

Influence of casein and glucose or starch supplementation in the rumen or abomasum on utilization of *Eragrostis curvula* hay by sheep

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In one experiment the effects of casein in the abomasum was compared to isonitrogenous quantities of casein or urea in the rumen. In another two experiments the effects of casein in the rumen or abomasum were compared to isocaloric quantities of glucose or starch or to treatments where casein was partially replaced by these carbohydrates. Casein and glucose or starch were administered to the rumen or abomasum twice daily as pulse doses in quantities varying between 30 and 80 g per day. Measurements include voluntary intake of hay, ADG, wool growth, non-ammonia N (NAN) flow to the abomasum, and N-retention. Results suggest that supplementation with casein instead of NPN in the rumen may be beneficial. Secondly, post-ruminal administration of casein increased ADG, wool growth and, on occasion, hay intake. Post-ruminal administered casein furthermore, increased NAN flow to the abomasum and N-retention. Thirdly, glucose or starch supplementation to the rumen or abomasum alone was not beneficial in terms of ADG, wool growth and N-retention. It is concluded that indispensable amino acids at the tissue level of sheep are limiting with such forages, rather than energy — a fact which should be taken into account with supplementation in practice.

In een eksperiment is die invloed van kaseïen in die abomasum vergelyk met isostikstof hoeveelhede kaseïen of ureum in die rumen. In twee verdere eksperimente is die invloed van kaseïen in die rumen of abomasum vergelyk met isokaloriese hoeveelhede glukose of stysel, of met behandelings waar kaseïen gedeeltelik verplaas is deur genoemde twee koolhidrate. Kaseïen en glukose of stysel is deur die rumen- of abomasale kanule twee keer per dag in enkel dosisse toegedien, teen hoeveelhede wat tussen 30- en 80 g per dag gewissel het. Vrywillige inname van hooi, GDT, wolgroei, nie-ammoniak-N(NAN)-vloei na die abomasum en N-retensie is gemeet. Die resultate dui daarop dat supplementering met kaseïen eerder as met NPN in die rumen voordele kan inhou. Tweedens, het abomasale toediening van kaseïen GDT, wolgroei en soms hooi-inname, verbeter. Verder het dit ook NAN-vloei na die abomasum en N-retensie verhoog. Derdens, was toediening van glukose of stysel in die rumen of abomasum alleen nie voordelig in terme van GDT, wolgroei of N-retensie nie. Die gevolgtrekking is gemaak dat onontbeerlike aminosure op weefselvlak van skape by hierdie tipe ruvoere beperkend is, eerder as energie, — 'n feit wat met supplementering in die praktyk in gedagte gehou moet word.

Keywords: Casein, *Eragrostis curvula*, glucose, sheep, starch, supplementation.

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Introduction

The primary objective of supplementing with protein and energy on veld pasture is to rectify possible nutrient shortages and to stimulate intake. Whether intake is increased depends on a number of conditions which have not been studied adequately.

Voluntary intake on low to medium quality pasture is hampered by distension of the rumen as a result of slow fermentation of the forage material. This may be offset partially by continuous addition of NPN to the rumen (Redman, Kellaway & Leibholz, 1980; Leng, 1984; Egan & Doyle, 1985), although others disagree (Romero, Siebert & Murray, 1976; Mizwicki, Owens, Poling & Burnett, 1980). Intake may be stimulated also by rumen-protected protein, because the amount of amino acids absorbed from the gut can alleviate or modify the effects of slow fermentation (Egan, 1965; Leng, 1981; 1984).

Stimulation of voluntary intake by rumen-protected protein has been shown on a number of occasions (Egan, 1965; Kempton & Leng, 1979), sometimes with very spectacular results (Lindsay & Loxton, 1981; Lee, Hennessy, Williamson, Nolan, Kempton & Leng, 1985). There have been convincing results to the contrary as

well (Redman *et al.*, 1980; Kung & Huber, 1983; Oke, Loerch & Deetz, 1986; Meissner & Paulsmeier, 1988). The present study further addressed the problem by using post-ruminal addition of casein instead of rumen-protected protein through the feed.

Some studies in the literature did not balance treatments effectively. This makes it difficult to distinguish whether intake and production responses were due to rumen-protected protein or to the inevitable energy increment. In the present investigation, care was taken to balance treatments on an isocaloric basis with glucose or starch.

