S.Afr.J.Anim.Sci., 1992, 22 (6)

Substitution of lucerne hay by untreated, urea-enriched and urea-ammoniated wheat straw in diets for sheep

T.S. Brand,* F. Franck and S.W.P. Cloete
Elsenburg Agricultural Development Institute, Private Bag, Elsenburg, 7607 Republic of South Africa

A.A. Brand
P.O. Box 101, Laaiplek, 7370 Republic of South Africa

Received 21 May 1991; revised 10 December 1991; accepted 11 August 1992

The aim of the study was to examine the effects of the substitution of lucerne hay by untreated, urea-enriched and urea-ammoniated wheat straw on (i) dry matter intake (DMI), digestibility and nitrogen balance, (ii) *in situ* degradability, (iii) rate of passage from the rumen and (iv) ruminal pH, volatile fatty acid and ruminal ammonia concentrations of sheep. Wheat straw was left untreated (WS), enriched with 1.5% urea (UWS), or ammoniated with 5.5% urea in a stack for 8 weeks (AWS). It was then hammermilled (18-mm screen) and used to substitute lucerne hay (LH) at LH : WS / UWS / AWS ratios of 100 : 0, 75 : 25, 50 : 50 and 25 : 75 in a 3 × 4 factorial experiment involving the three straw treatments and four substitution levels. These diets were fed to 24 adult SA Mutton Merino wethers in a voluntary dry matter (DM) intake, *in vivo* digestibility and nitrogen (N) balance trial. These procedures were replicated twice to get four replicates per treatment. Rate of passage from the rumen (using ^{51}Cr as external marker) and rumen parameters (pH, ammonia-nitrogen, volatile fatty acids), were investigated similarly. *In situ* organic matter (OM) degradability was determined according to a 3 × 3 factorial experiment, excluding LH from the analysis. Voluntary DMI declined linearly ($P \leqslant 0.01$) by 224 ($SE_b = 33$) g/sheep/d for each increment increase of WS/UWS/AWS. A quadratic tendency ($P = 0.09$) was also observed, indicative of some associative action between LH and the straws. This trend was consistent for all straw treatments. No significant effect of LH level on apparent DM and OM digestibility was found, while apparent NDF, ADF, hemicellulose and cellulose digestibility increased linearly ($P \leqslant 0.01$) with decreasing amounts of LH. Apparent N retention significantly ($P \leqslant 0.01$) decreased with decreasing amounts of LH in the diet. This trend could be attributed directly to higher N intake levels on the diets high in LH, as apparent N retention as percentage of N intake was independent of LH level. The effective OM degradability of the diet decreased linearly ($P \leqslant 0.01$) with a decline in level of LH with the substitution of LH by WS and UWS. When LH was substituted by AWS, there was also evidence of a quadratic response ($P \leqslant 0.01$), suggesting some associative action. Level of LH had no significant influence on outflow rate from the rumen, although it tended ($P \leqslant 0.12$) to decrease with increasing amounts of straw. Ruminal pH increased ($P \leqslant 0.05$) on diets including higher amounts of straw, while ruminal volatile fatty acid concentrations followed an opposite trend ($P \leqslant 0.01$). Ruminal ammonia N decreased ($P \leqslant 0.01$) with the substitution of LH by WS and AWS but it was curvilinearly influenced by the substitution of LH by UWS. In general, ammoniation improved ($P \leqslant 0.10$) voluntary DM intake, as well as the apparent digestibility coefficients of DM, OM and the various fibre fractions relative to untreated WS. Rate of passage from the rumen was significantly ($P \leqslant 0.05$) shorter on AWS than on untreated WS. This study accorded with previous results that ammoniation, in general, improves voluntary DMI, apparent digestibility of fibre fractions and apparent N retention. No conclusive associative effects between LH and WS, UWS or AWS were observed with regard to these factors or rumen parameters, apart from the results regarding voluntary DMI, and the *in situ* OM degradability in the case of AWS.

Die studie is uitgevoer ten einde die invloed van die vervanging van lusernhooi met onbehandelde, ureum-verrykte of ureum-geammonifiseerde koringstrooi op die (i) DM-inname, verteerbaarheid en stikstofbalans, (ii) *in situ* degradeerbaarheid, (iii) uitvloeitempo uit die rumen en die (iv) pH en vlugtige-vetsuur- en rumenammoniakkonsentrasies van skape te bepaal. Koringstrooi van dieselfde besending is óf onbehandeld gelaat (WS), met 1.5% ureum verryk (UWS), óf in 'n mied vir 8 weke met 5.5% ureum geammonifiseer (AWS). Die strooi en lusernhooi (LH) is daarna deur 'n hamermeul gemaal (18 mm-sif) en gebruik om LH stapsgewys, in verhouding van LH : WS / UWS / AWS van 100 : 0, 75 : 25, 50 : 50 en 25 : 75 te vervang. Die invloed van die vervanging van strooi met LH is in 'n 3 × 4-faktoriaalontwerp, met die drie tipes strooi en vier vervangingspeile, geëvalueer. Die diëte is aan 24 volwasse Suid-Afrikaanse Vleismerinohamels gevoer en DM-inname, *in vivo* verteerbaarheid en N-balans is bepaal. Die prosedure is herhaal ten einde vier waarnemings per behandeling te verkry. Uitvloeitempo uit die rumen (^{51}Cr as eksterne merker) en rumenparameters (pH, ammoniak-N en vlugtige-vetsuurkonsentrasies) is volgens dieselfde prosedure ondersoek. *In situ* organiese-materiaal(OM)-degradeerbaarheid is volgens 'n 3 × 3-faktoriaalontwerp bepaal, met LH uitgesluit uit die analise. DM-inname het reglynig ($P \leqslant 0.01$) afgeneem met 224 ($SE_b = 33$) g/skaap/d, met elke verhogingsvlak van WS/UWS/AWS in die dieet. 'n Kwadratiese neiging ($P = 0.09$) het ook voorgekom, wat op die bestaan van assosiatiewe effekte tussen LH en WS dui. Geen betekenisvolle invloed van LH-peil op skynbare DM- en OM-verteerbaarheid het voorgekom nie, terwyl NDF, ADF, hemisellulose- en sellulose-verteerbaarheid reglynig ($P \leqslant 0.01$) toegeneem het met dalende peile van LH. Skynbare N-retensie het betekenisvol ($P \leqslant 0.01$) afgeneem met dalende vlakke van LH. Hierdie neiging kon direk aan die hoër N-inname op die hoër LH-diëte toegeskryf word, aangesien skynbare N-retensie as persentasie van N-inname nie beïnvloed is nie. Die effektiewe OM-degradeerbaarheid van die diëte het reglynig afgeneem ($P \leqslant 0.01$) met 'n afname van LH in die dieet. By AWS het dit reglynig sowel as kwadraties ($P \leqslant 0.01$) afgeneem, wat dui op 'n mate van assosiatiewe werking tussen LH en AWS. Die peil van LH het geen invloed op die uitvloeitempo uit die rumen gehad nie, alhoewel dit geneig het ($P \leqslant 0.12$) om te daal met stygende peile strooi in die diëte. Rumen-pH het toegeneem ($P \leqslant 0.05$) by die diëte met hoër strooi-inhoud, terwyl vlugtige-vetsuurkonsentrasie die teenoorgestelde neiging ($P \leq 0.01$) getoon het. Rumenammoniak-N-konsentrasies het gedaal ($P \leq 0.01$) by die vervanging van LH deur WS en AWS, maar is kurvilineêr beïnvloed deur die vervanging van LH deur UWS. Oor die algemeen het ammonifisering DM-inname sowel as DM-, OM- en veselvertering relatief tot WS verhoog ($P \leq 0.10$). Uitvloeitempo uit die rumen was betekenisvol ($P \leq 0.05$) korter vir AWS as vir WS. Samevattend ondersteun die studie vorige resultate dat ammonifisering DM-inname, verteerbaarheid van die veselfraksies en skynbare N-retensie verbeter. Geen definitiewe assosiatiewe werking tussen LH en WS, UWS of AWS in terme van hierdie faktore of die gemete rumenparameters is waargeneem nie, behalwe die resultate betreffende vrywillige DMI, en die *in situ* OM-degradeerbaarheid by AWS.

