Tropical Animal Production 1984 9:299-310

MINIMUM RUMEN AMMONIA REQUIREMENTS FOR RUMEN DIGESTION OF NaOH TREATED MAIZE COBS AND *Pennisetum purpureum*

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Five sheep were given mature chopped *Pennisetum purpureum* forage *ad libitum* and each of four supplements based on NaOH treated maize cobs with no additional nitrogen (control), with 2% or 4% urea or with 22% soyabean meal. Measurements were made of rumen pH, rumen NH_3-N concentration and of the DM digestion in nylon bags incubated in the rumen of *Pennisetum purpureum* forage and of NaOH treated maize cobs. Maximum digestion of *Pennisetum purpureum* forage occurred at a rumen NH_3-N concentration between 32 and 90 mg NH_3-N/l, whereas the minimum NH_3-N concentration for maximum DM digestion of NaOH treated maize cobs was greater than 133 mg NH_3-N/l. Furthermore, when there was a deficiency of NH_3-N for microbial synthesis, the digestion of insoluble DM of NaOH treated maize cob was depressed more (from 36% to 16%) than that of *Pennisetum purpureum* forage (from 28% to 23%). In a second experiment, when four sheep were given the mature *Pennisetum purpureum* forage alone, the rumen NH_3-N was unlikely to be limiting for microbial activity.

Key words: Sheep, rumen ammonia, rumen digestion

The nitrogen (N) requirements of ruminants may be considered in terms of rumen fermentable N for microbial growth, and rumen undegraded dietary protein which is not fermented in the rumen and is digested in and absorbed from the small intestine (ARC, 1980). Since rumen fermentable N can be provided in diets at low cost in the form of non-protein N (NPN), it will usually be desirable to provide sufficient rumen fermentable N to maximise microbial growth and dry matter (DM) digestion. Furthermore, since NH_3-N is the principal microbial N substrate, an understanding of the microbial requirements for NH_3-N in the rumen is essential to a detailed understanding of dietary requirements for fermentable N.

The minimum rumen NH_3-N concentration required for maximum production of microbial N per unit of DM fermented has been estimated *in vitro* to be 30-50 mg NH_3-N/l (Allison, 1970; Satter and Roffler, 1977). In *in vivo* experiments Pisulewski *et al.* (1981) found that this NH_3-N requirement ranged from 22 to 84 mg NH_3-N/l depending on the diet. However, as Mehrez *et al.* (1977) have pointed out, the minimum ruminal NH_3-N concentration for maximum rate of DM concentration may be greater than the minimum concentration required for maximum microbial N synthesis per unit of DM fermented. In sheep fed barley grain the maximum rate of DM fermentation did not occur until the rumen NH_3-N concentration reached 200-250 mg NH_3-H/l.

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The present experiments were undertaken to examine the minimum NH_3-N concentration for maximum rate of rumen fermentation of two fibrous feedstuffs (NaOH treated maize cobs and *Pennisetum purpureum* forage), and also to determine whether rumen fermentable N was adequate when sheep were given mature chopped *Pennisetum purpureum* forage alone.

Materials and Methods

Animals and Management:

Experiment 1 : Five mature West African sheep (27-37 kg liveweight) were prepared with rumen cannulas (50 mm diameter) and held in metabolism crates in an open-sided shed. The sheep were given mature (60-65 d regrowth) *Pennisetum purpureum* forage at 120-130 % of expected feed intake. During 4 periods each of 14 days the sheep received 450 g/d (airdry) of each of the supplements given in Table 1. The supplements consisted of NaOH treated maize cobs with the addition of 0% (control), 2% of 4% urea or 22% soyabean meal. A changeover experimental design was used such that one diet was repeated within each period. The supplements and forage were given in separate feeders at 0830 h each day.

Table 1:
Composition of the 4 supplements used in Experiment 1.

	Supplement			
	Control	Urea A	Urea B	Soya
Composition (air-dry basis)				
NaOH (5%) treated ground maize cobs	93	91	89	71
Molasses	5	5	5	5
Soyabean meal	0	0	0	22
Urea	0	2	4	0
Minerals*	2	2	2	2
(Nx6.25) content (DM basis) %**	2.1	7.8	13.5	11.6

* Principally dicalcium phosphate.