The supplementation of glucose or starch to the rumen or lower gut offered the opportunity to study their effects in addition to those of casein. Utilization of ammonia for microbial protein synthesis is enhanced usually by easily fermentable carbohydrates, although cellulose digestion may be suppressed (Leng, 1981). However, if such carbohydrates were to be digested in the small intestine they would supply 11—30% more energy (Leng, 1981) than if fermented in the rumen. This should favour the use of energy supplements that can bypass the rumen.

Procedure

Experiment 1 — Comparison of isonitrogenous urea and casein in the rumen with casein in the abomasum

Four Merino-type wethers of about 35 kg live mass were equipped with rumen and abomasal cannulae and allocated to one of four treatments in a 4 × 4 Latin-square design. A standard average quality (*Eragrostis curvula*) hay was fed *ad lib* to sheep in all four treatments. The hay analysed 0,85% N in the dry matter and 55% *in vitro* digestibility of organic matter.

The four treatments were:

1. A control, where the hay was supplemented with a basal supplement only.
2. As 1, but with the addition of 10 g urea sheep⁻¹ day⁻¹ through the rumen cannula (+10 U_R).
3. As 1, but with the addition of 35 g casein sheep⁻¹ day⁻¹ through the rumen cannula (+35 C_R).
4. As 1, but with the addition of 35 g casein sheep⁻¹ day⁻¹ through the abomasal cannula (+35 C_A).

Treatments 2 to 4 were isonitrogenous.

The basal supplement consisted of 5 g urea, 24 g glucose, 8,15 g K₂SO₄, 15 g dicalciumphosphate, 11 g NaCl and 1 g of a commercial premix of vitamins and trace minerals. The basal supplement was administered in 150 ml water through the rumen cannula in two equal portions, 50% at 08h00 and 50% at 16h00. The casein supplement to the rumen was administered similarly, and to the abomasum in two equal portions in 150 ml 0,9% NaCl solution. On treatments with no abomasal supplement 150 ml 0,9% NaCl solution was administered into the abomasum only.

Each trial period lasted three weeks following one week of adaptation. Before being placed on the next treatment all sheep were returned to the control treatment to minimize carry-over effects. The following measurements were taken during an experimental period:

- Average daily gain (ADG) for two weeks;
- voluntary intake of organic matter (OMI) for one week, whereafter intake was standardized at a lower level for a nitrogen balance study;
- wool growth for three weeks;
- N and non-ammonia N (NAN) flow to the abomasum during the first week of the experimental period; and
- digestibility of organic matter (DOM) and N-balance during the third week of the experimental period.

The sheep were weighed after an over-night fast for calculation of ADG. Wool growth was measured from a standardized midrib patch of 100 cm². For estimating N and NAN, flow of digesta from the rumen to the abomasum was measured using Cr-EDTA as a marker (Faichney, 1980). N-balance was measured according to standard procedures.

Wool samples were cleaned by the following procedure: Samples were washed three times in hexane solution, then once with a liquid soap, rinsed thoroughly in water at room temperature, finally washed in alcohol and left at environmental conditions of 60% RH and 25°C until dry. Residual grass OM in the sample was

carefully removed with tweezers to determine clean fleece yield.

Nitrogen contents of hay, faeces and urine were determined by macro-Kjeldahl according to standard procedures. Rumen and abomasal NH₃-N concentrations were determined by the micro-Kjeldahl method as described by Markham (1942). Non-ammonia N, assumed to be an estimate of amino acid N flowing through the abomasum, was calculated as the difference between total N flow and NH₃-N flow, following correction for Cr absorption and overestimation of N by the single-marker method (Faichney, 1980). To avoid contamination with casein, abomasal samples for these determinations were spaced to the effect that any particular sampling period was at least 4 h post-administration.

The statistical analysis for a Latin-square design was described by Fisher & Yates (1963). However, two sheep were erroneously allocated in the third and fourth periods, so a fifth period was introduced to ensure allocation of all sheep to all treatments. Hence, the data were interpreted as unbalanced and consequently analysed by the general linear-models procedure utilizing least-square means. Treatments were compared using the Bonferroni *t* test.

Experiment 2 — Comparison of glucose in the rumen with isocaloric glucose and casein in the abomasum

Five Merino-type wethers of about 35 kg live mass, equipped with rumen and abomasal cannulae, were allocated to one of five treatments in a 5 × 5 Latin-square design which was conducted simultaneously with Experiment 1.