Keywords: Ammoniation, *in situ* degradation, lucerne hay, rate of passage, rumen ammonia, sheep, volatile fatty acids, wheat straw.

* To whom correspondence should be addressed.

Introduction

Lucerne hay is valued highly as a feedstuff for sheep, but is expensive and difficult to obtain in some years. Wheat straw, on the other hand, is normally abundant in the grain-producing areas. The degree to which straw can be utilized as a feedstuff for sheep is, however, limited due to a poor nutrient content (O'Donovan, 1983) and low inherent digestibility (Leng, 1982). Increasing the straw content of diets also reduced dry matter (DM) intake and production (O'Donovan, 1983; Brand *et al.*, 1991b).

Chemical upgrading of low-quality roughages by ammonia (Sundstøl *et al.*, 1978) has been frequently investigated over the past few years. Several studies (Cloete & Kritzinger, 1984; Brand & Cloete, 1988; Brand *et al.*, 1991a) indicated that ammoniation increases the nutritive value and utilization of wheat straw. Romero *et al.* (1970) found that the addition of urea to wheat straw diets increased feed intake and suggested that the role of urea was to increase feed intake presumably through an increased rate of fermentation. Leng (1982) therefore accentuated the need for a continuous supply of urea in the rumen, and suggested that the optimum rumen ammonia-nitrogen concentration for maximal microbial synthesis is in the order of 5—8 mg/100 ml (Satter & Roffler, 1977). The higher intake due to ammoniation may decrease retention time in the rumen (Thornton & Minson, 1972; 1973). This will tend to increase the ratio of microbial cells to volatile fatty acids (VFA). Under these circumstances less energy substrates and more microbial protein would be available from rumen fermentation (Leng, 1982), which would probably increase the efficiency of utilization of feed. Oji *et al.* (1979), on the other hand, reported an increase in the total VFA concentrations in the rumen due to the ammoniation of roughages.

Several researchers found positive associative effects between lucerne hay and poor-quality roughages in terms of digestibility (Hunt *et al.*, 1985), intake (Soofi *et al.*, 1982), VFA, ruminal ammonia-nitrogen (NH_3-N) levels, rate of passage from the rumen and *in sacco* rate of fibre digestion (Ndlovu & Buchanan-Smith, 1985) as well as daily gain (Brandt & Klopfenstein, 1986a). Leng (1982) postulated that there is a strong possibility that a small supplement of lucerne hay may have a beneficial effect on microbial growth, by providing essential co-factors.

The objectives of this study were therefore to determine the extent of improvement in the utilization of untreated, urea-enriched or urea-ammoniated wheat straw by the addition of lucerne hay, and to quantify the improvement in the utilization of wheat straw due to ammoniation. The effects of the substitution of lucerne hay by untreated, urea-enriched or urea-ammoniated wheat straw on (i) dry matter intake, digestibility and nitrogen balance, (ii) *in situ* degradability, (iii) rate of passage from the rumen and (iv) ruminal pH, VFA and ruminal ammonia concentrations of sheep were therefore examined.

Experimental procedure

The same batch of wheat straw was divided and treated as follows:

1. Wheat straw (no treatment applied) (WS).
2. Wheat straw enriched with 1.5% urea. Solution applied and dried prior to mixing the diets (UWS).
3. Wheat straw ammoniated with 5.5% urea in a stack, covered with a plastic sheet for 8 weeks and dried afterwards (AWS) (Cloete & Kritzinger, 1984).

Lucerne hay (LH) as well as WS, UWS and AWS were hammermilled through a 18-mm screen, whereafter LH was substituted in a stepwise fashion by WS, UWS and AWS. The diets were composed to contain 100, 75, 50, and 25% lucerne hay combined with 0, 25, 50, or 75% of either WS, UWS or AWS, and were thoroughly mixed in a horizontal mixer.

Metabolism trial

Twenty four rumen-cannulated South African Mutton Merino (SAMM) wethers were used as experimental animals (2 per diet) and the procedure was replicated twice. The wethers were randomly allocated to the different diets and subjected to a 45-day trial period, consisting of a 21-day adaption period, a 10-day period to calculate intake, followed by 4 days at 85% of *ad libitum* intake prior to a 10-day collection period, during which faeces and urine were collected. During the collection period, the experimental animals were fed at a level of 85% of *ad libitum* intake in two equal portions at 8:00 and 16:00. They had free access to water at all times. Representative feed, faeces and urine samples were taken daily and pooled for analysis. Dry matter (DM), organic matter (OM) and crude protein (CP) contents of feed and faeces samples as well as the N content of urine samples were determined (AOAC, 1984). Lignin, acid detergent fibre (ADF), neutral detergent fibre (NDF), cellulose and hemicellulose contents were determined as described by van Soest (1963) and van Soest & Wine (1967). Apparent digestibility coefficients were calculated for DM, OM, CP, ADF, NDF, cellulose and hemicellulose, while apparent N retention was also obtained.

