EFFECTS OF LEUCAENA (*L. LEUCOCEPHALA* CV TARRAMBA) LEUCAENA WITH UREA OR LEUCAENA WITH SUCROSE SUPPLEMENTATION ON INTAKE AND DIGESTIBILITY OF RHODES GRASS (*CHLORIS GAYANA* CV CALLIDE) HAY FED TO SHEEP

I W. KARDA
Fakultas Peternakan, Universitas Mataram, Nusa Tenggara Barat 83125
Telephon 633603

ABSTRACT

This experiment was conducted to assess the intake and digestibility of low quality rhodes grass (*Chloris gayana*) hay cv. Callide offered to sheep. Four rumen fistulated sheep were assigned to four dietary treatments with 21 days for each period namely, rhodes grass hay was fed alone *ad lib.* (treatment 1), rhodes grass hay was supplemented with 200 g/d oven-dried leucaena (treatment 2), rhodes grass hay was supplemented with 200 g/d leucaena plus 8 g/d urea (treatment 3) and rhodes grass hay was suplemented with 200 g/d leucaena plus 150 g/d sucrose (treatment 4). This experiment used a latin square design and variables measured were dry matter, organic matter intake and digestibilities of OM, NDF and N by the method of total collection, N balance, ruminal parameters such as rumen pH and ammonia, and rumen microbial protein synthesis. Differences between means were analysed by the General Linear Model procedure of the Statistical Analysis System. The results showed that although leucaena supplementation increased total DM intake total OM and digestible OM intake did not change. Supplementation increased ammonia-N concentration, N balance and efficiency of microbial N synthesis, especially with urea ($P<0.05$). It can be concluded that leucaena supplementation of 16% of DMI may need additional N source such as urea for better microbial N synthesis, however the need for ruminally fermentable organic matter may not be required unless rumen ammonia-N is not limiting.

Keywords: grass hay, leucaena, intake, digestibilities, sheep

PENGARUH SUPLEMENTASI LAMTORO (*L. LeucocephalaA* cv. tarramba) BAIK DENGAN TAMBAHAN UREA ATAU SUKROSE TERHADAP KONSUMSI DAN KECERNAAN RUMPUT RHODES KERING (*Chloris gayana cv. callide*) OLEH TERNAK DOMBA

ABSTRAK

Penelitian ini dilaksanakan untuk mempelajari konsumsi pakan dan kecernaan dari rumput rhodes kering yang diberikan kepada domba. Empat ekor domba dewasa yang difistula rumennya diperlukan dalam penelitian ini yang dialokasikan ke dalam empat macam perlakuan pakan dengan rancangan penelitian bujur sangkar latin. Pada setiap periode penelitian yang berlangsung selama 21 hari, keempat perlakuan tersebut adalah rumput rhodes diberikan tanpa

tambahan (perlakuan 1), rumput rhodes dengan tambahan 200 g/h daun lamtoro kering (perlakuan 2), rumput rhodes ditambah 200 g/h daun lamtoro kering dan 8 g/h urea (perlakuan 3), dan rumput rhodes dengan tambahan 200 g/h lamtoro kering dan 150 g/h sukrose (perlakuan 4) yang disemprotkan ke dalam rumput kering. Peubah yang diamati adalah konsumsi bahan kering, bahan organik dan kecernaan *in vivo* dari bahan organik, serat detergen netral dan nitrogen, keseimbangan N, konsentrasi ammonia-N, pH rumen, dan efisiensi sintesis protein mikroba rumen. Perbedaan rataan dari setiap peubah dianalisis dengan analisis variansi melalui prosedur *general linear model* dari SAS (Statistical Analysis System). Hasil penelitian menunjukkan bahwa meskipun konsumsi total bahan kering meningkat, total konsumsi bahan organik dan konsumsi bahan organik tercerna tidak berbeda ($P>0.05$). Di pihak lain, suplementasi lamtoro meningkatkan konsentrasi ammonia rumen, keseimbangan N dan efisiensi sintesis mikroba N ($P<0.05$), terutama dengan tambahan urea. Dari penelitian ini dapat disimpulkan bahwa suplementasi lamtoro 16% dari konsumsi bahan kering pakan, memerlukan tambahan urea agar sintesis mikroba rumen berlangsung lebih baik, sedangkan tambahan bahan organik yang terfermentasi dalam rumen mungkin tidak diperlukan kecuali sampai ammonia rumen tidak bersifat sebagai pembatas.

Katakunci: rumput kering. lamtoro, konsumsi pakan, kecernaan, domba

INTRODUCTION.

