

## The digestion of perennial ryegrass (*Lolium perenne* cv. Melle) and white clover (*Trifolium repens* cv. Blanca) by grazing cattle

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1. Pure swards of perennial ryegrass (*Lolium perenne* cv. Melle) as a primary growth (May), trimmed primary growth (early June) and regrowth (late June), and white clover (*Trifolium repens* cv. Blanca) as a mature primary growth (July) and vegetative regrowth (August), were grazed by twelve Friesian steers (mean body-weights throughout experiment 152–231 kg) at daily allocation rates of forage which provided dry matter (DM) intakes ranging from 28 to 36 g/kg body-weight).

2. Total nitrogen contents of the three ryegrasses declined with season (37–20 g/kg DM), but *in vitro* organic matter (OM) digestibilities were relatively constant (0.76–0.79). The clovers had higher N (average 36.6 g/kg DM) and lower fibre and water-soluble carbohydrate contents than the grasses, and *in vitro* OM digestibilities of 0.70 and 0.76 respectively.

3. Duodenal digesta samples were obtained, using a portable sampling apparatus from the animals grazing the pasture, and estimates of the flow of nutrients into the small intestine were derived using two indigestible markers which were continuously infused into the rumen using a portable infusion apparatus. Forage intakes were calculated from estimates of faecal output of indigestible OM and the predicted *in vivo* OM digestibilities of the forages consumed. Coefficients of variation for OM flow to the small intestine and OM intake were 11.8 and 10.9% respectively.

4. The apparent digestion of OM in the rumen ranged from 722 to 741 g/kg digestible OM intake and from 681 to 711 g/kg digestible OM intake for the grass and clover diets respectively. Substantial losses of ingested N before the small intestine were measured on all diets except the regrowth ryegrass. Losses were 0.30 and 0.40 g/g N intake on the primary growth ryegrass and the regrowth clover respectively; N contents were 37 and 39 g/kg DM respectively.

5. Comparison of the values obtained at pasture with that obtained when similar diets were offered to housed cattle (Beever *et al.* 1985) indicated that combined relations relating duodenal OM and non-ammonia-N (NAN) flows to OM and N intakes respectively could be established for each diet. For two diets (primary-growth grass and regrowth clover) the relations were curvilinear; for the remaining diets, the derived relations were linear.

6. The effects of forage species, stage of maturity and variations in the stem:leaf ratio in the grasses, on nutrient supply in grazing animals are discussed, and a combined relation for all grazed and harvested forages examined in this and in a previous study reported by Beever *et al.* (1985) is established. The relation  $Y = 1.430 - 0.0169X$  ( $r = 0.72$ , residual SD 0.140), where  $Y$  is NAN flow/N intake (g/g) and  $X$  is N content in the crop (g/kg OM), indicates that net losses of N across the rumen will occur on diets which contain more than 25.5 g N/kg OM.

7. Possible limitations in the techniques available to measure nutrient digestion and supply in grazing ruminants are discussed.

In recent years, considerable advances have been made in the nutrition of ruminant livestock, aided by the development and use of techniques to partition digestion within the alimentary tract between fermentative (reticulo-rumen) and endogenous enzymic (intestines) modes. This research has led to changes in our concepts of nutrient digestion and supply in ruminants, especially with respect to protein (e.g. Agricultural Research Council, 1980).

This work has, however, concentrated on conserved forages and diets containing a high proportion of concentrates. Published work with fresh forage has been restricted to a limited number of experiments mainly conducted with housed animals (Beever *et al.* 1974,

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1976, 1978; MacRae & Ulyatt, 1974; Ulyatt & MacRae, 1974; Ulyatt & Egan, 1979; Verite *et al.* 1984). Due to technical difficulties there have been few attempts to examine nutrient digestion and supply in ruminants grazing fresh forages (Corbett *et al.* 1976; Cruikshank *et al.* 1985; Beever *et al.* 1986*b*). However, workers in Australia and the UK have developed portable infusion pumps (Corbett *et al.* 1976; Evans *et al.* 1981*b*) and duodenal sampling apparatus (Evans *et al.* 1981*b*) suitable for the measurement of nutrient flow into the small intestine of either sheep or cattle at pasture. The results presented in the present paper are from an experiment using such approaches with grazing cattle, the objective being to quantify the effects of forage species (grass *v.* white clover) and the ratio leaf:stem of grasses of similar digestibilities on nutrient digestion and supply. The diets used in the present study were similar to those described by Beever *et al.* (1985) which were harvested daily and offered to cattle indoors. In the present paper the findings on nutrient supply obtained from both studies are compared, and the suitability of using values obtained with a cut forage to evaluate grazed forage is examined.