Degradability study

The OM degradability of the 10 differently composed diets (comprising LH and LH substituted by 25, 50, or 75% WS, UWS, or AWS) were determined by the *in situ* techniques described by Ørskov & McDonald (1979) and Ørskov *et al.* (1980). Polyester bags (14 × 9 cm) with a 53-µm mesh were used. Diet samples (2 g) were milled through a Wiley mill with a 1-mm sieve, and placed into the bags. The bags containing samples were then placed in lukewarm water (35—40 °C) for 1 min, whereafter they were inserted into the rumens of three rumen-fistulated sheep, for periods of 2, 4, 12, 24, 36, 48, or 72 h. This procedure was replicated, giving a total of six observations for each variable studied. The sheep received lucerne hay (milled through an 18-mm screen) *ad libitum* during the experimental period. At the end of each incubation period, the bags were removed from the rumen, rinsed under running tap water until no further colour washed out from the bags, and dried in a force-draught oven at 65 °C for 48 h. Contents of bags were removed and residual OM (AOAC, 1984) was determined. The percentage of material degraded was determined according to the model of Ørskov & McDonald (1979):

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

where p = the actual degradation of time t; a = the intercept of the degradation curve at time zero; b = the potential degradability of the insoluble fraction; and c = the rate constant for the degradation of the fraction described by b.

Iterative least-squares procedures were used for the determination of the non-linear functions (Stratgraphics, 1986). The effective degradability (P) was calculated from the equation:

$$P = a + \frac{bc}{c + k}\ ,$$

where k = the outflow rate from the rumen. Fractional outflow rates of 0.02 and 0.05 were used in the calculations. No provision for lag time was made due to a relative short lag time (± 20 min), which would not affect the effective degradation (McDonald, 1981).

Rate of passage

The rate-of-passage study was executed as described with the metabolism trial. The wethers were adapted to the different diets, at *ad libitum* intake, for 14 days. Thereafter, approximately 45 g of feed, marked with ^{51}Cr mordant (Udén *et al.*, 1980) was placed into the rumen of each sheep, and mixed thoroughly with rumen contents. Each sheep received between 0.1 and 0.2 mCi ^{51}Cr. Faeces were collected every 8 h for 10 successive days. Radioactivity in the faeces was determined using a Gamma counter and was expressed in cpm ^{51}Cr per gram dry matter. The mean retention time ($\bar{Rt}$) of the marked particles was calculated from the excretion curve, using the following equation (Pienaar & Roux, 1984):

$$\bar{Rt} = \frac{1}{N}\ \Sigma\{\,n[t^1 + t) \div 2]\}\ ,$$

where n = ^{51}Cr counts excreted between t and t^1, representing the sum of these counts for successive intervals until n = 0, and N = total number of marker counts excreted. The rate of passage was calculated as the inverse of $\bar{Rt}$.

Rumen parameters

Ruminal samples from four rumen-fistulated sheep per diet (procedure described in the metabolism trial) were collected at the end of the rate-of-passage study. Samples were collected at three times (2, 4, and 8 h post feeding) and pooled for analyses. The pH of the samples were measured immediately after sampling. The ruminal fluid samples were then separated from the solid fractions by a 0.5-mm sieve. Samples for rumen NH_3-N determination (50 ml) were acidified with 10 ml 1N H_2SO_4 and frozen. Rumen ammonia concentrations in the rumen fluid were determined by an Auto Analyser with Industrial Method no 334-74 W/B (1977) (Technicon Industrial Systems, Tarrytown, New York). Samples for VFA determination were centrifuged at 3 000 revolutions per minute (r.p.m.) for 20 min. 10% NaOH (1 ml) was added to 9 ml of the supernatant liquid, to prevent fermentation, and frozen at –15°C. Prior to analysis, the samples were treated with a 25% metaphosphoric acid solution per 5 ml of sample, and centrifuged at 3 000 r.p.m. for 10 min. Acetic, propionic, isobutyric, n-butyric and isovaleric acid concentrations were determined using a gas chromatography by the procedure described by Cottyn & Boucque (1968).

Statistical analysis

The results were analysed according to standard procedures for a 3 × 4 factorial design (Snedecor & Cochran, 1980), involving the three straw treatments and four LH substitution levels. Initially, the replication of the experiment over two periods was also included as a factor. In the absense of a significant influence or significant interactions involving period, it was excluded from the final analyses. Where no significant ($P > 0.05$) interaction occurred between main effects, the degrees of freedom for levels of LH substitution were partitioned into orthogonal polynomials, depicting linear and quadratic trends. In the presence of significant ($P \leqslant 0.05$) interactions between straw treatment and substitution level, a nested design was used. In this analysis, orthogonal polynomials were fitted within straw treatments. Regression coefficients describing responses to LH substitution were expressed on a per increment substitution basis, i.e. LH levels of 100%, 75%, 50%, or 25% were coded as 1, 2, 3, or 4. In case of the OM degradability study, the LH diet was excluded from the statistical analysis, which was conducted according to a 3 (type of straw) × 3 (level of substitution) factorial design.

Results and Discussion

Chemical composition

Substitution of LH by WS resulted in a marked reduction in CP content of the diet (Table 1), due to the much lower CP levels in WS compared to LH (3.4% vs. 17.0% CP respectively). A similar trend was observed in the case of UWS and AWS, but the magnitude of the reduction was reduced, owing to a smaller difference in CP content between LH and UWS (11.7% CP) or AWS (8.0% CP). Substitution of LH by wheat straw generally resulted in increased NDF, ADF, hemicellulose and cellulose contents, while lignin content tended to decline (Table 1). Corresponding results, involving 60:40 roughage: concentrate diets and the substitution of LH by AWS, were obtained previously (Brand *et al.*, 1990).