** Calculated from the composition of constituents.

Experiment 2 : Four sheep, including three used in Experiment 1, were given the *Pennisetum purpureum* forage *ad libitum* with minerals (10 g/d) and salt (5 g/d) sprinkled over the forage.

Measurements :

In each experiment daily measurements were made of forage and supplement DM offered and refused.

On day 12 of each period rumen digesta were sampled at 0800 h, 1100 h, 1400 h, 1700 h, 2000 h and 0800 h using a plastic tube 35 mm in diameter which could be occluded at the lower end with a conical stopper. The pH was determined immediately, and the digesta were then filtered through cloth to separate rumen liquid which was acidified (pH$\leq$ 4, 5M H_2SO_4) and stored at 5°.

The NH_3-N in rumen liquid was released by steam distillation under alkaline conditions, collected into boric acid (2% w/v) and titrated with 0.005 M H_2SO_4.

In Experiment 1 the DM digestibilities in the rumen of dried *Pennisetum purpureum* forage and of NaOH (5%) treated maize cobs were determined using duplicate nylon bags (cloth 12μ m pore size, nylon filter cloth HS 013, Henry Simon Ltd., Cheshire, U.K.) incubated in the rumen for 9 h and 24 h (Ørskov *et al*, 1980). Nylon bags containing each feedstuff were also soaked in 0.15 M NaCl for 3 h before washing to determine the respective proportions of soluble DM; digestion in the rumen of insoluble DM could therefore be calculated.