## EXPERIMENTAL

### *Forages*

A description of the two pasture species *i.e.* perennial ryegrass (*Lolium perenne* cv. Melle) and white clover (*Trifolium repens* cv. Blanca) and the pre-experimental and experimental management of the two swards has been given by Beever *et al.* (1985).

Briefly, the two forages were offered to growing cattle in the following sequence comprising five experimental periods: 1 (2 to 23 May 1979) primary-growth ryegrass (R1); 2 (24 May to 13 June 1979) trimmed primary-growth ryegrass (R2); 3 (14 June to 4 July 1979) regrowth ryegrass (R3); 4 (5 July to 1 August 1979) mature white clover (W1); 5 (2 to 30 August 1979) regrowth white clover (W2). Fertilizer management of both swards, establishment of grazing plots, and the stages of growth of the forages and post-grazing management were as indicated by Beever *et al.* (1985).

### *Animals and their management*

Twelve castrated Friesian calves, aged 3–4 months and weighing 130 kg (average) were prepared with a PVC rumen cannula (*i.d.* 13 mm) and a PVC 'T'-piece duodenal cannula (*i.d.* 20 mm) anterior to the bile duct. In addition two animals aged 12 months and weighing on average 330 kg, which had been previously prepared with similar rumen and duodenal cannulas, were maintained with the experimental group in order to assess whether indigestible markers infused into the twelve experimental animals were ingested to any appreciable extent as a result of faecal contamination of the pasture.

During the experiment, the calves were allowed to graze continuously apart from two occasions each day (approximately 45 min each) at 08.30 and 16.00 hours, when they were taken to an adjacent shed for removal of collected digesta samples (as appropriate), checking and adjustment of harnesses and infusion and sampling equipment, and general animal maintenance including recording of body-weight. In addition, during periods 4 and 5 when white clover was being examined, all animals were dosed orally at these times with 5 ml Poloxalene (Smith, Kline & French; Welwyn, Herts; 980 ml/l) to prevent legume bloat. Animals had free access to fresh water at all times and mineral blocks were provided, except for the last 2 weeks of each experimental period when measurements of feed intake and duodenal flow were made. All animals were weighed weekly.

### *Pasture management and feed allocation*

The objective was to provide sufficient forage daily to allow a dry matter (DM) intake within the range 18–26 g/kg body-weight, which coincided with the feeding levels adopted

by Beever *et al.* (1985) in the concurrent indoor experiment. This was estimated from Baker (1978) to require a daily allowance of standing crop of 56 g DM/kg body-weight, with measurements of standing crop being made by cutting quadrats of the crop to ground level.

Two systems of pasture allocation were used during the experiment. In period 1 (R1), each plot was internally divided by electric fences into three subplots each approximately 12 m wide, and each subplot was grazed in turn. The amount of DM on offer was estimated from six quadrats (each 0.24 m<sup>2</sup>) cut to ground level each morning from the new day's allocation, and DM content was estimated using a modified microwave oven. The length of subplot required to provide 56 g DM/kg body-weight was then calculated and this was divided into two equal portions by an electric fence, to provide morning and afternoon feed allocations. This system of feed allocation resulted in a high stocking density and considerable wastage of pasture occurred through trampling. For periods 2 and 3, this procedure was changed by increasing the width of the plot to 24 m and forage allocation was based on DM yields determined over the 3 d before each measurement period. When the required length of plot had been determined, this was not varied for the duration of the measurement period; furthermore, the calves were allowed free access to the allocation of each previous day as a rest area to encourage loafing-lying off the experimental feed. This alternative management was found to reduce trampling of the offered forage, and resulted in a more satisfactory utilization of the forage.

Similar systems of pasture management were used for periods 4 and 5 (W1 and W2 respectively) but the width of each plot was increased to 30 m.

#### *Sampling methods*

*Forages.* Samples were taken according to the procedures described previously, and used to determine DM yields of the offered crops by drying in a forced-draught oven for 24 h at 103°. Forage residues were taken after grazing each day during each digesta collection period between 08.30 and 10.00 hours by means of six quadrats cut to ground level, and samples of the resulting forage were dried for 24 h at 103°.

Samples representative of the consumed forage were obtained from the pasture offered in the morning and afternoon, before grazing, by hand cutting at the residual grazing height observed on the paddock from the previous day. These samples were immediately frozen and subsequently bulked over each collection period on an equal DM basis.