Many trials have been conducted to test the value of leucaena leaf for ruminants either as a sole diet (Yates, 1983) or as a protein supplement to low and moderate-quality roughages (Moran *et al*., 1983; Bamualim *et al*., 1984; Elliott *et al*, 1985; van Eys *et al*., 1986; Bonsi *et al*., 1995). In most trial it was found that supplemental protein from leucaena leaves promoted high levels of animal production because leucaena was capable of meeting the minimum N requirements for ruminants. Some feed protein in leucaena may escape digestion in the rumen and provide additional protein for absorption in the small intestine (Norton *et al*. 1995; Leng and Devendra 1995). According to these authors protection against digestion in the rumen may be afforded by heat denaturation of protein during drying or by complex formation with tannins during mastication and ruminal metabolism. Ahn *et al*. (1989) reported that protein degradability of dried leucaena leaf incubated in the rumen of sheep for 24 h was lower than that of the freeze dried leaf (73 v 83 %). This suggests that drying protects protein from degradation in the rumen, and consequently the dried leucaena leaf may supply a lower level of ruminally available nitrogen than the freeze dried leaf when used as supplements.

However, major limitations to the use of *Leucaena leucocephala* (Peru, Cunningham, and Hawaii cultivars) include poor tolerance of acid soils, poor adaptation to cool temperatures and frost, and susceptibility to the psyllid insect *(Heteropsylla cubana,* Shelton and Brewbaker 1994). These authors suggested that unless these major limitations are overcome, the great potential for leucaena during the 1970s and 1980s, will not be realised. Since then, interest in the use of other tree species for supplementing low-quality ruminant diets has arisen. A new

Leucaena leucocephala cv Tarramba has been released. This is expected to be more tolerant to the psyllid insect and of cool climates (Castillo *et al.*, 1997) than other lines within the species of leucaena. The nutritive value of the Tarramba cultivar has not been extensively investigated. Apart from the lack of detailed information on Tarramba nutritive value, there are two specific issues which may apply to all leucaena accession: 1). Drying the leaf may result in a lack of ruminally available nitrogen and 2). Supplements from tree foliage are suggested to have a lower metabolisable energy value compared to concentrate (Richards *et al., 1994).*

Therefore, in the present trial, dried-leucaena with or without urea or sucrose was used to supplement low quality rhodes grass hay offered to sheep in an attempt to study voluntary intake and digestibility. This trial was based on an hypothesis that dried-leucaena leaf may be deficient in ruminally available nitrogen, and its utilisation by animals may be increased with an additional energy source such as sucrose. The use of sucrose as a source of energy was based on the findings of Chamberlain *et al. (1993) who* reported that sugar supplements, particularly sucrose are clearly superior to starch as an energy source for the microbial fixation of nitrogen in the rumen of sheep given grass silage.

MATERIALS AND METHODS.

Animals and Dietary Treatments.

Four rumen fistulated crossbred sheep average body weight 38 ± 0.5 kg, were assigned to four dietary treatments in a latin square design. The four dietary treatments were rhodes grass *(Chloris gayana)* hay cv Callide fed alone *ad lib.*(treatment 1), rhodes grass was supplemented with 200 g/d ovendried leucaena (treatment 2), rhodes grass supplemented with 200 g/d leucaena plus 8 g/d urea (treatment 3), and rhodes grass supplemented with 200 g/d leucaena plus 150 g/d sucrose (treatment 4). Leucaena (leaflet and rachis) was harvested from plants which were grown at Beaudesert (west of Brisbane), and then dried (50 ^{0}C for 48 h) in a forced draught oven. Urea and sucrose were dissolved in water and then sprayed on to the hay.

Feeding Routines and Measurements.

Dried leucaena (leaflet and rachis) was chopped to a sufficient length to avoid wastage during feeding. The leucaena supplement was offered at 0800 h and consumed within 30 minutes, followed by the hay in two equal portions at 0830 and 1500 h. The daily allowance of hay was 20 % in excess of its previous day intake. Fresh water and multimineral blocks were also available throughout the trial. Each experimental period consisted of 10 days adaptation and 11 days of collection period. Rumen fluid samples were taken on days 16 and 17 by gentle suction from the ventral sacc of the rumen. About 45 mL was taken 0.5 h before feeding and at 3, 6, 12 h after feeding. Rumen liquor was strained through 6 layers of muslin cloth before measurement of pH then mixed with 0.5 mL of 5 M sulphuric acid and stored at -15 ^{0}C pending ammonia-N determination. Faecal and urine outputs were recorded during 5 day total collections (days 17 to 21). About

10 % of daily faecal outputs were taken and kept at -15 ^{0}C pending chemical analyses. Prior to analysis the 5 day bulked faecal samples were mixed and then sufficient amounts were dried and ground through a 1 mm sieve. For N balance studies, urine was collected into glacial acetic acid and an aliquot of 5 % was sampled and then kept at -15 ^{0}C. Urine for measurement of purine derivatives excretion was collected into 10% sulphuric acid. Five mL of strained urine plus 1.25 mL of allopurinol (internal standard) was diluted with 50 mL 0.1 M $NH_4H_2P0_4$ buffer solution. This sub-sample of urine was kept at -15 ^{0}C pending analysis (Balcells *et al*, 1991). Samples of feed offered, residues, and faeces were ground (1mm screen) prior to analysis.