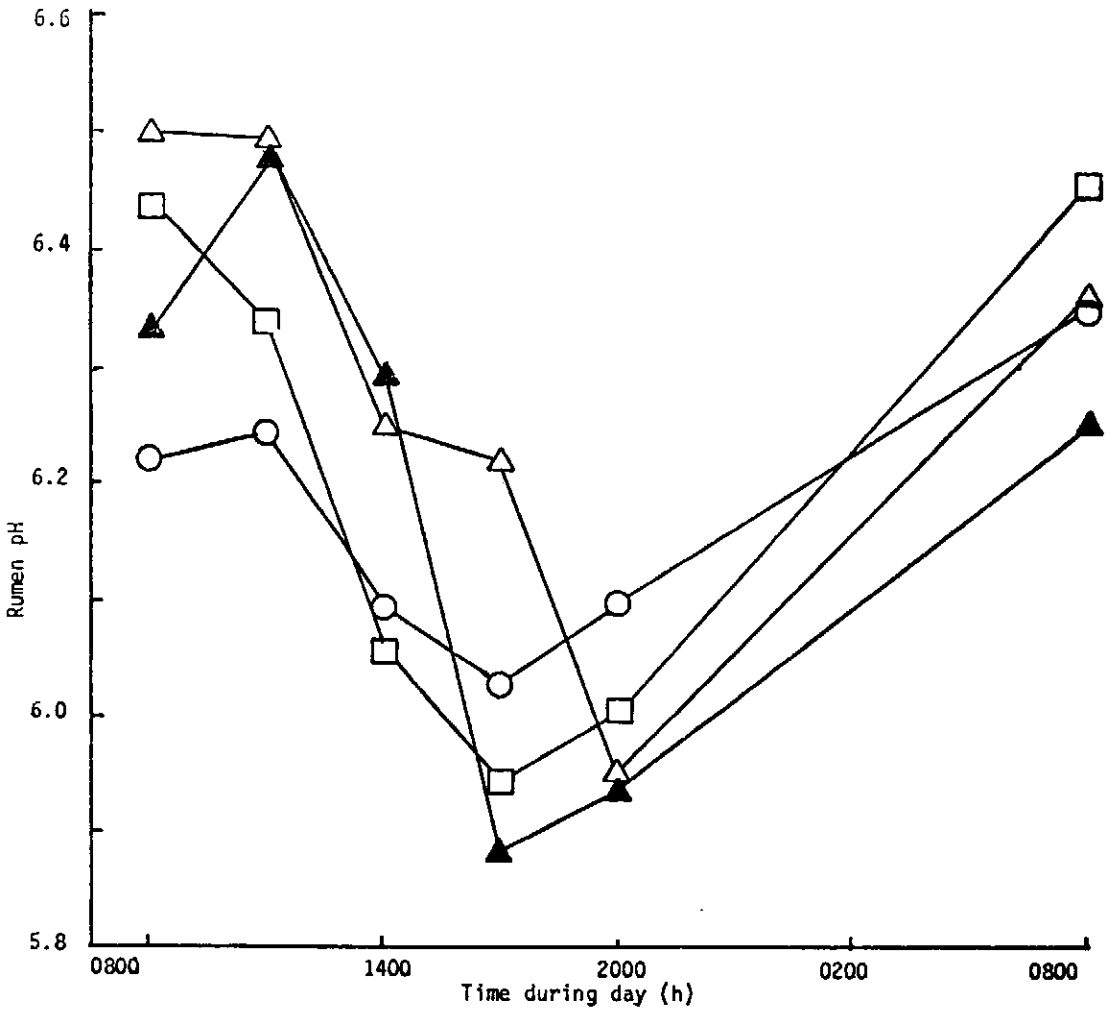
Results

Experiment 1 : The intake of forage and supplements were not different ($P > 0.05$) among treatments, although there was a tendency for the intake of forage to be lower in sheep given the control supplement than in those given the supplements containing urea or soyabean meal (Table 2).

There were no differences among the supplements in the mean daily pH (pH 6.2-6.3; Table 2). For all supplements there was a depression in rumen pH 9-12 h after feeding (Figure 1).

In sheep given the control supplement the mean ruminal NH_3-N concentration was 32 mg N/l (Table 2) and this concentration did not vary appreciably throughout the day (Figure 2). The rumen NH_3-N concentration was increased ($P < 0.01$) by the supplements containing urea or soyabean meal to 90-133 mg N/l, although there were no differences ($P > 0.05$) in NH_3-N concentrations among these latter supplements (Table 2). In the sheep given supplements containing urea there was a peak in the rumen NH_3-N concentration shortly after feeding followed by a decline in concentration to pre-feeding levels or less (Figure 2). In the sheep given the soyabean meal supplement the rumen NH_3-N concentration tended to decline 6-12 h after consuming the supplement, and then to increase to pre-feeding levels.

Figure 1:
Experiment 1: Rumen pH in sheep given at 0830 h Pennisetum purpureum *forage ad libitum plus 450 g/d of supplement of NaOH treated maize cobs with no added N (0), 2% urea (△), 4% urea (▲) or 22% soyabean meal (□)*



The proximal analysis of the two feedstuffs incubated in the nylon bags are given in Table 3. The solubilities of DM in 0.15 M NaCl were 24.7% and 24.3% for the NaOH treated maize cobs and the *Pennisetum purpureum* forage respectively.

The DM digestibility of the NaOH treated maize cobs after both 9 h and 24 h incubation was greater ($P < 0.01$) in sheep given the supplements

Table 2:
Experiment 1: Intake and measurements of rumen fermentation and digestion in sheep given Pennisetum purpureum forage ad libitum and 450 g/d of each of four supplements (n = 5).