*Duodenal digesta.* The twelve calves were divided into two equal groups (A and B) and collections of digesta from each group were staggered by approximately 4 d, similar to the procedures adopted by Beever *et al.* (1985) to maximize use of a limited number of automatic duodenal samplers. Infusions of markers into the reticulo-rumen (CrEDTA and ruthenium phenanthroline (Ru) (Faichney, 1975)) were commenced 13 d (group A) and 8 d (group B) before the end of each experimental period using the portable pumps described by Evans *et al.* (1981*a*) with dose rates as described by Beever *et al.* (1985); i.e. 120 mg Cr and 12 mg Ru/kg expected DM intake. After 5 d of continuous marker infusions, two 24 h continuous collections of duodenal digesta were undertaken using the apparatus described by Evans *et al.* (1981*b*). Collected samples of digesta were removed from each animal at 16.00, 21.00 and finally 09.00 hours on each day, subsequently bulked for each 24 h and processed for analysis according to the procedures adopted by Beever *et al.* (1985), by preparation of the appropriate whole and centrifuged digesta samples.

In addition, when the duodenal samples were removed from the experimental animals, spot samples of duodenal digesta were obtained from the two non-experimental steers. These samples were freeze-dried directly, without processing, to provide samples of whole digesta only.

*Faeces.* During the last 3 d of each infusion period, faecal material was obtained from all

calves by grab sampling at approximately 09.00 and 16.00 hours. Each sample was oven dried (24 h at 103°) and daily bulk samples for each animal were prepared on an equal DM basis.

#### *Sample analysis*

Before chemical analysis, all samples of feed, duodenal digesta and faeces were freeze-dried, then ground to pass through a 1 mm sieve. Samples of duodenal digesta and faeces for Ru analysis (3 g) were subsequently ball milled and finally pelleted (Evans *et al.* 1977).

Analysis of samples, as required, for organic matter (OM), gross energy, nitrogen, ammonia, cellulose, water-soluble carbohydrate, neutral-detergent fibre and *in vitro* OM digestibility were as described by Beever *et al.* (1985).

#### *Calculation of results*

The flows of digesta, and its constituents, into the small intestine of the calves were calculated according to the procedures proposed by Faichney (1975), and mean 24 h flows for each animal were obtained. Duodenal digesta samples obtained from the two non-experimental animals throughout the whole season contained no significant concentrations of Ru and Cr and, consequently, background corrections to the samples obtained from the experimental animals were not necessary. Estimates of faecal OM output were derived from faecal Ru content (expressed on an OM basis) and daily Ru dose rate, and these were used to calculate OM intake (OMI) according to the equation:

$$\text{OMI} = \frac{\text{faecal OM output}}{1 - D}, \quad (1)$$

where  $D$  is the estimated *in vivo* OM digestibility, calculated from:

$$D = 1.04X + 0.00246 \quad (r \ 0.871; \ n \ 45), \quad (2)$$

where  $X$  is the measured *in vitro* OM digestibility (g/kg total OM) of the samples taken to represent consumed forage. Eqn (2) was derived from *in vitro* and *in vivo* OM digestibility values obtained in the indoor feeding experiment reported by Beever *et al.* (1985) for diets corresponding to those used in the present study.

Owing to the nature of the experiments, the dietary treatments were totally confounded with time, such that the amounts of forage consumed increased as the season advanced in response to the increased live weight of the animals. The results are presented both in terms of total quantities of nutrients flowing through the alimentary tract and in relation to live weight as these were found, from the statistical analysis, to be the most appropriate ways of illustrating the experimental values. Alternative ways of expressing the values (e.g. in relation to metabolic body size, live weight<sup>0.75</sup>) were examined but dismissed on the basis that they did not result in any change in the conclusions which could be drawn from the study. To permit further examination of the experimental values by the reader, if deemed appropriate, the mean body-weights of the animals for each period-diet are presented.

#### *Statistical analysis*

Treatment means were compared in a split-plot analysis of variance after tests (Bartlett, 1937) showed that variances and correlations could be treated as equal over five growth periods. There were no main-plot treatments giving thirty-six subplot error df due to eight missing values (one in period 2 (R2), two in period 4 (W1) and five in period 5 (W2)). Further comparisons were made using regression analysis of nutrient flow on nutrient intake.

Table 1. *The botanical and chemical composition (g/kg dry matter (DM) unless stated) of the three perennial ryegrass (Lolium perenne cv. Melle; R) and two white clover (Trifolium repens cv. Blanca; W) diets offered to calves*

	Perennial ryegrass			White clover	
	R1	R2	R3	W1	W2
<b>Botanical composition</b>					
Leaf*	924	730	702	658	887
Stem	64	213	270	—	—
Dead	2	0	23	13	25
Flower head	—	—	—	327	85
Purity	990	943	995	998	997
<b>Chemical composition</b>					
Organic matter	908	912	913	922	902
Water-soluble carbohydrate	128	132	187	93	103
Cellulose	214	244	206	232	201
Neutral detergent fibre	396	447	405	316	241
Nitrogen	37.1	31.4	20.2	34.6	38.6
Gross energy (MJ/kg DM)	19.4	18.9	18.1	19.3	18.8
In vitro organic matter digestibility	786	764	787	695	756

\* Includes petiole for clover.