Chemical Analysis.

Feed offered and refused (hay and leucaena) were analysed for dry matter (DM, 100 ^{0}C for 24 h) and for organic matter (OM, 450 ^{0}C for 4 h). The N content of feed, refusal and urine were determined by the method of Dumas using a FP-2000 nitrogen analyser (Leco, USA). Neutral detergent fibre (NDF), and acid detergent fibre (ADF) contents of feed offered, residues and faecal samples, and acid detergent lignin (ADL) of feed offered were analysed using the Fibertec System (Tecator, Sweden) based on the methods of Goering and van Soest (1970). Acid detergent lignin (ADL) was determined by the 72 % sulphuric acid method of Goering and van Soest (1970). The condensed tannin content of leucaena was assayed by a modified butanol-HCI technique of Perez-Maldonado (1994). Rumen ammonia-N concentrations were determined by steam distillation using MgO and $CaCl_2$ (AOAC, 1984).

Purine derivatives in the urine were estimated by HPLC after filtering the diluted urine through Whatman cellulose nitrate membrane filters (25 mm, 0.2 μ) according to the method of Balcells *et al*. (1991). Rumen microbial purine absorbed from the intestine was estimated from purine derivative excretion in the urine according to Balcells *et al*.(1991), and finally, the rumen microbial N synthesis was estimated using the formula developed by Chen and Gomes (1992). $Y = 0.938X + (3.29\ e^{-0.14x})$, where Y is purine derivatives excreted in the urine and X is microbial purine absorbed, both in mmol/d, and the ruminal microbial N supply is formulated as follows:

$$\text{Rumen microbial N supply} = \frac{X \times 70}{0.83 \times 0.116 \times 1000} = 0.727\ X \text{ g per day}$$

Where X is microbial purine absorbed (mmol/d)
70 is the N content of purine (mg/mmol)
0.83 is the digestibility of purine
11.6 : 100 is the ratio of purine-N : total N in mixed rumen microbes.

Statistical Analysis.

Data were examined by analysis of variance using the general linear model procedure of the Statistical Analysis Systems (SAS, Institute Inc., 1988).

Significant differences between treatment groups were determined by least significant difference and were declared at P< 0.05.

RESULTS

The chemical composition of the basal diet and leucaena supplement is presented in Table1.

Table 1. Nitrogen (N), organic matter (OM), neutral detergent fibre (NDF), acid detergent fibre (ADF) and acid detergent lignin (ADL) contents of the hay and leucaena.

Constituents (% DM)	Hay	Leucaena
DM (%)	94.3	90.2
Nitrogen	1.0	2.8
Organic matter	91.5	86.4
Neutral detergent fibre	73,0	31.3
Acid detergent fibre	39,8	17.5
Acid detergent lignin	7.8	8.9
Condensed tannin	nd	5.0
Solubility(gfkg DM)		
N	24	42
OM	10	33

nd, not determined.

Supplementation did not increase hay dry matter intake, but total dry matter intake was increased by either leucaena, leucaena plus urea or leucaena plus sucrose diet (P<0.05). However, total organic matter and digestible organic matter intake was not affected by supplementation (P>0.05). Further, supplementation did not improve apparent digestibilities of diet constituents, except apparent and true digestibility of N were improved by the leucaena plus urea diet (P<0.05). In contrast, leucaena plus sucrose depressed NDF digestibility compared to the other three diets (P<0.05). Nitrogen intake was significantly increased by supplementation. Although faecal and urinary nitrogen were higher in the supplemented than the control sheep, nitrogen retention was improved by leucaena or leucaena plus urea but not by leucaena plus sucrose diet, compared to the control diet. However, when N retained was expressed as a percentage of N intake the difference among dietary treatments was not significant (Table 2).

Table2. Intake, digestibility, and N balance of sheep offered rhodes grass hay only *ad lib*.(RG), rhodes grass hay supplemented with leucaena (RGL), rhodes grass hay supplemented with leucaena and urea (RGLU), or rhodes grass hay supplemented with leucaena and sucrose (RGLS).