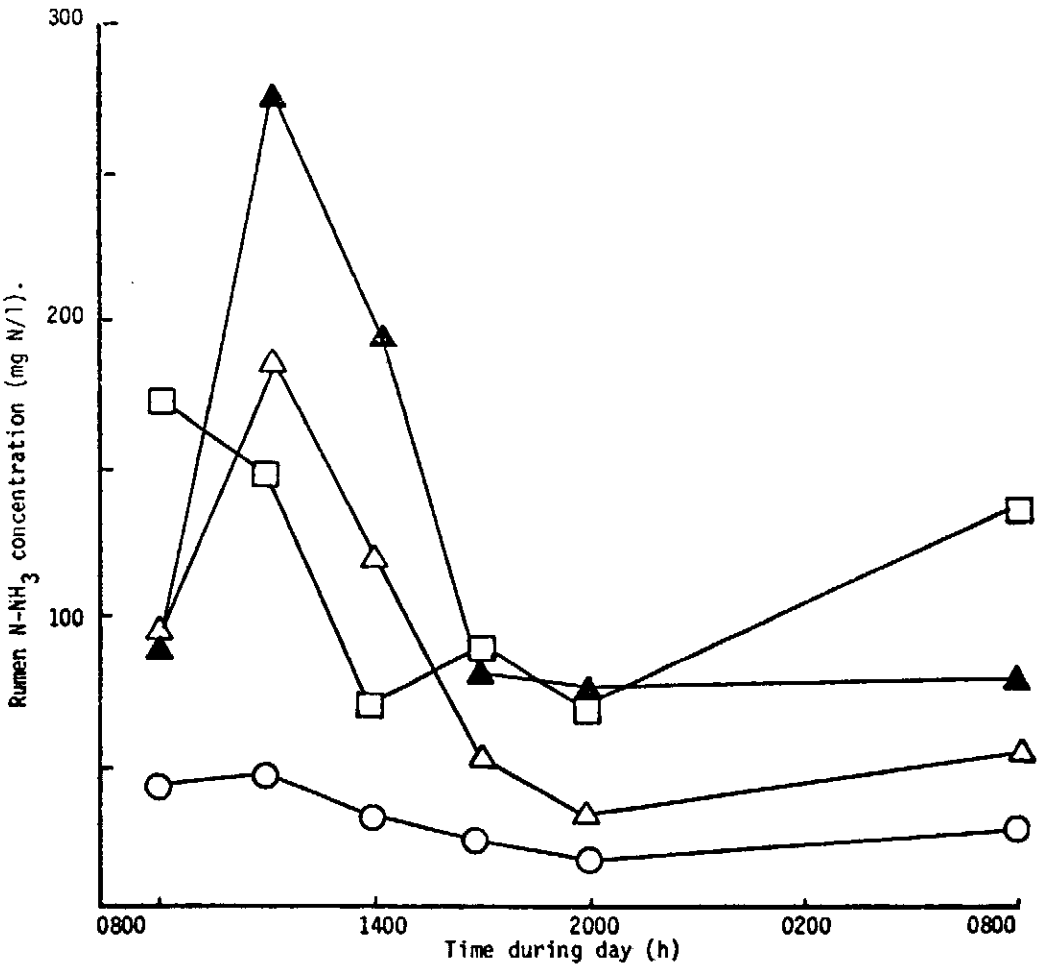
	Control	Urea A	Urea B	Soya	Sx	Prob.
Intake (g DM/d)						
Forage	414	526	558	584	43	NS
Supplement	316	378	298	370	35	NS
Total	730	904	856	954	68	NS
Rumen measurements						
pH	6.2	6.3	6.2	6.3	0.07	NS
NH_3-N (mg N/l)	32	90	133	114	21.4	**
Nylon bag digestion						
Maize cobs+ 9 h	28	33	32	33	0.9	**
24 h	36	48	54	51	2.1	**
Forage 9 h	26	31	31	28	0.8	**
24 h	42	47	46	45	.19	NS

+ Maize cobs ground and treated with 5% NaOH.

containing urea or soyabean meal than for the sheep given the control supplement (Table 2); however there were no differences ($P > 0.05$) among the supplements containing the urea or soyabean. There was a similar tendency for the DM digestibility of the *Pennisetum purpureum* forage to be greater for the supplements containing the additional N (Table 2), although this was only significant ($P < 0.01$) for the 9 h incubation.

The relationship between the rumen NH_3-N concentration and the DM digestibility of each of the feedstuffs are shown in Figures 3 and 4. It was considered that an appropriate model for this relationship was that there should be a progressive increase in DM digestibility until the NH_3-N requirements of the rumen microbes are reached, but no further increase in DM digestion with further increases in rumen NH_3-N concentration (Mehrez *et al.*, 1977). For *Pennisetum purpureum* forage it appeared that rumen DM digestion increased as rumen NH_3-N concentration increased from 32 to 90 mg NH_3-N/l, after which there was no further increase in rumen DM digestibility. Therefore the minimum NH_3-N concentration for maximum rate of DM digestion of this forage was between 32 and 90 mg NH_3-N/l. The DM digestion of the NaOH treated maize cobs apparently increased linearly from 32 to 133 mg NH_3-N/l (Figure 4), and the minimum NH_3-N concentration for maximum rate of DM digestion of this feedstuff was therefore in excess of 133 mg N/l.

