate to propionate in the rumen (Chilliard 1993). The possible mechanisms involved in these changes include the coating effect on fibre digestion, anti-microbial effects on microbial populations and a reduction in Ca available for microbial activity (Jenkins 1993). Most important, infusion of lipids into the rumen of sheep depresses protein degradation and consequently decreases rumen NH_3 concentrations, ultimately increasing intestinal N flow (Ikwuegbu and Sutton 1982; Jenkins and Fotouhi 1990). These effects probably result from reductions in protozoal population in the rumen and in recycled bacterial N, or from increases in dilution rate of rumen particulate digesta (Jenkins 1993).

Maize feeding tended to decrease ($P=0.09$) PUN concentrations in this study. In general, a high energy intake reduces the PUN level by controlling the amount of NH_3 released in and absorbed from the rumen (Topps and Thompson 1984). Sultan and Loerch (1992) noted that when intakes of N and DM by lambs fed wheat straw-based diets were constant, lambs fed at a higher energy level had a lower PUN concentration than those fed at a lower one 4 and 8 h after feeding. Freeman *et al.* (1992) also reported that when supplements supplied 0.52 g CP/kg $W^{0.75}$.d, steers given a maize-CSM-based supplement had a lower ruminal NH_3 concentration than those given a CSM-based supplement, which likely resulted from differences in the degradation rate of supplemental N and in ammonia utilisation by ruminal microbes degrading starch. In the present study, maize feeding also decreased the N output in faeces and urine, and increased microbial protein yield. These suggested an improved utilisation of NH_3 by microorganisms in the gut.

7.4.4 Animal performance

7.4.4.1 Live-weight gain

In the current study, when both SFM and CSM supplied an average amount of 3.0 g CP/kg $W^{0.75}$.d, only CSM supplementation increased average daily gain by 350% ($P<0.05$), which was associated with an increase in N retention by 538% ($P<0.05$). This result was contradictory to the results of Osuji *et al.* (1993) which showed that when both supplements supplied 5.0 g CP/kg $W^{0.75}$.d, sheep fed maize stover receiving SFM grew faster than those receiving CSM (54 vs 44

g/d), and the lower growth rate of sheep given CSM was associated with a poor intestinal digestibility of CSM protein. Usually protein sources with a low rumen degradability are more capable of improving live-weight gain in animals than those with a high degradability (Preston and Leng 1987). However, von Keyserlingk and Mathison (1993) found that when N intake by sheep was similar, sheep supplemented with canola and fish meal concentrates with rumen degradabilities of 78.3 and 59.0%, respectively, had similar growth rates. Likewise, Huston *et al.* (1993) noted that when offered at nearly isonitrogenous levels, a protein supplement with a rumen degradability of 82.5% was as effective as one with a degradability of 58.0% in stimulating live-weight gain in grazing Angora kids.

In this study, as shown in Table 7.7, the estimated ME intakes were the same (5.1 MJ/d) and the calculated duodenal protein flows (67 vs 74 g/d) were similar in sheep given SFM and CSM, but only CSM feeding significantly stimulated live-weight gain in sheep. The reason for this is unclear.

In addition, as shown in Table 7.8, the feed conversion efficiency (live-weight gain/total DM intake) in sheep supplemented with CSM was higher ($P<0.05$) than that in the control animals. These results indicated that CSM feeding was more effective in stimulating growth rate of sheep than SFM feeding.

Table 7.8. *Feed conversion efficiency (FCE, g/kg) in sheep given a basal diet of pasture hay supplemented with protein meals and maize grain.*

	Protein supplement			Maize supplement	
	0	SFM	CSM	0	200 g
FCE	11.7^{a}±6.9	41.3^{ab}±8.3	68.9^{b}±7.6	23.3±8.4	58.0±7.0

$^{a, b}$: Means bearing different superscripts in the same row within protein supplement or maize supplement differ ($P<0.05$).

SFM feeding increased intake and digestibility of N, and microbial protein yield, but it did not significantly increase N retention or growth rate of sheep in the present study. Coombe (1985) reported that sheep fed a SFM-straw-based diet had higher intake, digestibility and retention of N, and higher live-weight gain than those fed a urea-straw-based diet. In the current study, although SFM had a higher fat content than CSM, estimated ME intakes by sheep supplemented with SFM and CSM were the same (5.1 MJ/d, Table 7.7). Fats can be used to increase energy density of ruminant diets, to prevent acidosis and to manipulate the fatty acid content of meat and milk fat (Chilliard 1993). Ludden *et al.* (1995) reported that fat offered at 5% of diet DM had no effect on growth rate of lambs given corn- or soybean hull-based diets. Chilliard (1993)

concluded that addition of fats at 3 to 8% of diet DM either increased or had no effect on growth rate of steers, and fats offered at 7 to 14% of diet DM did not affect growth rate.

The present study showed that wool production, rather than live-weight gain, was improved by SFM supplementation. As shown in the Appendix, when all sheep received an equal amount of 31.8 g CP/h.d from supplements, the sheep given SFM as the only supplement and those given maize as the only supplement had similar live-weight gains (21 vs 28 g/d). Results of CASRP (1996) also showed that supplementation of Xinjiang Merino ewes grazing autumn pastures with different levels of SFM (40, 80 and 120 g/h.d) did not improve either live-weight gain or wool production.