Table 1 Chemical composition of the experimental diets (DM basis)

	Chemical composition							
Diet[1]	Dry matter	Organic matter	Crude protein	Acid detergent fibre	Neutral detergent fibre	Hemi-cellulose	Cellulose	Lignin
100 LH	92.1	90.9	17.0	39.0	48.7	9.7	31.5	7.5
75 LH : 25 WS	92.4	92.0	13.6	42.1	55.9	13.5	35.1	7.2
50 LH : 50 WS	92.7	93.2	10.2	45.2	61.1	17.4	38.8	6.9
25 LH : 75 WS	93.0	94.3	6.8	48.4	70.3	21.2	42.4	6.6
75 LH : 25 UWS	92.4	92.0	15.7	42.1	55.9	13.5	35.1	7.2
50 LH : 50 UWS	92.7	93.2	14.4	45.2	63.1	17.4	38.8	6.9
25 LH : 75 UWS	93.0	94.3	13.1	48.4	70.3	21.2	42.4	6.6
75 LH : 25 AWS	92.4	92.0	14.8	41.9	56.0	14.1	34.6	7.1
50 LH : 50 AWS	92.7	92.5	12.5	44.8	63.2	18.5	37.6	6.8
25 LH : 75 AWS	93.0	93.3	10.3	47.8	70.6	22.8	40.7	6.5

[1] LH = lucerne hay; WS = untreated wheat straw; UWS = urea-enriched wheat straw; AWS = urea-ammoniated wheat straw.

Table 2 Dry-matter intake, DM, OM, NDF, ADF, hemicellulose and cellulose digestibility and N balance of differently combined diets in which lucerne hay was substituted (25%, 50%, 75%) by untreated, urea-enriched and urea-ammoniated wheat straw

	Level of lucerne hay				*SE*	
Parameter	100%	75%	50%	25%	Mean	LSD*
Dry-matter intake (g/d)	2 248	1 957	1 591	1 556	74	228
Apparent digestibility of:						
Dry matter (%)	61.3	59.4	59.1	59.1	0.9	2.77
Organic matter (%)	61.8	59.8	59.4	59.5	0.9	2.77
Neutral detergent fibre (%)	48.4	52.1	55.0	60.6	1.0	3.08
Acid detergent fibre (%)	50.6	51.1	54.1	59.6	1.0	3.08
Hemicellulose (%)	39.1	53.3	59.3	68.6	1.9	5.85
Cellulose (%)	60.9	61.3	63.1	66.7	1.2	3.70
Nitrogen balance:						
Nitrogen intake (g/d)	45.5	35.9	24.7	19.1	1.2	3.70
Nitrogen excretion (g/d)						
Faeces	12.1	10.5	8.0	7.4	0.4	1.23
Urine	25.5	19.6	13.0	8.3	0.7	2.16
Apparent nitrogen retention (g)	7.9	6.0	4.4	3.5	0.8	2.47

* $P \leq 0.05$.

Metabolism trial

The effect of LH level (100%, 75%, 50%, and 25%) on the results of the voluntary intake and digestion trial is presented in Table 2, while the effect of type of straw (WS, UWS and AWS) is presented in Table 3. No interactions ($P > 0.05$) were observed between LH substitution level and straw type, except in the case of apparent CP digestibility, and these results are presented separately in Table 4. Dry-matter intake (DMI) was linearly reduced ($P \leq 0.01$) by 224 ($SE_b = 33.1$) g/d for each increment increase in WS inclusion level. This trend accounted for 92% of the variance in DMI associated with LH substitution levels. Although not significant, there was evidence of a quadratic response ($P = 0.09$), contributing another 5.1% to the variance accounted for by the diets, and suggesting the possibility of limited associative action between LH and wheat straw with regard to DMI. The linear regression of DMI on LH level was the following:

$$\text{DMI} = 1\,838\ \text{g} - 244.2\,(x - 2.5)$$

where x represents the inclusion levels of 1(25%), 2(50%), 3(75%), and 4(100%). The equation for the quadratic response was the following:

$$\text{DMI} = 1\,758\ \text{g} - 244.2\,(x - 2.5) + 63.96(x - 2.5)^2.$$

These results were, however, in general agreement with results reported by Brand *et al.* (1990; 1991b), who found linear decreases in DMI as LH was substituted by AWS or WS in

Table 3 Dry-matter intake, DM, OM, NDF, ADF, hemicellulose and cellulose digestibility and N balance of untreated, urea-enriched and urea-ammoniated wheat straw fed in combination with lucerne hay to sheep

	Type of straw [1]				
Paramenter	WS	UWS	AWS	*SE* mean	LSD*
Dry-matter intake (g/d)	1 748	1 819	1 946	64	192
Apparent digestibility of:					
Dry matter (%)	58.2	60.1	60.9	0.78	2.34
Organic matter (%)	58.7	60.5	61.2	0.76	2.28
Neutral detergent fibre (%)	51.8	53.4	57.3	0.89	2.67
Acid detergent fibre (%)	52.5	54.9	54.4	0.87	2.60
Hemicellulose (%)	48.3	51.6	65.2	1.69	5.07
Cellulose (%)	61.9	62.8	64.3	1.00	3.00
Nitrogen balance:					
Nitrogen intake (g/d)	26.4	33.4	34.1	1.05	3.15
Nitrogen excretion (g/d)					
Faeces	8.5	9.1	11.1	0.35	1.05
Urine	13.3	19.1	17.3	0.62	1.86
Apparent nitrogen retention (g)	4.6	5.2	5.7	0.72	2.16

[1] WS = untreated wheat straw; UWS = urea-enriched wheat straw; AWS = urea-ammoniated wheat straw.

* $P \leq 0.05$.

Table 4 Apparent crude protein digestibility of 10 different diets in which lucerne hay was substituted (25%, 50%, 75%) by untreated, urea-enriched and urea-ammoniated wheat straw

Diet [1]	Crude protein digestibility (%)*
100 LH : 0 WS	72.6
75 LH : 25 WS	70.1
50 LH : 50 WS	64.9
25 LH : 75 WS	50.9
100 LH : 0 UWS	72.7
75 LH : 25 UWS	73.3
50 LH : 50 UWS	72.2
25 LH : 75 UWS	72.8
100 LH : 0 AWS	74.6
75 LH : 25 AWS	67.3
50 LH : 50 AWS	65.3
25 LH : 75 AWS	56.3

[1] LH = lucerne hay; WS = untreated wheat straw; UWS = urea-enriched wheat straw; AWS = urea-ammoniated wheat straw.