## RESULTS

### *Composition of crops offered*

The botanical composition of the five pasture diets as offered is given in Table 1. Species purity of all swards was high (> 943 g/kg DM), but there were marked differences between the grasses in leaf:stem ratios (14.4, 3.4 and 2.6 for R1, R2 and R3 respectively). The primary-growth clover (W1) had a higher proportion of flower head to leaf than the regrowth clover (W2).

There were differences in chemical composition between the ryegrass diets. R2 contained more cellulose and neutral-detergent fibre but had less N than R1. R3 had a much lower content of N than either R1 or R2, but similar cellulose and neutral-detergent fibre contents to R1 and a higher water-soluble carbohydrate content than the other grasses. The in vitro OM digestibility for R2 was marginally lower than that for R1 and R3. Cellulose and neutral-detergent fibre contents were higher on W1 than W2, and both clovers had high N contents but water-soluble carbohydrate contents were lower than those for the grasses. In vitro OM digestibility of the primary-growth clover was lower than the regrowth clover and the three grass diets.

### *Nutrient intake, digestion and supply*

As indicated earlier, some problems were encountered within the experiment with respect to pasture growth and allocations. Consequently, the estimated mean DM intakes (i.e. R1 32, R2 28, R3 28, W1 29, and W2 34 g/kg live weight) were all higher than the mean intakes achieved with the housed cattle (Beever *et al.* 1985).

In Table 2, the mean quantities of OM and N consumed, OM and non-ammonia-N (NAN) entering the small intestine, and rumen losses of apparently digestible OM (g/kg digestible OMI (DOMI)) and ingested N(g/d) are given. Mean OMI for the five diets ranged from 4.37 to 7.17 kg/d, and duodenal OM flows (from 1.72 to 3.04 kg/d) reflected these differences, but the values for the clover diets were significantly higher ( $P < 0.01$ ) than

Table 2. Mean quantities of organic matter (kg/d) and nitrogen (g/d) consumed, flowing into the small intestine (as non-ammonia-N; NAN) and present in the faeces (organic matter only) for three perennial ryegrass (*Lolium perenne* cv. *Melle*; R) and two white clover (*Trifolium repens* cv. *Blanca*; W) swards offered to grazing cattle

(Values for the extent of rumen digestion of digestible organic matter and total N, and NAN flow/unit N intake are also given)

	Perennial ryegrass			White clover		SEM
	R1	R2	R3	W1	W2	
Mean body-wt of animals (kg)	152.4	171.2	189.1	208.2	231.0	2.01
Organic matter						
Consumed (kg/d)	5.02	4.37	4.82	5.53	7.17	0.152
Entering small intestine (kg/d)	1.97	1.72	1.85	2.70	3.04	0.078
In faeces (kg/d)	0.83	0.82	0.79	1.39	1.36	0.034
Apparent digestibility (g/kg intake)	835	815	836	752	808	
Extent of rumen digestion of digestible organic matter (g/kg apparently digested)	722	741	738	681	711	19.49
N						
Consumed (g/d)	205.0	151.3	106.5	208.5	306.7	5.64
Entering small intestine (as NAN) (g/d)	140.7	115.6	122.0	151.3	184.1	5.24
Rumen loss of N* (g/d)	64.3	35.7	15.5	57.2	122.6	6.54
NAN flow (g/kg N intake)	0.70	0.77	1.14	0.72	0.60	0.035

\* Includes duodenal  $\text{NH}_3\text{-N}$ .

those recorded for the grass diets. Coefficients of variation for OM flow to the small intestine averaged 11.8% on all diets, compared with 10.9% for estimates of OM intake. On the grass diets, OM digestion in the rumen in relation to DOMI showed little variation from the mean value of 734 g/kg DOMI whilst the value observed on W2 (711 g/kg) was marginally but not significantly ( $P > 0.05$ ) lower. In contrast, the value for W1 (618 g/kg) was significantly ( $P < 0.05$ ) lower than the values for the three grasses but not significantly ( $P > 0.05$ ) different from the value obtained on W2. There was a threefold range in daily N intakes for the five diets, but duodenal NAN flows showed a narrower range due in part to large losses of N across the reticulo-rumen, especially on diets R1, W1 and W2, which all had high dietary N contents. On diet R3, which had the lowest dietary N content, there was an increase in NAN flow into the small intestine relative to the amount consumed. NAN flow per unit N intake ranged across the diets from 0.60 (W2) to 1.14 (R3).