The five treatments were:

1. A control, where the same *E. curvula* hay and basal supplement as in Experiment 1 were fed with the addition of 10 g urea sheep⁻¹ day⁻¹.
2. As 1, but with the addition of 80 g glucose sheep⁻¹ day⁻¹ through the rumen cannula (+80 Gl_R).
3. As 1, but with the addition of 80 g glucose sheep⁻¹ day⁻¹ through the abomasal cannula (+80 Gl_A).
4. As 1, but with the addition of 40 g glucose and 40 g casein sheep⁻¹ day⁻¹ through the abomasal cannula (+40 Gl_A +40 C_A).
5. As 1, but with the addition of 80 g casein sheep⁻¹ day⁻¹ through the abomasal cannula (+80 C_A).

The control treatment here corresponded with Treatment 2 in Experiment 1. Treatments 1 to 3 were isonitrogenous and Treatments 2 to 5 were isocaloric.

All procedures, measurement techniques adopted and parameters observed were as described for Experiment 1, whereas the statistical analysis was as prescribed for a 5 × 5 Latin-square design (Fisher & Yates, 1963).

Experiment 3 — Comparison of starch in the rumen with isocaloric combinations of starch plus casein in the rumen and abomasum

This experiment was designed as a 4 × 4 factorial. Four Merino-type wethers varying in live mass from 27 to 49 kg were used per treatment but the animals were

blocked for live mass before allocation. All animals were equipped with rumen and abomasal cannulae.

Sheep had access *ad lib* for six weeks to average quality *E. curvula* hay as in Experiments 1 and 2. In addition they received the following supplements in one of four treatments:

1. 17 g urea and 90 g starch sheep⁻¹ day⁻¹ through the rumen cannula (17 U_R 90 S_R);
2. 60 g casein and 30 g starch sheep⁻¹ day⁻¹ through the rumen cannula (60 C_R 30 S_R);
3. 10 g urea and 20 g starch sheep⁻¹ day⁻¹ through the rumen cannula, plus 25 g casein and 45 g starch through the abomasal cannula (10 U_R 20 S_R; 25 C_A 45 S_A); and
4. 10 g urea and 20 g starch sheep⁻¹ day⁻¹ through the rumen cannula, plus 70 g casein through the abomasal cannula (10 U_R 20 S_R; 70 C_A).

Treatments 1 to 3 were isonitrogenous and 1 to 4 isocaloric.

Sheep received a basal supplement through the rumen cannula as well. The basal supplement consisted of 8,15 g K₂SO₄, 12 g dicalciumphosphate, 11 g NaCl and 1 g of a commercial vitamin and trace mineral mixture. All supplements were divided 50:50 into a morning portion given at 08h00 and an afternoon portion given at 16h00. Supplements to the abomasum were administered in a 150 ml solution of 0,9% NaCl and sheep on treatments with no abomasal supplement received a similar volume of 0,9% NaCl.

Parameters observed during the six-week trial were ADG, voluntary intake of hay OM, clean wool yield and fibre diameter of the wool. Live masses and intakes were recorded every week to enable study of the pattern of change with time. Wool growth was measured through total shearing of the sheep at the start and termination of the trial, while fibre diameter was measured in a Wira Wool Fineness meter No. 4147.

The results were analysed statistically with a one-way analysis of variance procedure employing Duncan's *t* test for individual treatment differences.

Results

Comparison of isonitrogenous urea and casein in the rumen with casein in the abomasum

Results are presented in Table 1.

Voluntary intake of hay was not significantly influenced by treatment. Wool production of sheep on the +35 C_A treatment tended to be higher. ADG was significantly higher on the +35 C_A treatment while other treatments did not differ.

Flow of NAN at the abomasum was in proportion to N intake for the treatments Control, +10 U_R and +35 C_R, and not significantly different from one another, while significantly more NAN was recorded for treatment +35 C_A.

For digestibility and N-balance measurement, intake was lowered to decrease sheep effects (Table 1).

The apparent digestibility of OM did not differ significantly between treatments. N-retention was changed, mainly as a result of differences in urinary excretion of N. N-retention did not differ significantly between the Control and +10 U_R, but was significantly greater for the +35 C_R and +35 C_A treatments. N-retention also was significantly higher when casein was supplemented in the abomasum than when it was supplemented in the rumen (Table 1).