* *SE* mean = 1.30%; LSD ($P \leq 0.05$) = 5.13%.

diets for growing lambs. Hunt *et al.* (1988) similarly reported no significant associative effect in voluntary DMI when LH was substituted by UWS or AWS. Although not directly comparable, it should be stated that Soofi *et al.* (1982) found positive associative effects with regard to DMI between LH and soybean stover. No significant effects of lucerne hay level were found on DM or OM digestibility. The digestibilities of NDF, ADF, hemicellulose and cellulose, however, increased linearly ($P \leq 0.01$) with increasing wheat straw levels in the diet. These trends ranged from 1.90 (SE_b = 0.51) units in the case of cellulose, to 9.45 (SE_b = 0.88) for hemicellulose, the trends accounting for ≥ 87% of the variance introduced by LH substitution levels. N retention (g/d) on the other hand, significantly ($P \leq 0.01$) decreased with decreasing levels of lucerne hay in the diet, which could probably be attributed to the higher N intake, as apparent N retention as percentage of N intake (17–18%) was independent of LH levels. The results found in this study were in agreement with results found by Hunt *et al.* (1988), who also observed no indication of associative action between wheat straw and lucerne hay for DM and NDF digestibilities. It is, however, in contrast to the results of other low-quality roughages, reported by Soofi *et al.* (1982) and Hunt *et al.* (1985). They found positive associative effects between LH and soybean stover and between LH and fescue hay respectively.

Dry-matter intake did not differ between the different types of straw, although intake tended ($P = 0.10$) to be improved relative to WS by urea ammoniation of wheat straw (Table 3). In general, the apparent digestibilities of DM ($P \leq 0.05$), OM ($P \leq 0.07$), NDF ($P \leq 0.01$), ADF ($P \leq 0.08$), hemicellulose ($P \leq 0.01$) and cellulose ($P \leq 0.22$) were also improved in AWS, when compared to untreated WS. Nitrogen retention was unaffected ($P > 0.05$) by straw treatment, but tended to be improved by either urea enrichment (13.0%) or ammoniation (23.9%). These tendencies could be ascribed to the higher ($P \leq 0.05$) N intake on the latter diets, since apparent N retention as percentage of N intake (16—17%), was similar for the different types of straw. The tendencies in this study generally accord with results by Cloete *et al.* (1983) and Cloete & Kritzinger (1984), but the magnitude of the differences in the present study was markedly smaller than in previous studies. This observation could possibly be ascribed to the masking of effects due to straw treatment by the inclusion of LH in the diets evaluated in the present study.

The effect of the substitution of LH by WS, UWS or AWS on apparent CP digestibility was only significant in the case of WS and AWS (Table 4). Apparent CP digestibility linearly decreased ($P \leq 0.01$) by respectively 7.0 ($SE_b = 0.6$) and 5.7 ($SE_b = 0.6$) units with increasing amounts of WS and AWS in the diet. These trends accounted for respectively 88% and 94% of the variance associated with the substitution of LH by WS or AWS. In the case of UWS, no significant effect of substitution level was found. This lack of a decline in CP digestibility was cancelled by a higher nitrogen excretion level in the urine of sheep on this diet (Table 3), thus resulting in no advantage in favour of UWS in the apparent N-balance results (Table 2). Cloete & Kritzinger (1984) similarly found a significant ($P \leq 0.01$) higher CP digestibility with urea-supplemented wheat straw in comparison to untreated and ammoniated wheat straw. In their study, the advantage due to higher N digestibility results was also cancelled by increased urinary-N losses.

Degradability study

Statistical appraisal of the results of the OM-degradability study suggested that level of lucerne hay and type of straw interacted ($P \leq 0.01$) for most of the depended variables and results were therefore given in terms of the different diets (nine diets in which LH was substituted by WS, UWS and AWS with LH *per se* as reference) (Table 5). The non-linear parameters (a, b and c) as well as the effective degradation (P at $k = 0.02$/h and $k = 0.05$/h) differed significantly ($P \leq 0.05$) between diets. The solubility at time zero (a) declined linearly ($P \leq 0.03$), while the potential degradability of the insoluble fraction (b) increased linearly ($P \leq 0.01$), with increasing levels of the differently treated straw in the diets. A quadratic response ($P \leq 0.01$) was also observed in the case of a (for WS and AWS) and b (for WS). The rate constant (c) declined linearly ($P \leq 0.001$) with increasing amounts of straw in the diet, while a quadratic response ($P \leq 0.02$) was found with (AWS and UWS), which indicated that with an increasing amount of lucerne hay, the diet was degraded more rapidly. The effective degradation (P) decreased linearly ($R^2 > 0.81$) with a decline in lucerne hay in the diet, suggesting that increased wheat straw levels in the diet suppressed OM fermentation. In the case of LH substitution by AWS, a quadratic response ($P \leq 0.01$) was also obtained, suggesting some associative action. The lack of a quadratic response with OM degradability in case of the UWS and WS substituted diets indicated that no associative effects occurred as was found for apparant OM digestibility (Table 2). Results of the 100% lucerne hay diet *per se*, were in agreement with the results obtained with the substituted diets and indicated a higher soluble fraction (a), lower potential degradable fraction (b), more rapid degradation (c) and higher effective degradation (P) than the substituted diets. The improved OM degradation with increasing levels of lucerne hay in the diet corresponds well

Table 5 Organic matter degradability of 10 differently combined diets in which lucerne hay was substituted by untreated, urea-enriched and urea-ammoniated wheat straw as estimated by the model of Ørskov & McDonald (1979)

	Non-linear parameters[1]			Effective degradation[2] (%) at fractional outflow rate of	
Diet[3]	a	b	c	0.02/h	0.05/h
100 LH[4]	29.4	40.5	0.153	65.1	59.7
75 LH : 25 WS	24.6	43.1	0.114	61.2	54.4
50 LH : 50 WS	25.7	40.0	0.075	57.1	49.5
25 LH : 75 WS	19.3	49.0	0.051	54.5	44.0
75 LH : 25 UWS	25.6	44.1	0.117	63.2	56.4
50 LH : 50 UWS	25.4	45.5	0.059	59.3	49.9
25 LH : 75 UWS	23.3	50.1	0.045	57.9	46.9
75 LH : 25 AWS	28.8	43.6	0.117	65.9	59.2
50 LH : 50 AWS	20.3	50.8	0.068	59.5	49.5
25 LH : 75 AWS	17.6	61.2	0.049	60.8	47.7
SE	0.69	1.31	0.005	0.69	0.69
LSD ($P \leq 0.05$)	1.98	3.73	0.014	1.97	1.97

[1] Calculated by the iterative least-squares procedures fitting the model $p = a + b(1 - e^{-ct})$.

[2] Calculated by the equation $P = a + \frac{bc}{c + k}$ for $k = 0.02$, $k = 0.05$.

[3] LH = lucerne hay; WS = untreated wheat straw; UWS = urea-enriched wheat straw; AWS = urea-ammoniated wheat straw.