Figure 2:
Experiment 1: Rumen N-NH$_3$ concentration in sheep given at 0830 h Pennisetum purpureum *forage ad libitum plus 450 g/d of supplement of NaOH treated maize cobs with no added N (0), 2% urea (△), 4% urea (▲) or 22% soyabean meal (□)*



Experiment 2: The voluntary intake of the forage alone was 683 ± SE 24 g/d of DM.

The variations throughout the day of the ruminal pH and NH_3-N concentration are shown in Figure 5. The pattern of rumen pH was similar to that observed in Experiment 1, but the mean pH (pH 6.8 ± SE 0.1) was higher than that in Experiment 1 (pH 6.2 - 6.3; Table 2).

Mean rumen NH_3-N concentration over 24 h was 121 ± 14 mg NH_3-N/l although the concentration was less than 90 mg NH_3-N/l for approximately 7 h (Figure 5).

Figure 3:
Experiment 1: The relationship between mean rumen N-NH_3 concentration and the digestion in nylon bags for 9 h (△) or 24 h (▲) of Pennisetum purpureum *forage*

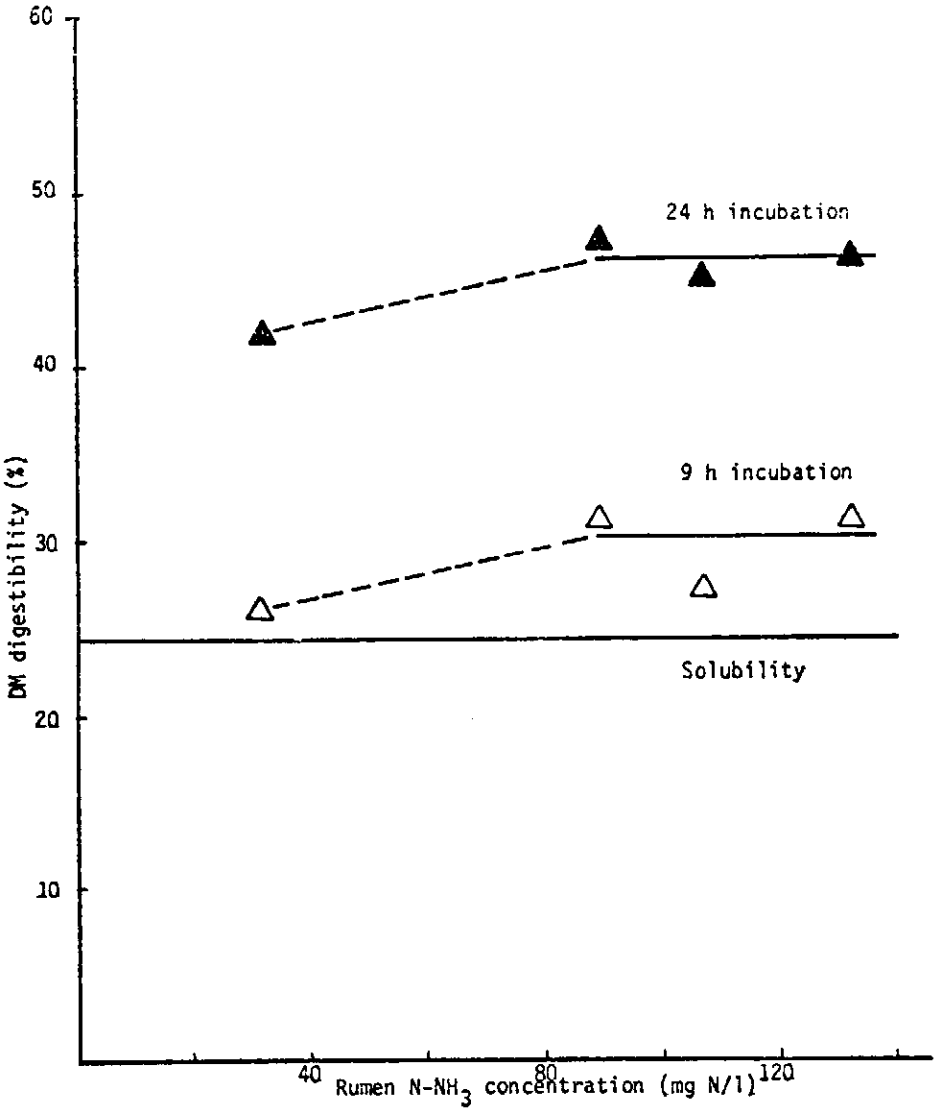


Table 3:
Proximal analysis of the forage offered and refused in Experiment 2, and the feedstuffs incubated in the nylon bags in Experiment 1.

	Experiment 2		In nylon bags - Experiment 1	
	Forage offered	Forage refused	NaOH treated maize cobs	*Pennisetum purpureum*
Dry matter	-	-	93.3	87.2
Organic matter	91.2	92.5	92.3	88.0
N x 6.25	6.8	6.5	2.3	9.4
Neutral detergent fibre	78.2	80.8	79.4	71.8
Acid detergent fibre	-	-	49.0	48.7
Lignin	-	-	10.0	10.3

Discussion

Experiment 1 clearly demonstrated that there were differences between the two feedstuffs examined in the minimum rumen ammonia concentrations required for maximum DM digestion. The minimum concentration for digestion of *Pennisetum purpureum* forage, between 32 and 90 mg NH_3-N/l, was within the range observed for maximum production of microbial N per unit of DM fermented (Allison, 1970; Satter and Roffler, 1977). The much greater rumen NH_3-N concentration (in excess of 133 mg NH_3-N/l) required for digestion of the NaOH treated maize cobs is in agreement with the results of Mehrez *et al.* (1977) which indicated that 200-250 mg NH_3-N/l was required for maximum rate of digestion of barley grain. Furthermore a deficiency of rumen NH_3-N reduced the rumen digestion of insoluble DM much more for the NaOH treated maize cobs (from 36% to 16%) than for the *Pennisetum purpureum* forage (from 28% to 23%). These differences between feedstuffs may have been associated with the much lower N content of the NaOH treated maize cobs (0.4% N) than of the *Pennisetum purpureum* forage (1.5% N) per unit of fermentable DM. Microorganisms closely associated with particulate matter during the digestion process (Cheng and Costerton, 1980) could presumably obtain at least part of their N substrate from the *Pennisetum purpureum* forage. However the microbes closely associated with the NaOH treated maize cobs would presumably have to obtain more of their N substrate from the liquid

Figure 4:
Experiment 1: The relationship between mean rumen N-NH_3 concentration and the digestion in nylon bags for 9 h (□) or 24 h (■) of NaOH treated maize cobs