Parameters	**RG alone**	**RGL**	**RGLU**	**RGLS**	**Lsd (P<0.05)**
Ingredient intake (g DM/d)					
Hay	924[a]	912[a]	899[a]	726[a]	311
Leucaena	-	186	186	186	-
Urea	-	-	7	-	-
Sucrose	-	-	-	126	-
Food constituent intake (g/d)					
Total DM	924[a]	1098[c]	1092[c]	1038[b]	49
Total OM	872[a]	1020[a]	1005[a]	940[a]	162
Total DOM	464[a]	542[a]	532[a]	490[a]	85
Digestible constituent contents (g/kg DM)					
OM	532[a]	531[a]	523[a]	502[a]	32
NDF	540[b]	499[b]	486[b]	392[a]	58
N	537[a]	564[a]	659[b]	510[a]	83
N balance (g/d)					
N intake	9.5[a]	14.6[c]	17.6[d]	12.3[b]	1.2
Faecal N	4.4[a]	6.4[b]	6.0[b]	6.1[b]	0.7
Urrinary	1.6[a]	2.2[b]	3.3[c]	1.6[a]	0.2
N retained (g/d)	3.7[a]	6.2[b]	8.3[c]	4.7[ab]	1.5
N retained (% N intake)	38.1[a]	42.4[a]	47.3[a]	39.1[a]	18.3

RG = rhodes grass, L = leucaena, U = urea, S sucrose. "b, **Means within row with dissimilar notations are difference (P<0.05).**

Urinary excretion of purine derivates increased due to leucaena supplementation and this further increased by addition of urea. However, addition of sucrose resulted in similar urinary purine derivatives excretion compared to the leucaena only diet or the control diet (P>0.05). Consequently, microbial N supply to the intestine was increased by leucaena supplementation and urea further increased this parameter compared to the other three diets. When microbial N synthesis was expressed in gram per kilogram digestible organic matter apparently digested in the rumen (DOMR), only the leucaena plus urea diet increased the efficiency of microbial N synthesis (P<0.05, Table 3).

Table 3. Urinary excretion of purine derivatives (PD), microbial N supply and efficiency of microbial N synthesis of sheep offered rhodes grass hay only *ad lib.* (RG), rhodes grass hay supplemented with leucaena (RGL), rhodes grass hay supplemented with leucaena and urea (RGLU), or rhodes grass hay supplemented with leucaena and sucrose (RGLS).

Parameters	RG alone	RGL	RGLU	RGLS	Lsd (P<0.05)
Urinary excretion of PD (mmol/d)	13.5[a]	17.6[b]	21.1[c]	15.9[ab]	2.5
Microbial N supply (g/d)	10.1[a]	13.4[b]	16.3[c]	12.1[b]	1.9
Efficiency of microbial N synthesis (g/kg DOMR)[1]	33.6[a]	38.4 [a]	48.1[b]	37.9[a]	8.2

RG = rhodes grass, L = leucaena, U = urea, S = sucrose, DOMR[1] = digestible organic matter apparently digested in the rumen = 0.65 DOMR (SCA, 1990).

Rumen ammonia-N concentration from rumen fluid sampled at 3, 6 and 12 h after feeding was increased by addition of urea and reduced by sucrose supplementation. On the other hand, rumen pH was higher on leucaena plus sucrose diet when samples of rumen fluid were taken at 12 h after and half an hour before feeding on the following day, and over the 24 h compared to control diet (Table 4).

Table 4. Rumen ammonia-N concentration and pH of sheep offered rhodes grass hay only *ad lib*.(RG), supplemented with leucaena (RGL), with leucaena and urea (RGLU), or with leucaena and sucrose (RGLS).

	Treatments				
Time after feeding (h)	RG alone	RGL	RGLU	RGLS	Lsd (P<0.05)
Rumen ammonia-N (mg/L)					
3 (11:00)	56^{ab}	74^{b}	178^{c}	15^{a}	42
6 (14:00)	26^{ab}	45^{b}	134^{c}	8^{a}	21
12 (20:00)	10^{a}	16^{a}	98^{b}	7^{a}	10
24 (08:00)	29^{a}	62^{b}	94^{c}	9^{a}	24
over a 24 h period[1]	30^{b}	49^{c}	126^{d}	12^{a}	12
Rumen pH					
3 (11:00)	6.25^{a}	6.48^{c}	6.48^{c}	6.38^{b}	0.10
6 (14:00)	6.15^{a}	6.33^{b}	6.45^{b}	6.40^{ab}	0.16
12 (20:00)	6.00^{a}	6.18^{a}	6.15^{a}	6.38^{b}	0.18
24 (08:00)	6.53^{a}	6.65^{ab}	6.68^{b}	6.73^{b}	0.15
over a 24 h period[1]	6.23^{a}	6.41^{b}	6.44^{b}	6.47^{b}	0.08

RG = rhodes grass, L = leucaena, U = urea, S = sucrose.Means within rows with different superscripts are significantly different (P<0.05). 1, unweighted means of individual-sampling. Feeding time was at 08:00 h.