Comparison of glucose in the rumen with isocaloric glucose and casein in the abomasum

Results are shown in Table 2.

Voluntary intake of hay was not influenced significantly by abomasal supplementation of casein but was suppressed significantly by glucose supplementation in both the rumen and abomasum. The suppression caused

Table 1 Effect of N source and site of supplementation on N flow and utilization by sheep on average quality *E. curvula* hay

	Control	+10 U _R *	+35 C _R *	+35 C _A *	SE _m
Hay OMI (g day ⁻¹)	741	758	767	777	7,6
ADG(g)	15,5 ^a	-1,25 ^a	17,8 ^a	53,8 ^b	11,4
Wool (mg 100cm ⁻² day ⁻¹)	66,5	71,8	57,8	83,0	5,4
N intake from hay (g day ⁻¹)	6,74	6,89	6,98	7,07	0,7
+ N supplem.(g day ⁻¹)	9,04 ^a	13,8 ^b	14,0 ^b	14,1 ^b	1,2
NAN at abomasum (g day ⁻¹)	5,20 ^a	5,65 ^a	6,05 ^a	10,0 ^b	1,1
Digestibility and N-balance					
Hay OMI (g day ⁻¹)	492	500	524	536	10,2
Apparent DOM (%)	60,7	58,3	61,3	63,5	1,0
Total N intake (g day ⁻¹)	6,57 ^a	11,3 ^b	11,4 ^b	11,6 ^b	1,1
N in faeces (g day ⁻¹)	2,70	2,92	2,93	2,89	0,05
N in urine (g day ⁻¹)	4,32 ^a	8,68 ^c	7,86 ^{bc}	7,06 ^b	0,9
N retained (g day ⁻¹)	-0,45 ^a	-0,31 ^a	0,62 ^b	1,65 ^c	0,4

^{abc} Values in the same line with different superscripts differ at the 5% level of probability.

* U_R = urea in rumen; C_R = casein in rumen; C_A = casein in abomasum.

Table 2 Effect of glucose and casein and site of supplementation on N flow and utilization by sheep of average quality *E. curvula* hay

	Control	+80 GI _R *	+80 GI _A *	+40 GI _A +40 C _A	+80 C _A *	SE _m
Hay OMI (g day ⁻¹)	830 ^b	655 ^a	663 ^a	768 ^{ab}	783 ^{ab}	34,6
ADG(g)	24,6 ^{ab}	-1,20 ^a	-26,6 ^a	88,8 ^b	81,0 ^b	19,4
Wool (mg 100cm ⁻² day ⁻¹)	88,1 ^{ab}	80,4 ^a	92,6 ^{ab}	90,7 ^{ab}	111 ^b	15,4
N intake from hay (g day ⁻¹)	7,54 ^b	5,96 ^a	6,10 ^a	6,99 ^{ab}	7,33 ^{ab}	0,3
+ N supplem.(g day ⁻¹)	13,9 ^b	12,5 ^a	12,6 ^a	18,5 ^c	24,0 ^d	2,3
NAN at abomasum (g day ⁻¹)	6,75 ^a	6,05 ^a	5,20 ^a	14,8 ^b	20,5 ^c	2,9
Digestibility and N-balance						
Hay OMI (g day ⁻¹)	476 ^b	480 ^b	450 ^a	477 ^b	478 ^b	5,6
Apparent DOM (%)	62,1	59,5	59,1	61,4	60,7	0,6
Total N intake (g day ⁻¹)	10,7 ^b	10,9 ^b	10,0 ^a	16,0 ^c	21,3 ^d	2,2
N in faeces (g day ⁻¹)	2,74 ^a	3,02 ^{ab}	3,34 ^b	2,98 ^{ab}	2,84 ^{ab}	0,1
N in urine (g day ⁻¹)	6,96 ^a	7,50 ^a	7,74 ^a	11,5 ^b	15,2 ^c	1,5
N retained (g day ⁻¹)	1,14 ^{ab}	0,40 ^{ab}	-1,04 ^a	1,56 ^{bc}	3,18 ^c	0,7

^{abc} Values in the same line with different superscripts differ at the 5% level of probability.