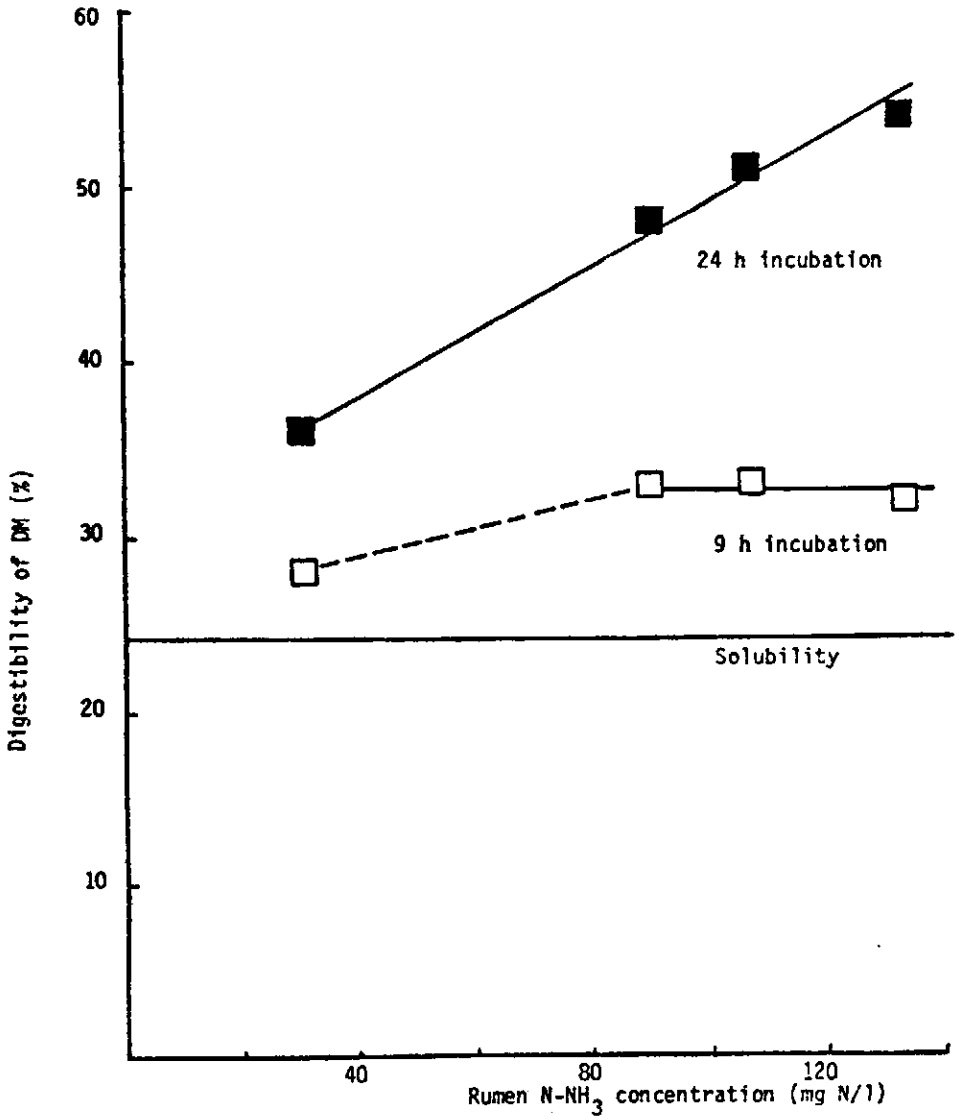