#### DISCUSSION

The main objectives of the present experiment were to evaluate the portable infusion and duodenal sampling apparatus as described by Evans *et al.* (1981 *a, b*) to estimate nutrient flow in grazing animals and to compare the results obtained with those derived from similar cattle fed indoors on known amounts of the same forages harvested daily by cutting. Additionally, the use of contrasting forage species, and the deliberate establishment of different leaf:stem values within the three ryegrasses provided the opportunity to examine further the effects of forage composition (both chemical and physical) on nutrient supply.

*Techniques and methodology in measuring nutrient flow*

The apparatus described by Evans *et al.* (1981*a, b*) was used for the first time under rigorous field conditions, and, from an intended total of 572 animal days of infusion of markers into the rumen with the peristaltic pumps, only 23 d (4%) were unsatisfactory, due in part to battery failure, split infusion tubes and occasional misplacement of the harnesses which secured the pumps. Equally, 92% ( $n$  110) of the planned duodenal collections ( $n$  120) were successfully completed, and reasons why ten duodenal collections had to be discarded were related to digesta blockages, tubing disconnected from the duodenal cannulas or occasional sampler malfunction (e.g. valve remaining open). All such faults were rapidly corrected and overall success rate was good. The health of the animals and the levels of feed consumption were satisfactory at all times; during the experiment the calves gained an average of 80 kg body-weight, equivalent to a daily gain of 0.73 kg.

*Limitations on nutrient-supply values derived from grazing animals*

Estimation of nutrient flow to the small intestine of grazing animals will be affected by the accuracy of the marker techniques adopted and adequacy of the sampling procedures used, in a similar manner to the estimation of nutrient flow in stall-fed animals. Additionally, performance and reliability of the automatic infusion and sampling apparatus may place another constraint on the determination of such values, as discussed previously. The usefulness of such values in the nutritional evaluation of grazed forage, however, is greatly enhanced if measurements of feed intake and faecal output can be obtained in parallel with estimates of nutrient flow. To obtain such values, a variety of techniques has been proposed (Langlands, 1975). One essential prerequisite is an estimation of the composition of the consumed forage. In this regard, animals with oesophageal fistulas would have been appropriate in the present study, but bearing in mind the degree of surgical modification required to estimate nutrient flow, the establishment of a further fistula was considered inappropriate. Thus in the present study hand-plucked samples of the offered forage were taken as representative of the consumed forage. Clearly this could constitute a major limitation on the interpretation of the values obtained in the present study.

From estimates of *in vitro* digestibility of the forage samples considered to be representative of the consumed forages, *in vivo* values of digestibility were calculated using the relation obtained from a concurrent indoor-feeding experiment (Beever *et al.* 1985). It is difficult to comment on the suitability of this approach, but it is possible that the estimates may have been subject to bias as the cattle grazing the forages had considerable opportunity to exert selective preferences within the sward, and during periods 2–5 to graze from the allocation of the previous day. In the concurrent indoor-feeding experiment (Beever *et al.* 1985), grab samples of faeces were obtained using the same infusion and sampling protocol adopted in the present experiment. Estimates of DM intake were calculated using the feed:faeces ratio method (Langlands, 1975) with measurements of *in vitro* OM digestibility being used to predict *in vivo* OM digestibility. These predicted OMI ( $Y$ , g OM/d) were subsequently related to measured OMI ( $X$ , g OM/d) obtained in the indoor experiment, and the following regression equation was derived:

$$Y = 0.98(\text{SE } 0.08) X + 274(\text{SE } 322) \quad r = 0.87.$$

From this it may be calculated that for a predicted intake ( $Y$ ) of 5000 g OM/d, based on the feed:faeces ratio method, the true OMI ( $X$ ) would be 4822 g/d, indicating an over-prediction of intake equivalent to 3.7% of the true value.

In the present study, faecal OM output was calculated from faecal dilution of an indigestible marker (Ru) which was infused continuously into the rumen, and estimated

forage *in vivo* digestibility calculated as described previously. There are several potential sources of error in these procedures. Whilst the marker used was considered acceptable as judged by the criteria given by Kotb & Luckey (1972), the timing of the faecal samples could lead to errors in the estimation of faecal output. In the present study only two sampling times (09.00 and 16.00 hours each day) were used, as these times have been shown to give representative faecal samples with respect to marker concentrations when chromic oxide administered twice daily is used (Langlands, 1975). However, the pattern of marker concentration in faeces when the marker is administered continuously has not been established, and it was not possible in the present study to identify the most appropriate times for faecal sampling, within each day. More frequent faecal sampling, and increasing the number of successive days of sampling, may have improved the estimation of faecal output, and hence intake, but at the same time could have seriously disturbed the grazing pattern of the animals.