* GI_R = glucose in rumen; GI_A = glucose in abomasum; C_A = casein in abomasum.

a reduction in ADG, *albeit* non-significantly, in comparison to the Control. ADG was increased significantly with casein addition to the abomasum, with no difference whether 40 g casein or 80 g casein per day was supplemented (Table 2). Wool production was increased significantly on the +80 C_A treatment in comparison to the +80 GI_R treatment.

Because of the lower intake of hay, N intake was slightly lower for the +80 GI_R and +80 GI_A treatments. This tended to decrease NAN flow to the abomasum (Table 2). The supplementation of 40 g casein per day to the abomasum (+40 GI_A; +40 C_A) significantly increased NAN flow, while a further significant increase was realized when 80 g casein per day (+80 C_A) was supplemented.

As in Experiment 1, intake of hay was reduced to measure digestibility and N-balance, so intakes did not differ except where 80 g glucose was supplemented to the abomasum (Table 2). Despite the reduced feed supply sheep ate significantly less hay on this treatment.

The apparent digestibility of OM did not differ significantly between treatments. More N was excreted in the faeces on the +80 GI_A treatment in comparison to all other treatments whilst more N was excreted in the urine when casein was administered to the abomasum. N-retention tended to be lowest on the +80 GI_A treatment and was highest with the +80 C_A treatment.

Comparison of starch in the rumen with isocaloric combinations of starch plus casein in the rumen and abomasum

Results are shown in Table 3 and displayed in Figure 1.

Although sheep were blocked for live mass to decrease animal variation, the attempt was only partially successful. Consequently intake of hay and ADG were expressed relative to metabolic mass in Table 3.

Voluntary intake of hay did not differ significantly with starch or casein addition to the rumen, or when a portion of the starch and casein was shifted to the abomasum (25 C_A 45 S_A). Hay intake increased significantly when casein administration to the abomasum reached 70 g per day. This difference in intake was realized mainly during the first three weeks, whereafter it tapered off (Figure 1).

ADG tended to increase when casein replaced starch whether in the rumen or abomasum (Table 3), although only the change with abomasal administration was significant. The highest ADG was realized with 70 g casein addition per day to the abomasum but, as with intake of hay, the main difference was during the first three weeks of treatment (Figure 1). In contrast, ADG with 25 g casein and 45 g starch in the abomasum, initially not different from the two ruminal treatments, became significantly greater after four weeks.

Wool growth increased significantly when 70 g casein was supplemented in the abomasum though none of the other treatments differed. Treatments did not affect fibre diameter significantly (Table 3).

Discussion

Limitations to experimental design and procedure

A factorial design would probably be more appropriate than a Latin Square when studying ruminal and post-ruminal supplementation. A factorial design is not limited by carry-over or period effects which decrease the sensitivity of tests. A second advantage of a factorial design is a relatively short study period. For the Latin Square, sheep have to cope in metabolism crates for long periods which wear them down considerably. We tried to minimize such effects by shortening adaptation periods somewhat and by feeding them in small pens during the period of standardization (see Procedure).

Table 3 Effect of starch and casein and site of supplementation on production parameters of sheep fed on average quality *E. curvula* hay

	17 U _R * 90 S _R	60 C _R * 30 S _R	10 U _R 20 S _R * 25 C _A 45 S _A	10 U _R 20 S _R * 70 C _A	SE _m
Hay OMI (g w ^{0.75} day ⁻¹)	52,0 ^a	56,9 ^{ab}	58,1 ^{ab}	68,7 ^b	3,4
(g day ⁻¹)	(775 ^a)	(845 ^{ab})	(796 ^{ab})	(991 ^b)	(48,7)
ADG(g.w ^{-0.75} day ⁻¹)	2,74 ^a	4,36 ^{ab}	6,85 ^{bc}	9,29 ^c	1,4
(g day ⁻¹)	(35,7 ^a)	(63,1 ^{ab})	(96,5 ^{bc})	(135 ^c)	(17,9)
Wool (g day ⁻¹)	6,03 ^a	7,40 ^{ab}	7,36 ^{ab}	9,49 ^b	0,7
Fibre diameter (μ)	22,3	19,8	20,8	22,4	0,8

^{abc} Values in the same line with different superscripts differ at the 5% level of probability.

* U_R = urea in rumen; S_R = starch in rumen; C_R = casein in rumen; S_A = starch in abomasum; C_A = casein in abomasum.