DISCUSSION

The lack of response in terms of increasing the voluntary intake of low quality grass hay or the total diet digestibility, due to leucaena supplementation, may be related to the level of protein of the basal diet, or to the degradability of leucaena protein in the rumen, and/or to the level of supplement offered. Supplementation with legumes could be expected to increase intake of basal diet when it contains less than 20 g N/kg DOM (Egan 1986). In the work reported here the N content of the basal diet was close to this value (19.5 v 20 g N/kg DOM). The increase intake of alkali-treated rice straw (4% protein, DM basis) by up to 30% by both Ongole cattle and buffaloes has been reported by Moran *et al*. 1983. Doyle (1989)reported a linear increase in total OM intake by sheep fed rice straw basal diets (3.6% protein, DM basis) supplemented with 7 to 21% dry leucaena. In contrast, Van Eys (1986) observed lower intake of napier grass (12% protein, DM basis) by goats supplemented with gliricidia or leucaena at 15% of the dry matter intake of napier grass than that of controls receiving no legume supplement. Although total dry matter intake, intake of cell wall constituents or digestibility in

Van Eys's trial did not change due to supplementation, average daily gain for control goats was reported -1g/day as compared with 21g/day for supplemented goats. The characteristics of low quality roughages such as their physical nature, susceptibility to microbial digestion and nutrient content, which are important in intake regulation, also play a role in determining the nature of responses to supplementation (Doyle, 1989).

Although it is suggested that the responses obtained in the present work were largely influenced by the protein content of the basal diet, it is also possible that they may be attributed to the level of leucaena supplementation. The level of leucaena supplementation in this trial was 186g DM/d (16% total DM intake), whereas Norton *et* al. (1995) have suggested that at least 30 to 50% leucaena in the diet (or equivalent to 0.8 to 1.2% body weight daily) is required for optimum performance of cattle, sheep and goats offered low quality basal diets. Alternatively, the nature of the supplement itself (degradability, states of hydration, etc) may be also important in determining the type of responses to supplementation. For example, Ash (1990) demonstrated an increase in N retention by goats offered guinea grass hay supplemented with gliricidia or sesbania leaf, but not with albizia leaf. Albizia was reported to have a lower protein degradability in the rumen compared to gliricidia and sesbania. Bamualim *et al.* (1984) reported that dry leucaena supplement given to sheep fed a low quality basal hay did not change intake of the hay. When fresh leucaena was fed to goats given the similar quality of hay, intake of the hay increased compared to the control diet.

Obara *et al.* (1994) and Sutoh *et al.* (1996) have reported that positive responses to sucrose supplementation of sheep given lucerne, and improvements in animals given concentrate with fodder tree-supplemented diets have been reported (eg. Richards *et al.,* 1994). However, in this experiment sucrose supplementation reduced NDF digestibility, contrasting to the observations of Obara *et al.* (1994) and Sutoh *et al.* (1996) who reported no change in NDF digestibility by sheep offered lucerne hay cubes supplemented with sucrose at 14-15% of DM intake. The cause of this difference is not known but may be associated with differences in the cell wall composition of the basal diet used (a tropical grass v a legume). However, similar decreases to those reported here in NDF and ADF digestibilities were observed by Khalili and Huhtanen (1991) in cattle fed grass silage supplemented with sucrose at 1.0 kg per day, whether it was fed twice daily or as a continuous intraruminal infusion. Clearly, in the present trial the reduction in NDF digestibility was not associated with lower rumen pH due to addition of readily fermentable carbohydrate since rumen pH on the sucrose supplemented diet was higher than the control and leucaena alone diets.

Reduction in fibre digestion associated with the fermentation of readily soluble carbohydrate is not caused by lower rumen pH alone, but rather many factors may be involved (Mould *et al*., 1983). Feeding easily fermentable carbohydrates depresses forage DM degradation and this may be not alleviated by decreasing rumen pH. This concept is also supported by the adverse effect of sugar infusion in cattle given silage-based diets (Rooke *et al*., 1987), although rumen pH was maintained at the high level (6.8 and 6.7). Alternatively, the decrease in NDF digestibility in this trial with addition of sucrose to the leucaena-supplemented diet may be associated with lower activity of cellulolytic

bacteria as rumen ammonia-N concentration on this diet was lower than the other three treatments. A lower rumen ammonia-N concentration with sucrose supplemented than unsupplemented diets has also been reported by Chamberlain *et al.* (1993) in sheep fed grass silage *ad lib.* leading to more efficient in using N in the silage for microbial protein synthesis.