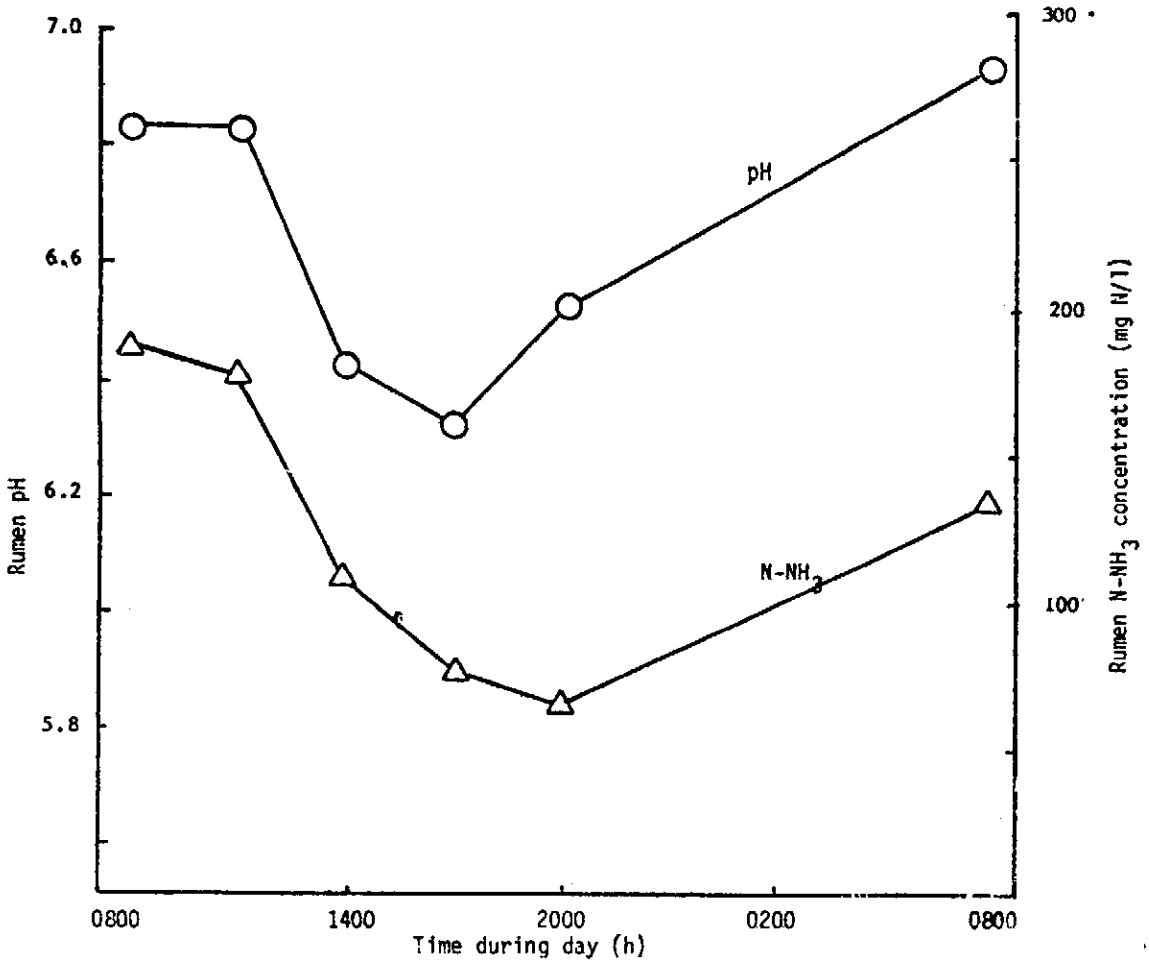