Despite the limitations in the approaches adopted, the coefficients of variation in estimates of OM flow to the small intestine, and OMI, appeared to be satisfactory. In relation to the live weights of the animals, the estimated DM intakes were satisfactory (range 28–34 g/kg live weight), and exceeded the highest plane of restricted feeding used by Beever *et al.* (1985), possibly reflecting the high forage allowances given in the present experiment. Furthermore, the findings relating to the digestion of OM (Table 2), indicating that between 681 and 741 g/kg DOMI were apparently digested in the rumen on all diets, are consistent with values previously reported for such diets (Beever *et al.* 1986*a*). On this basis, the values presented in Table 2 appeared to be acceptable and suitable for further examination in relation to the objectives of the study as stated earlier.

#### *Comparison of cut and grazed forage*

Whilst the intention of the present experiment was to promote similar forage DM intakes to those achieved in the indoor study (Beever *et al.* 1985), problems were encountered with the allocation of the pasture and the estimated intakes were higher than those recorded for the stall-held cattle. Some indication of this effect is seen in Fig. 1 where values relating NAN flow to OMI for both the indoor and outdoor studies are presented. Furthermore, some differences in chemical composition between the mechanically harvested and grazed forages were observed. The grasses given indoors had a higher proportion of stem with slightly higher contents of carbohydrate constituents and lower N contents, whilst the harvested clovers had higher N and lower water-soluble carbohydrate contents than the corresponding grazed forages. However, all these differences were small, and whilst it may be inappropriate to ignore them, they were not considered at this stage to be of great nutritional significance.

Higher intakes in the grazing trial provided an extended range for the response variables and overall regression analysis was used to test whether curvilinearity was significant. In order to decide whether weighted overall analysis was required, separate regression equations (on a body-weight basis) describing the amounts of OM and NAN entering the small intestine ( $Y$ ) in relation to OMI and total N intake ( $X$ ) respectively were calculated for the corresponding forages offered indoors or grazed *in situ* (i.e. for each individual diet-period), and Bartlett's (1937) test of homogeneity of variances was applied to test the error variances obtained for each forage given indoors or grazed.

With the values available from the present study, the choice of components to include in the regression equations in order to test the values for homogeneity was limited. Previous studies have reported similar relations (Hogan & Weston, 1970) and research has shown that the flows of OM and NAN into the small intestine are important nutritional indices in relation to the extent of rumen digestion and nutrient supply respectively. Furthermore,



## Digestion of grazed forages

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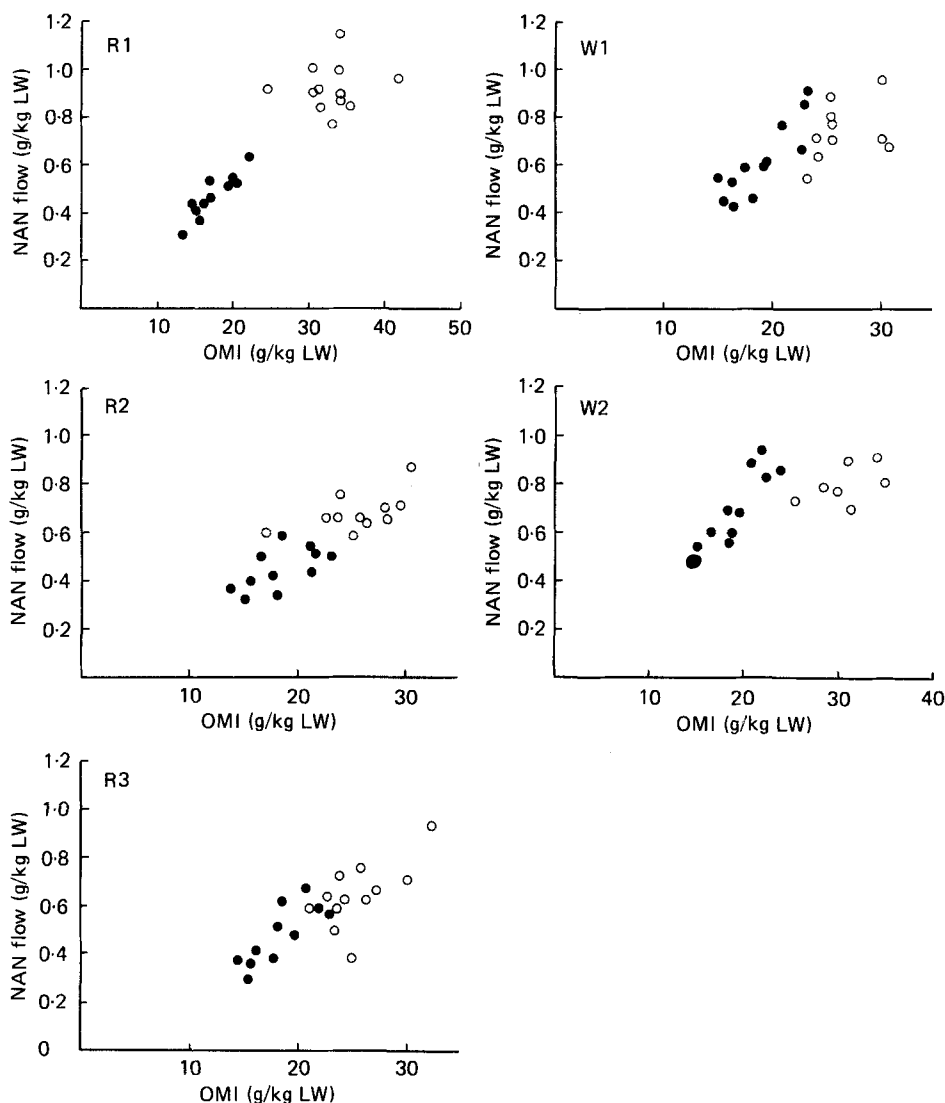