Rumen ammonia-N concentration was increased by leucaena supplementation to the level which is thought to be sufficient for fibre digestion (50 mg NH_3-N/L) in *vitro* (Satter and Slyter, 1974). Addition of urea further increased rumen ammonia-N concentration to the level of 126 mg/L. Sucrose supplementation at 14-15% of DM intake has also been reported to reduce rumen ammonia-N concentration in sheep offered lucerne hay cubes compared to the control diet (172 vs 224 mg/L, Sutoh *et al*., 1996; 173 vs 274 mg/L, Obara *et al*., 1994). The low rumen ammonia-N concentration due to addition of sucrose was not indicative of more efficient capture of ammonia-N by the rumen microbes in the presence of an additional energy source such as sucrose. This was judged by no increase in microbial protein supply on this diet when compared to the other three diets. The low level of rumen ammonia-N concentration may be associated with two possible reasons: Firstly, drying the leaf may insolubilise protein in the leaf, and consequently reduce ammonia-N concentrations. Secondly, it may be associated with the present of tannins in leucaena.

In practice, the microbial crude protein yield is unlikely to exceed about 300 g (48g N. kg DOMR-', SCA 1990). In this trial, the highest efficiency of microbial crude protein synthesis was achieved when grass hay was supplemented with leucaena. plus urea, and the magnitude was close to the figure adopted by SCA (1990). Many factors have been known to influence the efficiency of microbial N synthesis in ruminants these are the amount of N and other nutrients recycled into the rumen, the supply and nature of energy-yielding substrates available to the microorganisms and the fractional outflow rates from the reticulo-rumen. A similar efficiency of microbial N synthesis (44.8g N. kg $DOMR^{-1}$) was reported by Doyle (1989) in sheep offered rice straw *ad lib.* supplemented with 15% dry leucaena. McSweeney *et al.* (1998) reported a lower efficiency of microbial N synthesis of 28.6g N. kg $DOMR^{-1}$ in sheep offered buffel grass hay *ad libitum* supplemented with 30% calliandra, with or without addition of 40g polyethylene glycol 4000/d.

On the other hand, the efficiency of microbial N supply of the unsupplemented sheep offered rhodes grass hay *ad libitum* in this trial was higher than the value reported by Bolam *et al.* (1998) in cattle fed the same quality rhodes grass hay *ad libitum* (23 v 19g N. kg $DOMR^{-1}$). In contrast, Bamualim *et al.* (1984) have reported a higher efficiency of microbial N synthesis in goats offered spear grass hay alone *ad libitum* than that was observed in this trial (31.4 v 23g N. kg $DOMR^{-1}$). Likewise, McSweeney *et al.* (1998) have reported a higher efficiency of microbial N synthesis in sheep offered buffel grass hay alone *ad libitum* (29.3 v 23g N. kg $DOMR^{-1}$). Again, the difference in the efficiency of microbial N supply may be related to the many factors as described earlier.

Leucaena supplementation increased the N retention by the sheep, similar to the observation of Moran *et al. (1983)* in Ongole cattle or buffaloes offered rice straw basal diets supplemented with leucaena. Similarly, N retention was increased by animals fed low quality diets supplemented with fodder trees such as

leucaena (Bonsi *et al.* 1995), sesbania (Ash, 1990), *Cratylia argentea* alone or C. *argentea with Flemingia macrophylla* (Fassler and Lascano, 1995). Addition of urea to the leucaena supplemented diet increased N retained by 43% compared to the leucaena diet alone, suggesting improved N utilisation by the sheep when urea was provided along with leucaena. This was probably because of the increased efficiency of microbial N synthesis on this diet compared to other treatments.

However, leucaena supplementation has been associated with an increase in the faecal NDF-N. This may be associated with the presence of condensed tannin in leucaena-supplemented diets. Tannins in fodder trees have been reported to increase NDF-N during its passage through the digestive tract (Reed *et al.*, 1990; Fassler and Lascano, 1995; Degen *et al.*, 1997).

CONCLUSION

The implications from the results of this trial are that with supplementation of about 16% of DMI of dry leucaena, the need for additional ruminally available N source, is necessary for better microbial N synthesis. This is evident from the result of another trial that when leucaena supplementaton increased to 25% of DMI addition of urea did not improve the efficiency of microbial N supply measured in g per kg DOMR over urea-unsupplemented diet (Karda and Dryden, 2001). It also appears that ruminally fermentable organic matter may not be required unless rumen ammonia-N concentrations are not limiting.