[4] Diet excluded from statistical analysis.

with results reported by Ndlovu & Buchanan-Smith (1985), who also found increased rates of DM disappearance when brome grass, barley straw and corncobs were supplemented with lucerne hay. Ørskov *et al.* (1980) related the potential DM intake of roughages to OM degradability. In our study, mean OM degradability figures on the respective diets were highly correlated with mean DM-intake figures (r = 0.92; n = 10). These results appear to implicate that DM intake on diet high in wheat straw was restricted by a low degradability potential, with the possible exception of the diets containing AWS.

Rate of passage

Results of the rumen outflow study are presented in Table 6.

Table 6 Mean retention time and outflow rates from the rumen of 10 differently combined diets in which lucerne hay was substituted by untreated, urea-enriched and urea-ammoniated wheat straw as estimated by the equation of Pienaar & Roux (1984)

Text component	Mean retention time (h)	Mean outflow rate (h)
Level of lucerne hay:		
100%	53.50	0.019
75%	57.30	0.018
50%	57.60	0.018
25%	58.70	0.017
SE	2.30	0.0007
LSD ($P \leq 0.05$)	6.64	0.002
Type of straw: [1]		
WS	59.70	0.017
UWS	57.50	0.018
AWS	53.20	0.019
SE	1.90	0.0007
LSD ($P \leq 0.05$)	5.82	0.0019

[1] WS = untreated wheat straw; UWS = urea-enriched wheat straw; AWS = urea-ammoniated wheat straw.

Level of lucerne hay had no significant influence on outflow rate as well as retention time, although retention time tended ($P \leq 0.12$) to increase as LH was substituted by increasing amounts of wheat straw. Ndlovu & Buckhanan-Smith (1985) reported that the rumen retention time of barley straw was unaffected by LH supplementation, while it was reduced when corncobs were treated similarly. Low DM-intake levels on diets high in wheat straw (Table 2) could be attributed to a decreasing tendency in rumen outflow rates, although it appeared to be less important than DM degradability. Type of straw, however, significantly ($P \leq 0.05$) affected retention time, which was significantly shorter for ammoniated wheat straw compared to untreated wheat straw. These results accorded with those published by Oji *et al.* (1979). Baumgardt *et al.* (1976) related the potential intake of low-quality roughages to physical factors. This study supports the contention that ammoniation beneficially influences DM intake through a reduction in rumen retention time (Tables 3 & 6).

Rumen parameters

Ruminal pH values of sheep on the different diets are presented graphically in Figure 1. The pH of the rumen contents varied within the range of 6.27 and 6.85 on the diets, and there was a tendency for pH to increase on diets containing higher levels of wheat straw. The UWS diets maintained a higher pH than WS or AWS diets throughout. The pH generally followed a linear response ($P \leq 0.01$) with increasing levels of straw. The regression coefficients ranged from 0.052 (SE_b = 0.02; $P \leq 0.05$) units in the case of WS to 0.14 (SE_b = 0.02; $P \leq 0.01$) in the case of UWS. These trends accounted for $\geq 68\%$ of the variance associated with the substitution of LH. The lower pH found on the diets high in LH was probably an indication of increased fermentation (Weidemeier *et al.*, 1983), while the higher pH on the UWS diet and 50% and 75% AWS diets could possibly be ascribed to the effect of NH_3 on rumen pH. The higher fermentation rate on diets high in LH was largely confirmed by the DM-degradability study (Table 5). The reduction in total VFA concentration (Figure 3) could also have led to the increase in pH (Kaufmann, 1977).

Type of straw and level of lucerne hay interacted ($P \leq 0.01$) in the case of rumen NH_3-N concentration and the results are therefore presented separately in Figure 2. Rumen NH_3-N concentration declined linearly by respectively 5.3 and 6.7 ($R^2 \geq 0.93$; SE_b = 1.2; $P \leq 0.01$) mg NH_3-N/100 ml rumen fluid with decreasing levels of LH where it was substituted by WS or AWS. In the case of UWS, a quadratic response curve was associated with 99.7% of the variation introduced by LH substitution level. Higher levels of rumen ammonia-N concentrations were maintained in the case of AWS than with WS. These results were in accordance with the N intakes found with different levels of lucerne hay substitution and types of straw (see Tables 2 & 3), except when LH was substituted by UWS. The quadratic response curve in this case could be ascribed to the readily available NPN provided by the urea included in this diet, in combination with NH_3-N provided by the degradation

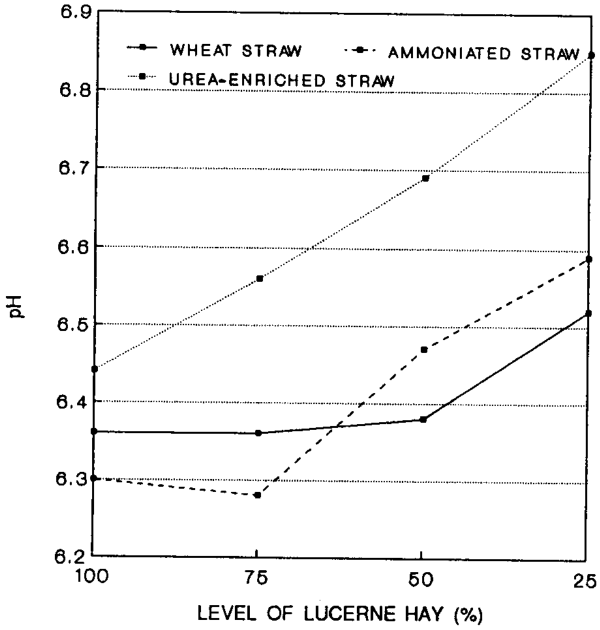

Figure 1 Ruminal pH of sheep which received diets in which lucerne hay was gradually substituted by unprocessed, urea-enriched and urea-ammoniated wheat straw.

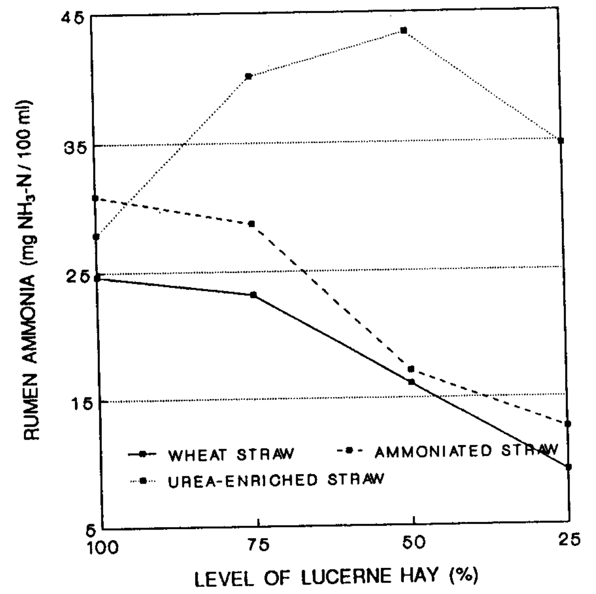

Figure 2 Ruminal ammonia nitrogen concentrations of sheep which received diets in which lucerne hay was gradually substituted by unprocessed, urea-enriched and urea-ammoniated wheat straw.