Figure 5:
Experiment 2: Rumen pH (0) and NH_3-N concentration (△) in sheep given at 0830 h Pennisetum purpureum forage alone ad libitum



phase of rumen digesta, and therefore are likely to be more susceptible to a low concentration of soluble N substrates in the liquid phase of rumen digesta. Feed particle N can be used directly as a microbial N substrate in sheep given a diet of oaten chaff and sugar the proportion of microbial ^{15}N derived from rumen $^{15}NH_3$-N was lower in microbes associated with particulate matter (51%) than those in the fluid phase (75%) (Cottle, 1980). Similarly Dixon and Parra (1983) observed a much greater reduction in the rumen digestion of maize cobs (with or without

NaOH treatment) than of *Pennisetum purpureum* and *Cenchrus ciliaris* forages due to dietary concentrate supplementation, and suggested that this was due to a greater dependence on microbial nutrients (other than NH_3-N) present in rumen fluid by the microbes digesting the maize cobs.

The soyabean meal supplement was included to determine whether provision of dietary true protein than urea N increased rumen DM digestion. Many rumen microbial species require and utilize amino acid and peptide N (Allison, 1969; Nolan *et al.*, 1976; Maeng *et al.*, 1976) and there are several experiments that suggest that microbial growth may be stimulated when dietary NPN is replaced by true protein (Hume *et al.*, 1970; Ben-Ghedalia *et al.*, 1978; Teather *et al.*, 1980). However in Experiment 1, the increased DM digestion due to the soyabean meal supplementation could be attributed enterely to the increased rumen NH_3-N concentration. This suggests that in those sheep given diets where a considerable proportion of the diet consisted of true protein from the forage, whether NPN or true protein supplements were used was not of importance.

Experiment 2 showed that in sheep given mature chopped *Pennisetum purpureum* forage containing 6.8% (Nx6.25) the rumen NH_3-N concentration was above the minimum concentration required for maximum rate of DM digestion for most of the day. This is consistent with the experiment of Chicco *et al.* (1971) where growth rate of cattle given green chopped *Pennisetum purpureum* forage was not improved by NPN supplementation.

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Received June 25, 1984