ACKNOWLEDGEMENT

I would like to thank firstly my supervisor, Dr. Gordon Dryden for his guidance during the course of my study and also to Ms Kaye Vockenson, Mr Felix Gorbacz, Mr Tony Chen, Mr Bob Engelbright and Mr Andy Goodwin of the School of Veterinary Science and Animal Production for their guidance in the laboratory analyses. Secondly, my gratitude is due to Mr Alan Lisle for his guidance in the Statistical Analysis, Dr. Ross Gutteridge and Mr James Hales for providing gliricidia leaves used in the trials. I also express my gratitude to Professor John Ternouth for fistulating the sheep used in these trial. Lastly, I would like to acknowledge AusAID for providing financial supports required by this study and our Rector Professor dr Dr Mulyanto who gave me permission to take this study.

REFERENCES

Ahn, J-H., Robertson, B. M., Elliott, R., Gutteridge, R. C., and Ford, C. W. 1989. Quality assessment of tropical brawse legumes: tannin contents and protein degradation. *Anim. Feed Sci. and Technol*., **27**: 147.

Ash, A. J. 1990. The effect of supplementation with leaves from the leguminous trees Sesbania grandiflora, Albizia chinensis and Gliricidia sepium on the intake and digestibility of Guinea grass hay by goats. *Anim. Feed Sci. and Technol.*, **28**: 225-232.

Balcells, J., Guada, J. A., Castrillo, C., and Gasa, J. 1991. Urinary excretion of allantoin and allantoin precursors by sheep after different rates of purine infusion into the duodenum. *J. Agric. Sci. Camb*., **116**: 309.

Bamualim, A., Weston, R. H., Hogan, J. P., and Murry, R. M. 1984. The contribution of *Leucaena leucocephala* to post ruminal digestible protein for sheep fed tropical pasture hay supplemented with urea and mineral. *Proc. Aust. Soc. Anim. Prod*., **15**: 255.

Bamualim, A., Stachiw, S., Jones, R. J., and Murry, R. M. 1984. The effect of fresh leucaena as a supplement on the utilisation of pasture hay by goats. *Proc. Aust. Soc. Anim. Prod*., **15**: 259.

Bolam. M. J., Connors, M. T., McLenan, S. R., and Poppi, D. P. 1998. Variability in microbial protein supply under different supplementation strategies. *Proc. Aust. Soc. Anim. Prod*., 22: 389.

Bonsi, M. K. L., Osuji, P. O., and Tuah, A. K. 1995. Effect of supplementing teff straw with different levels of leucaena or sesbania leaves on the degradabilities of teff straw, sesbania, leucaena, tagasaste and vernonia and on certain rumen and blood metabolites in Ethiopian Menz sheep. *Anim. Feed Sci. and Technol*.,**52**: 101.

Castillo, A. C., Cuyugan, O. C., Fogarty, S., and Shelton, H. M. 1997. Growth, psyllid resistance and forage quality of *Leucaena laucocephala*, *L. pallida*, *L. diversifolia*, and the hybrid of *L. leucocephala* x *L. pallida*. Trop. Grassland, **31**: 188.

Chamberlain, D. G., Robertson, S., and Choung, Jai-Jun. 1993. Sugars versus starch as supplement to grass silage: Effects on rumen fermentation and the supply of microbial protein to the small intestine, estimated from the urinary excretion of purine derivatives in sheep. *J. Sci. Food Agric*., **63**: 189.

Chen, X. B., and Gomes, M. J. 1992. Estimation of microbial protein supply to sheep and cattle based on urinary excretion of purine derivatives. Occasional publication 1992. Rowett Research Institute, Bucksburn, Aberdeen, AB2 9 SB, UK.

Degen, A. A., Blanke, A., Becker, K., Kam, M., Benyamin, R. W., and Makkar, H. P. S. 1997. The nutritive value of *Acacia saligna* and *A. salicina* for goats and sheep. *J. Anim. Sci*., **64**: 253.

Doyle, P. T. 1989. Supplementation of rice straw with dry leucaena. Aust. *J. Agric. Res*., **40**: 381.

Egan, A. R. 1986. Principle of supplementation of poor quality roughages with nitrogen. In: R. M. Dixon (ed). Ruminant feeding system utilising fibrous agricultural residues-1985. IDP of Australian Universities and Colleges, Canberra, pp. 49-57.

Elliott, R., Norton, B. W., Milton, J. T. B., and Ford, C. W. 1985. Effect of molasses on mimosine metabolism in goats fed fresh and dried leucaena with barley straw. Aust. J. *Agric. Res*., **36**: 867.

Fassler, O. M., aand Lascano, C. E. 1995. The effect of mixtures of sun-dried tropical shrub legumes on intake and nitrogen balance by sheep. Trop. Grassland, **29**: 92.