In Xinjiang Province, supplementation of sheep for wool production is unlikely to be economic due to the low price of clean wool. However, the local price for mutton is higher than that for wool, so supplementation of sheep for meat production is likely to be economic. Even with SFM, which is the least expensive supplement available in Xinjiang, a considerably greater response would be required to make supplementation economically viable. Treatments which reduce degradability of SFM protein may improve the feeding value of SFM for growth. For example, Coombe (1985) demonstrated that formaldehyde treatment reduced rumen degradability of SFM from 69.5 to 32.5% in sheep fed lucerne and wheaten chaff, and sheep fed oat straw receiving formaldehyde-treated SFM had a lower ruminal NH_3 level and retained more N than those receiving untreated SFM when both supplements supplied an equal amount of N. Similarly, Faichney *et al.* (1994) reported that formaldehyde treatment of SFM reduced ruminal NH_3 concentration by 30%, and increased N retention by sheep by 150%. The treatment of SFM and use of protected SFM in Xinjiang Merino sheep deserve further study.

An increase of 140% ($P<0.05$) in live-weight gain in sheep supplemented with maize was also noted in the present study, which was associated with increases in digestibility of DM and N, N retention and microbial protein yield from the rumen. Energy supplementation usually increases the proportion of propionate in the VFA in rumen fluid, but decreases methane production from the rumen (Van Soest 1994), and increases digestibility of OM and DM (Sultan and Loerch 1992; Osuji *et al.* 1995). The increased growth rate (Osuji *et al.* 1993; McLennan *et al.* 1995; Osuji *et al.* 1995) and milk yield (Osuji *et al.* 1995) of animals receiving energy supplements are considered to originate from the increased VFA production, microbial protein and lipid yield from the rumen and from a higher efficiency of energy utilisation in the host animal (Nolan and Leng 1989; Howard *et al.* 1992; Osuji *et al.* 1993; Van Soest 1994; Osuji *et al.* 1995).

Sibanda *et al.* (1993) suggested that energy supplements with a low rumen degradability, such as maize grain, should be offered with protein supplements with a intermediate or low rumen degradability (e.g., CSM) to enhance the utilisation of ruminal NH_3 by rumen microbes.

However, in the present study, microbial protein production in sheep given maize grain was similar whether they were supplemented with SFM or CSM (7.0 vs 6.2 g N/d, see Appendix), despite the higher protein degradability of the SFM.

7.4.4.2 Clean wool production

In this study, although protein degradability of SFM was higher than that of CSM and should have provided less augmentation of amino acid supply to the intestine, feeding SFM, not CSM, increased wool production. Coombe (1985) reported that sheep fed a SFM-straw-based diet had higher wool growth rate than those fed a urea-straw-based diet, but the efficiency of wool growth was similar for the two diets. In contrast, Huston *et al.* (1993) reported that, when supplemental energy intake by grazing Angora kids was constant and supplemental protein intake was around 7.3 g/kg $W^{0.75}$.d, fibre length growth rate was higher for a higher degradability protein supplement (82.5%) than for a lower degradability one (58.0%), but clean fleece weight and fibre diameter were unaffected by the difference in protein degradability.

Although the calculated duodenal protein flows (67 vs 74 g/d) did not differ significantly between sheep supplemented with SFM and CSM, SFM supplementation stimulated wool growth by 30% ($P<0.05$). This might be related to the higher content of sulphur-containing amino acids (S-AA) in SFM. SFM is higher in methionine (7.6 vs 5.2 g/kg) and cystine (9.2 vs 6.4 g/kg) compared with CSM (McDonald *et al.* 1988). In this study, the net microbial N flows from sheep given SFM and CSM were nearly the same (5.1 vs 5.3 g N/d, Table 7.6). The calculated total rumen-undegraded protein supplies to sheep given SFM and CSM were also similar (34.8 and 40.9 g/d), and the amounts of rumen-undegraded protein from SFM and CSM (31.4 vs 27.5 g/d) accounted for 90 and 67% of the total rumen-undegraded protein supplies in sheep receiving SFM and CSM, respectively. Therefore, the amount of S-AA in duodenal protein flow in sheep offered SFM might be a little higher than that in those offered CSM. This might be responsible for a higher wool production in sheep given SFM since the supply of intestinal amino acids, in particular S-AA, largely determines wool production (Reis 1979; Reis and Sahlu 1994).

Maize feeding increased clean wool production by 50% ($P<0.05$, Table 7.4) without affecting fibre diameter. These results were consistent with the study of Huston *et al.* (1993) who found that, when maize grain was given to grazing Angora kids at a level equivalent to 50% of their maintenance energy requirement, clean fleece weight was increased, but fibre diameter was unaffected. The increased wool production in response to maize supplementation in the present study was associated with an increase of 74% ($P<0.05$, Table 7.6) in microbial protein yield and a higher duodenal protein flow (54.4 vs 71.7 g/d, Table 7.7).

Increasing post-ruminal energy supply to sheep has a minor effect on wool growth (Kempton 1979; Reis and Sahlu 1994), staple strength and protein composition of wool fibres (Reis 1991). Even though the bulb cell in wool follicles utilises both glucose and acetate for the synthesis of wool fibres (Black and Reis 1979), Black (1987) estimated that the maximum daily growth rate of 20 g clean wool required 0.43 MJ of energy, which represented only about 9% of the basal metabolic rate of a 40 kg sheep. Hoaglund *et al.* (1992) also reported that feeding pregnant ewes at 80% or 100% of ME requirement had no effect on greasy wool production and fibre diameter.

7.5 Conclusion

Supplementation with CSM rather than SFM improved N retention and live-weight gain in sheep fed low digestibility pasture hay. Therefore, CSM appeared to be an effective supplement for profitable meat production in Xinjiang Merino sheep. CSM may be used to improve the nutritional status of grazing sheep, in particular during the winter when the availability and quality of pasture forages are low. Production in sheep in agricultural areas, which feed on straw, stalk and other low quality agricultural by-products and are occasionally given energy supplements, may be improved by CSM supplementation. For lot-feeding sheep given concentrate diets, incorporation of CSM into their diets may further improve their productivity.