The disadvantage of a factorial design is physical. A total of at least 16 rumen and abomasal cannulated sheep would have been required in Experiment 1 and simultaneously 20 in Experiment 2 which cannot be accommodated if infusions and digesta flows are to be measured. Therefore, we decided upon Latin Squares in the flow studies (Experiments 1 and 2) and a factorial design in Experiment 3 to test the trends in ADG, wool growth and voluntary intake. There are, without doubt, limitations to this approach.

Administration of casein and glucose or starch to the rumen or abomasum was done in two equal pulse doses rather than more conventional regular infusions (Egan, 1965; Redman *et al.*, 1980). This was thought to be more in accordance with supplementation in practice. Additionally, in the case of casein, a pulse dose may evoke elevation of growth hormone and insulin levels (Clark, 1974; Barry, Manley, Davis & Redekopp, 1982) which should favour a positive response to voluntary intake. On the negative side, though, glucose administration in this way may have flooded the small intestine causing a

slight diarrhoea. This could have been responsible for the observed reduction in ADG and N retention (Table 2), and one is therefore reluctant to draw conclusions from this particular treatment.

Postruminal casein and voluntary intake of hay

Intake was not influenced by abomasal administration of casein in Experiments 1 and 2 (Tables 1 and 2), but was initially increased in Experiment 3 (Table 3). These conflicting results support the literature where some reports show increases in intake with casein infusion or rumen-protected protein (Egan, 1965; Kempton & Leng, 1979; Lindsay & Loxton, 1981; Lee *et al.*, 1985) whereas others do not (Redman *et al.*, 1980; Kung & Huber, 1983; Oke *et al.*, 1986; Meissner & Paulsmeier, 1988).

Of interest is the initial response observed in Experiment 3, while at a later stage the intake response tapered off. One is inclined to assume that this peculiar result relates to amino acid requirements. Young growing sheep were used in Experiment 3 and intake was stimulated, presumably, when the requirements for indispensable amino acids were still high but the need declined when the requirements were met. This suggests that the magnitude of the intake response and its duration may be a function of nutritional requirements. There is some support for this postulate in dairy cows (Clay & Satter, 1979) and in young growing steers (Hennessy, Williamson, Lowe & Baigent, 1981).

Postruminal casein vs casein and urea in the rumen

In Experiment 1 no response in intake of hay, ADG, wool production or digestibility was observed when either urea or casein was supplemented to the rumen. This may be due to the fact that N intake on the Control was already adequate within the limitations imposed by the fermentation potential of the hay. Egan & Doyle (1985) found positive responses to these parameters when N intake was increased from 6,28 g day⁻¹ to 10,22 g day⁻¹ or to 14,08 g day⁻¹ by urea infusion. Nitrogen intake on our Control diet was 9,04 g day⁻¹, part of which was supplied by 5 g urea, and increased to about 14 g day⁻¹ on the supplemented treatments. If 10 g N

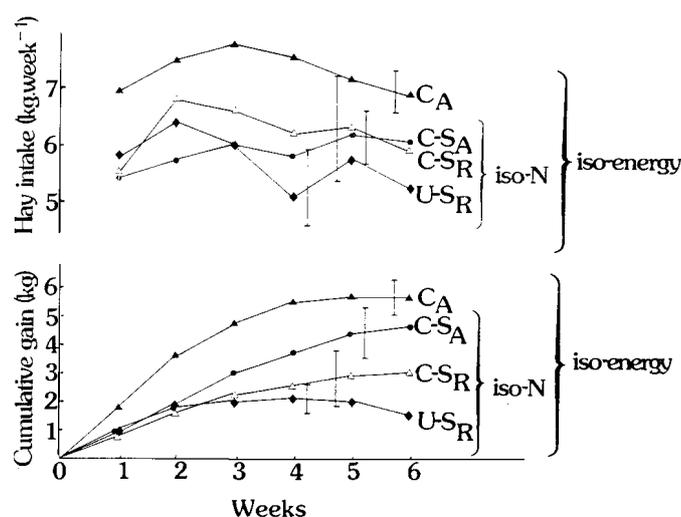


Figure 1 Pattern of hay intake and weight gain over 6 weeks of sheep receiving supplements of casein and starch in the rumen and abomasum.

day⁻¹ is adequate on low to medium quality forages the Control diet at 7% crude protein equivalent in the dry matter may have supplied sufficient N.