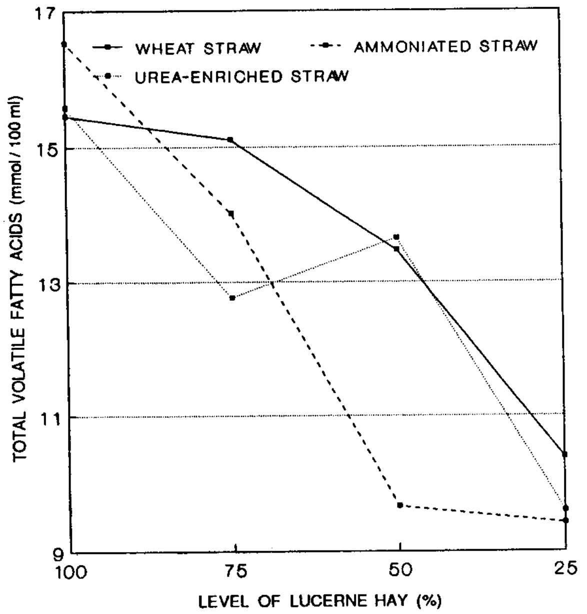

Figure 3 Ruminal volatile fatty acid concentrations of sheep on differently combined diets in which lucerne hay was gradually substituted by unprocessed, urea-enriched and urea-ammoniated wheat straw.

of CP in LH. Rumen NH_3-N levels may be influenced by several other factors like total CP intake, OM digestibility and outflow rate (Roffler & Satter, 1975a; 1975b). Rumen NH_3-N concentration was, however, in excess of the minimum requirement of 5—8 mg NH_3-N /100 ml rumen fluid proposed by Satter & Roffler (1977) on all diets.

No specific tendency in acetate:propionate ratio, as well as in percentage of individual volatile fatty acid (VFA) concentrations in relation to total VFA concentrations, was found. The ratio of acetic acid:propionic acid varied between 3.5:1 and 3.7:1, which was within the normal range for high-roughage diets (Ruckebush & Thivend, 1980). Due to the lack of differences in individual VFA concentration ratios, results are presented only in terms of total VFA concentrations.

The effects of level of LH substitution and straw type on total VFA concentration are graphically illustrated in Figure 3. The effects of type of straw and substitution level interacted ($P \leq 0.01$), but the substitution of LH resulted in a linear decline in VFA concentration regardless of type of straw. These trends ranged between −1.63 (SE_b = 0.38; $P \leq 0.01$) mmol total VFA/100 ml rumen fluid in the case of WS to −2.52 (SE_b = 0.38; $P \leq 0.01$) in the case of AWS, and accounted for ≥ 78% of the variation introduced by LH substitution level. The higher VFA concentrations found on the diets high in LH may be ascribed to the higher fermentation rate (Weidemeier *et al.*, 1983).This is clearly confirmed by the degradability study (Table 5). The tendency towards a lower total VFA concentration found on the 50% and 75% AWS diets, on the other hand, could be ascribed to the shorter retention time in the rumen (Table 6), and the enhancement of VFA utilization by ruminal micro-organisms (Leng, 1982; Brandt & Klopfenstein, 1986b). The VFA concentrations on the diets high in LH were, however, higher than the normal range (6—12 mmol/100 ml; Ruckenbusch & Thivend, 1980), while levels on the diets high in straw accorded with normal values.

Conclusions

No significant positive associative affects for LH with UWS and AWS on DM intake, digestibility, nitrogen balance, OM degradability (in the case of WS and UWS), rumen retention time and VFA concentrations were found. A significant ($P \leq 0.01$) positive associative affect was observed for OM degradability in the case of AWS, while a tendency ($P \leq 0.09$) towards positive associative action also occurred for DM intake. There were suggestions that an increased retention time in the rumen limited DM intake on diets containing WS relative to those including AWS. The degradation of OM decreased markedly with increasing levels of WS and UWS, implicating that the lower DM intake of diets containing high levels of wheat straw was associated with reduced rumen fermentation levels. This contention was supported by tendencies in ruminal pH levels and VFA concentrations.

Acknowledgements

Mrs Joan Collier and other personnel of the Animal Nutrition Laboratory at the Animal and Dairy Science Research Institute, Irene are thanked for the determination of volatile fatty acids and ruminal ammonia-N concentrations. The authors also thank the Elsenburg Animal Production Laboratory personnel for chemical analyses and Mr P. Adams and Mr. S Adams for feeding and care of the animals.

References

AOAC, 1984. Official methods of analysis. Association of Official Analytical Chemists, Washington DC.

BAUMGARDT, B.R., KRABILL, L.F., GROBBLE, J. & WANGENESS, J., 1976. Estimation feed intake for cattle, sheep and swine. In: Animal Nutrient Requirements and Computerisation of diets. Ed. Fonesbeck, P.V., Logan Utah 84322, USA.

BRAND, A.A. & CLOETE, S.W.P., 1988. Die benutting van ureumaangevulde en verskillende tipes geammoniseerde garsstrooi by skape. *S.-Afr. Tydskr. Veek.* 18, 118.

BRAND, A.A., CLOETE, S.W.P. & FRANCK, F., 1990. Substitution of lucerne hay by ammoniated wheat straw in growth diets for lambs. *S. Afr. J. Anim. Sci.* 20, 78.

BRAND, A.A., CLOETE, S.W.P. & FRANCK, F., 1991a. Supplementation of untreated wheat straw, urea-supplemented wheat straw and urea-ammoniated wheat straw with maize meal and/or fish meal for sheep. *S. Afr. J. Anim. Sci.* 21, 48.

BRAND, T.S., CLOETE, S.W.P., VAN DER MERWE, G.D. & FRANCK, F., 1991b. Wheat straw as roughage component in finishing diets of growing lambs. *S. Afr. J. Anim. Sci.* 21, 184.

BRANDT, R.T., JR & KLOPFENSTEIN, T.J., 1986a. Evaluation of alfalfa corn cob associative action. I. Interactions between alfalfa hay and ruminal escape protein on growth of lambs and steers. *J. Anim. Sci.* 63, 894.