Goering, H. K., and Van Soest, P. J. 1970. Forage fibre analysis. USDA Agriculttural Handbook No. 379, USDA, Washington, DC, pp. 1-20.

Karda, I W., and Dryden, G. McL. 2001. Effect of leucaena supplementation levels, and provision of urea, on utilisation by sheep of Rhodes grass hay. Aust. *J. Exp. Agric*., **41**: 1155.

Khalili, H., and Huhtanen, P. 1991. Sucrose supplementation in cattle given grass silage-based diet. 2. Digestion of cell wall carbohydrate. *Anim. Feed. Sci. and Technol*., **33**: 263.

Leng, R. A., and Devendra, C. 1995. Research priorities to improve livestock productin in Asia. In: Rowe, J. B. and Nolan, J. V. (eds). Recent advances in animal nurition in Australia 1995. University of New England. Armidale NSW 2351, Australia, pp. 160-171.

McSweeney, C. S., Palmer, B., Kennedy, P. M., and Krause, D. 1998. Effects of caliandra tannin on rumen microbial function. *Proc. Aust. Soc. Anim. Prod*., **22**: 289.

Moran, J. B., Satoto, K. B., and Dawson, J. E. 1983. The utilisation of rice straw fed to Zebu cattle and Swamp buffalo as influenced by alkali treatment and leucaena supplementation. Aust. *J. Agric. Res*., **34**: 73.

Mould, F.L., Orskov, E. R., and Mann, S. O.1983. Associative effects of mixed feeds. I. Effects of type and level of supplementation and the influence of the rumen fluid pH on cellulosis in vivo and dry matter digestion of variousroughages. *Anim.Feed Sci. and Technol*., 10: 15-30.

Norton, B. W., Lowry,J. B., and McSweeney, C. 1995. The nutritive value of leucaena species. In: H.M. Shelton, C.M. Piggin and J. L. Brewbaker (eds). Leucaena-Opportunities and Limitations. Proc.of a workshop held in Bogor, Indonesia, 24-29 Januari 1994. pp. 103-111.

Obara, Y., Fuse, H., Terada, F., Shibata, M., Kawabata, A., Sutoh, M., Hodate, K., and Matsumoto, M. 1994. Influeence of sucrose supplementation on nitrogen kinetics and energy metabolism in sheep fed with lucerne hay cubes. *J. Agric. Sci. Camb*., **123**: 121.

Perez-Maldonado, R. A. 1994. The chemical nature and biological activity of tannins in forage legumes fed to sheep and goats. PhD thesis. The University of Queensland, Australia.

Reed, J. D., Soller, H., and Woodward, A. 1990. Fodder tree and straw diets for sheep. Intake, growth, and the effect of phenolics on nitrogen utilisation. *Anim. Feed Sci. and Technol*., **30**: 39.

Richards, D. E., Brown, W. F., Ruegsegger, G., and Bates, D. B. 1994. Replacement value of tree legumes for concentrates in forage-based diets. I. Replacemen value of *Gliricidia sepium* for growing goats. *Anim. Feed Sci. and Techno*l., **46**: 37.

Rooke, J. A., Lee, N. H., and Armstrong, D. G. 1987. The effect of intra ruminal infusion of urea, casein, glucose syrup and a mixture of casein and glucose syrup on nitrogen digestion in the rumen of cattle receiving grass silage diets. Br. *J. Nut*., **57**: 89.

SAS. 1988. SAS procedures guide (version 6.03 edition). SAS Institute Inc., Cary, NC., USA.

Satter, L. D., and Slyter, L. L. 1974. Effects of ammonia concentration on rumen microbial production *in vitro*. Br. *J. Nut*., **32**: 199.

SCA 1990. Feeding standard for Australian livestock. Ruminants. Standing Committee on Agrculture, CSIRO, Australia, pp. 129.

Sheelton, H. M., Lowry, J. B., Gutteridge, R. C., Bray, R. A., Wildin, J. H. 1991. Sustaining productive pasture in the tropics. 7 Tree and shrub legumes in improved pastures. Trop. Grassland, **25**: 119.

Sutoh, M., Obara, Y., and Miyamoto, S. 1996. The effect of sucrose supplementation on kinetics of nitrogen, ruminal propionate and plasma glucose in sheep. *J. Agric. Sci. Camb*., **126**: 99.

Van Eys, J. E., Mathius, I W., Pangsapasn, P., and Johnson, W. L. 111986. Foliage of tree legumes gliricidia, leucaena, and sesbania as supplement to napier grass diets for growing goats. *J. Agric. Sci. Camb*., **107**: 227.

Yates, N. G. 1983. The effect of a salt and trace element supplement on the growth of Javanese sheep offered *Leucaena leucocephala*. *Trop. Anim. Prod*. 50-52.