Fig. 1. Individual observations relating non-ammonia-nitrogen (NAN) flow to the small intestine to organic matter intake (OMI) for cattle grazing (○) or fed on harvested (●) perennial ryegrass (*Lolium perenne* cv. Melle; R1, R2 and R3) and white clover (*Trifolium repens* cv. Blanca; W1, W2) forages. LW, live weight.

OMI was used in preference to DM intake to avoid possible biases in the values which could have arisen from soil ingestion by the grazing animals or soil contamination of the consumed forage, both of which would elevate faecal ash and faecal DM output and hence estimates of feed intake.

It was concluded from the statistical analysis, that in all but one situation (OM, diet W2), no differences in variance between the regressions for the respective indoor and outdoor values could be established and consequently the indoor and outdoor results within each period (except W2) were combined and the best-fit, linear or quadratic relations were developed (see Figs. 2 and 3). For diet W2, the OM results were weighted by the reciprocal

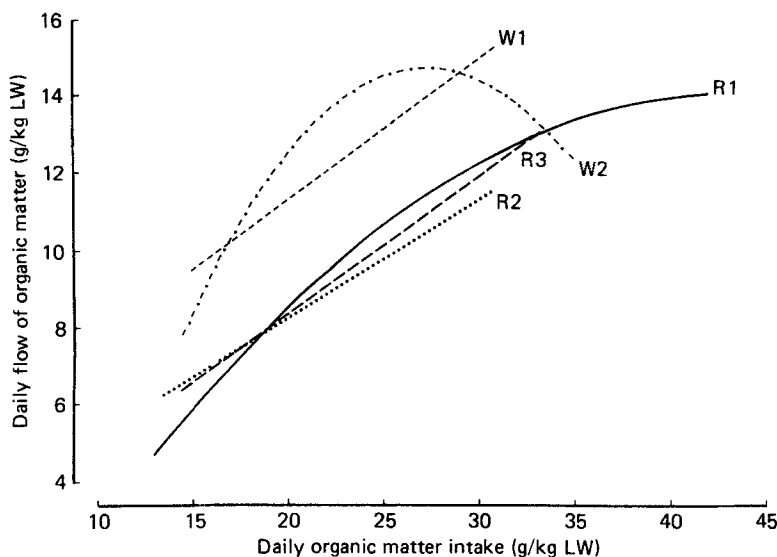


Fig. 2. The relations between organic matter flow to the small intestine ( $Y$ , g/kg live weight (LW) per d) and organic matter intake ( $X$ , g/kg LW per d) for cattle grazing or fed on harvested perennial ryegrass (*Lolium perenne* cv. Melle; R1, R2 and R3) and white clover (*Trifolium repens* cv. Blanca; W1 and W2) forages.

$$R1: Y = 0.865X - 0.0099X^2 - 4.876, r^2 0.90, \text{residual SD (RSD) } 1.048;$$

$$R2: Y = 0.307X + 2.059, r^2 0.734, \text{RSD } 0.940;$$

$$R3: Y = 0.351X + 1.304, r^2 0.581, \text{RSD } 1.440;$$

$$W1: Y = 0.359X + 4.083, r^2 0.560, \text{RSD } 1.549;$$

$$W2: Y = 2.277X - 0.0415X^2 - 16.512, r^2 0.783; \text{RSD } 1.116$$

of the variance obtained from separate regressions and, from overall regression analysis of the weighted data, a quadratic relation for the combined indoor and outdoor values was derived.