BRANDT, R.T., JR & KLOPFENSTEIN, T.J., 1986b. Evaluation of alfalfa corn cob associative action. III. The effect of mechanically separated alfalfa fractions on intake, digestibility and ruminal characteristics of ammonia-treated corn cob diets fed to sheep. *J. Anim. Sci.* 63, 911.

CLOETE, S.W.P., DE VILLIERS, T.T. & KRITZINGER, N.M., 1983. The effect of ammoniation by urea on the nutritive value of wheat straw for sheep. *S. Afr. J. Anim. Sci.* 13, 143.

CLOETE, S.W.P. & KRITZINGER, N.M., 1984. Urea ammoniation compared to urea supplementation as a method of improving the nutritive value of wheat straw for sheep. *S. Afr. J. Anim. Sci.* 14, 60.

COTTYN, B.G. & BOUCQUE, C.V., 1968. Rapid method for the gas-chromatographic determination of volatile fatty acids in rumen fluid. *J. Agric. Food Chem.* 16, 105.

HUNT, C.W., KLOPFENSTEIN, T.J. & BRITTON, R.A., 1988. Effect of alfalfa quality and wheat straw ammoniation on intake and digestibility of alfalfa – wheat straw combination diets fed to sheep. *Nut. Rep. Int.* 37, 675.

HUNT, C.W., PATERSON, J.A. & WILLIAMS, J.E., 1985. Intake and digestibility of alfalfa – tall fescue combination diets fed to lambs. *J. Anim. Sci.* 60, 301.

KAUFMANN, W., 1977. Influence of the composition of the ration and the feeding frequency on pH-regulation in the rumen and on feed intake in ruminants. *Livest. Prod. Sci.* 3, 103.

LENG, R.A., 1982. A theoretical discussion on the factors limiting production in cattle fed basal diets of straw. Proc. 3rd Bangladesh Conf. on Straw Utilization. pp. 79–104. Bangladesh Agricultural University, Mynenssingh, Bangladesh.

MCDONALD, I., 1981. A revised model for the estimation of protein degradability in the rumen. *J. Agric. Sci., Camb.* 96, 251.

NDLOVU, L.R. & BUCHANAN-SMITH, J.G., 1985. Utilization of poor quality roughages by sheep: Effect of alfalfa supplementation on ruminal parameters, fibre digestion and rate of passage from the rumen. *Can. J. Anim. Sci.* 65, 693.

O'DONOVAN, O.B., 1983. Untreated straw as livestock feed. *Nutr. Abstr. Rev.* 63(7), 441.

OJI, U.I., MOWATT, D.N. & BUCHANAN-SMITH, J.G., 1979. Nutritive value of thermo-ammoniated and steam treated maize stover. II. Rumen metabolites and rate of passage. *Anim. Feed. Sci. Technol.* 4, 187.

ØRSKOV, E.R., HOVELL, E.P. DE B. & MOULD, F., 1980. The use of the nylon bag technique for the evaluation of feedstuffs. *Trop. Anim. Prod.* 5, 195.

ØRSKOV, E.R. & MCDONALD, I., 1979. The estimation of protein degradability in the rumen from incubation measurement weighted according to rate of passage. *J. Agric. Sci., Camb.* 92, 499.

PIENAAR, J.P. & ROUX, C.A., 1984. Inferred mixing compartments in the sheep's digestive tract. *Can. J. Anim. Sci.* 64, 74.

ROFFLER, R.E. & SATTER, L.D., 1975a. Relationship between ruminal ammonia and non-protein nitrogen utilization by ruminants. I. Development of a model for predicting non-protein nitrogen utilization by cattle. *J. Dairy Sci.* 58, 1980.

ROFFLER, R.E. & SATTER, L.D., 1975b. Relationship between ruminal ammonia and non-protein nitrogen utilization by ruminants. II. Application of published evidence to the development of a theoretical model for predicting non-protein nitrogen utilization. *J. Dairy Sci.* 58, 1989.

ROMERO, V.A., SIEBERT, B.D. & MURRAY, R.M., 1970. A study on the effect of frequency of urea ingestion on the utilization of low quality roughage by steers. *Aust. J. Exp. Agric. Anim. Husb.* 16, 304.

RUCKEBUSCH, Y. & THIVEND, P., 1980. Digestive physiology and metabolism in ruminants. A.I.V. Publishing Company, Inc. Westport, Connecticut, USA.

SATTER, L.D. & ROFFLER, R.E., 1977. Influence of nitrogen and carbohydrate inputs on rumen fermentation. In: Recent advances in animal nutrition. Eds. W. Haresign & D. Lewis. London, Butterworths, p. 25.

SNEDECOR, G.W. & COCHRAN, W.G., 1980. Statistical methods (7th edn). The Iowa State University Press, Ames, Iowa, USA.

SOOFI, R., FAHEY, G.C., JR., BERGER, L.L. & HINDS, F.C., 1982. Digestibilities and nutrient intakes by sheep fed mixtures of soybean stover and alfalfa. *J. Anim. Sci.* 54, 841.

SUNDSTØL, F., COXWORTH, E. & MOWAT, D.N., 1978. Improving the nutritive value of straw and other low-quality roughages by treatment with ammonia. *World Anim. Rev.* 26, 13.

THORNTON, R.F., & MINSON, D.J., 1972. The relationship between voluntary intake and mean apparent retention time in the rumen. *Aust. J. Agric. Res.* 23, 871.

THORNTON, R.F. & MINSON, D.J., 1973. The relationship between apparent retention time in the rumen, voluntary intake and apparent digestibility of legumes and grass diets in sheep. *Aust. J. Agr. Res.* 24, 889.

UDÉN, P., COLUCCI, P.E. & VAN SOEST, P.J., 1980. Investigation of chromium, cerium and cobalt as markers in digesta rate of passage. *J. Sci. Food Agric.* 31, 625.

VAN SOEST, P.J., 1963. Use of detergents in the analysis of fibrous feeds. II. A rapid method of the determination of fibre and lignin. *J.A.O.A.C.* 46, 825.

VAN SOEST, P.J. & WINE, R.H., 1967. Use of detergents in the analysis of fibrous feeds. II. Determination of plant cell wall constituents. *J.A.O.A.C.* 50, 50.

WEIDEMEIER, R.D., MALES, J.R. & GASKINS, C.T., 1983. Effect of dietary crude protein on the dry matter digestibility of wheat straw diets in cattle. *J. Anim. Sci.* 57, 1568.