Nitrogen supplementation to the rumen also did not alter the production parameters when it was supplied as soluble natural protein (casein) instead of NPN (Table 1). There is evidence to suggest that slow release of amino acids in the rumen will increase efficiency of microbial protein synthesis (Hume, 1974; Maeng, van Nevel, Baldwin & Morris, 1976) although there is evidence also to the contrary (Redman *et al.*, 1980; Mackie, 1988). Whereas the level of casein supplementation may have been too low to cause an increase in NAN flow to the abomasum, a similar result was found by Meissner, Ponelat & Spreeth (1989) with a much higher level. Thus, either natural protein in the rumen does not increase microbial synthesis, or casein, being highly soluble in rumen fluid, is not conducive to slow release of amino acids.

Casein in the rumen, though, did increase N-retention in comparison to urea in Experiment 1 (Table 1); casein supplementation to the rumen in Experiment 3 (Table 3) also improved production, *albeit* not significantly. The improvement in N-retention resulted from less N excreted in the urine, suggesting more efficient utilization of absorbed amino acids at the tissue level when casein was a substrate in the rumen. It is possible that with added casein, more S-containing amino acids were incorporated into microbial amino acids that subsequently were absorbed from the small intestine.

In contrast to N supplementation in the rumen, casein administration to the abomasum increased ADG, wool growth and N-retention in all three experiments. Non-ammonia N flow at the abomasum also was increased in proportion to level of casein administered (Tables 1 and 2). This suggests that microbial protein flow to the abomasum had been increased which would support the observations of Leng (1984). He maintained that fermentable N to the rumen may be supplied directly through a ruminally degraded N source or indirectly through urea-N secretions from the blood. These secretions may originate from rumen-protected amino acids absorbed from the small intestine. Because urea in this manner can be regularly circulated fermentable N is always available for microbial synthesis. Therefore, the value of rumen-protected protein is not only the increased supply of indispensable amino acids to the tissues, but also the regular secretion of N to enhance microbial synthesis.

Glucose and starch supplementation in the rumen and abomasum

The utilization of fermentable N in the rumen for microbial synthesis can be enhanced by supplying easily fermentable carbohydrates, provided sufficient N is available to minimize the depression in cellulose digestion (Leng, 1981) and pH remains above 6.0. In both Experiments 2 and 3 these conditions should have been met. Yet, no response was obtained when either glucose or starch was introduced to the rumen. In fact, with

glucose, intake and consequently ADG was reduced. One explanation for the lack of response may be the type of energy supplement. Hennessy *et al.* (1981) reported that digestibility and utilization of 'native pasture hay' when urea was given, increased more when fed with molasses rather than without. But Hemsley & Moir (1963) and Faichney (1965) showed that addition of sucrose did not enhance urea utilization by sheep on straw diets.

Posttruminally the supplementation of carbohydrates should increase energy availability by 11–30% (Leng, 1981). In Experiments 2 and 3 carbohydrates were administered into the abomasum through glucose and starch, respectively, but no response was observed. Slight diarrhoea was observed with the glucose treatment which may indicate that the absorptive capacity of the small intestine was exceeded. On the other hand, the starch supplementation should have been well within the limits of starch digestion or glucose absorption (Ørskov, 1986). Presumably, carbohydrate supplementation to the abomasum did not cause a response in our studies because supply of digestible energy was not the first limiting factor for production.

Results in the literature on this issue are confusing. Requirements of growing ruminants for glucose and amino acids appear to be interdependent. Kempton, Hill & Leng (1978) showed that when glucose was provided with only low levels of rumen-protected protein, glucose supplementation depressed growth due to reduced feed intake. But when requirements for rumen-protected amino acids had been met, an additional response was obtained by providing posttrimal glucose. In contrast, Leng, Economides & Ball (1978) by continuous infusion of glucose into the duodenum, showed that a response can be achieved whether rumen-protected amino acids were supplied or not. Similar results were reported by Lindsay, Davies & Leibholz (1978).

According to the hypothesis of interdependence between glucose and amino acids, 40 g glucose plus 40 g casein were administered in Experiment 2 into the abomasum (Table 2) and 45 g starch plus 25 g casein in Experiment 3 (Table 3).

Although the additional energy may have contributed to the responses, ADG and wool growth clearly were more closely related to the supply of amino acids by casein. It is concluded, therefore, that indispensable amino acids are limiting on such hays, rather than energy, a fact which should be borne in mind in practical supplementation.

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