From this overall analysis of the results of the grazing experiment combined with the values obtained with cut forage, a further basis on which to examine the effect of forage species, stage of maturity of the forage and level of feeding on nutrient digestion and supply in young cattle was developed.

#### Digestion of fresh forages

From the combined relations presented in Fig. 2 it can be seen that on the legume diets, between 2 and 4 g extra OM/kg live weight per d entered the small intestine compared with the grass diets; the effect being similar in magnitude to the response to forage species reported earlier by Beever *et al.* (1986*a*). In the present experiment, no attempts were made to characterize the origin of the extra OM flow, although it would appear reasonable to conclude on the basis of findings presented by Beever & Siddons (1986) that a major part could be attributed to an increased synthesis of microbial protein within the rumen.

Despite distinct leaf:stem ratios and differing chemical compositions, the regressions for OM presented for the three ryegrasses (Fig. 2) were virtually indistinguishable, with OM flow to the small intestine averaging between 0.36 and 0.40 of OMI at the two extremes of OMI achieved (i.e. 15 and 32 g OM/kg live weight).

In contrast, the N digestion values presented in Fig. 3 indicated a broad compatibility

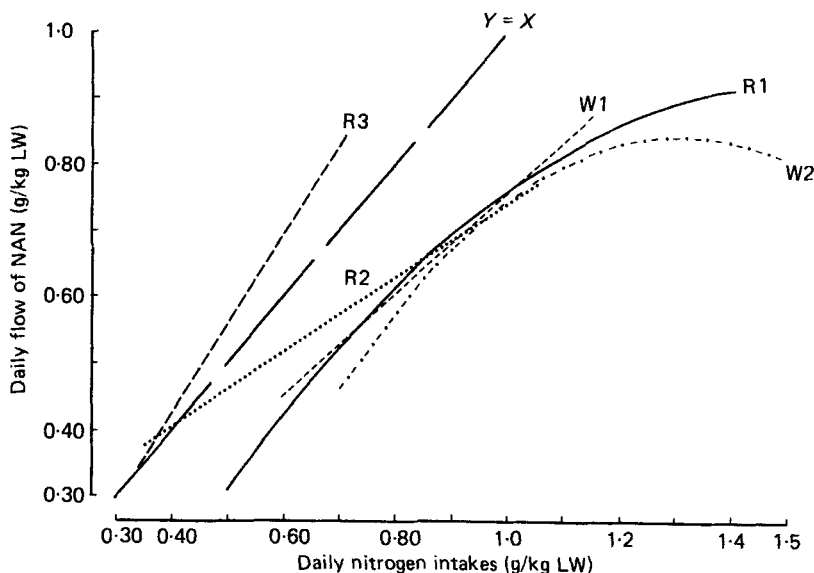


Fig. 3. The relations between non-ammonia-nitrogen (NAN) flow to the small intestine ( $Y$ , g/kg live weight (LW) per d) and N intake ( $X$ , g/kg LW per d) for cattle grazing or fed on harvested perennial ryegrass (*Lolium perenne* cv. Melle; R1, R2 and R2) and white clover (*Trifolium repens* cv. Blanca; W1 and W2) forages.

$$R1: Y = 1.746X - 0.564X^2 - 0.423, r^2 0.892, \text{residual SD (RSD) } 0.085;$$

$$R2: Y = 0.506X + 0.183, r^2 0.789, \text{RSD } 0.068;$$

$$R3: Y = 1.348X - 0.117, r^2 0.656, \text{RSD } 0.090;$$

$$W1: Y = 0.765X - 0.006, r^2 0.507, \text{RSD } 0.102;$$

$$W2: Y = 2.684X - 1.024X^2 - 0.913, r^2 0.712, \text{RSD } 0.083.$$

with respect to forage species and stage of growth, except for diet R3. With all other diets, NAN flow to the small intestine was less than N intake and the extent of this loss increased as N intake increased, especially on diets R1 and W2 where quadratic equations were established. This effect is in broad agreement with the values presented by Ulyatt & Egan (1979) from a collation of values obtained on the digestion of forages by sheep. In contrast, diet R3, which was characterized by a low N content, gave small net gains of N across the rumen at all levels of intake achieved, although it can be seen from Fig. 3 that high N intakes corresponding to those seen on the other four diets were not achieved with this diet.

When all the indoor and outdoor results for all diets were combined and duodenal NAN flow (expressed as g/g N intake,  $Y$ ) was related to N concentration in the forage OM (g/kg,  $X$ ), the following equation was obtained:

$$Y = 1.430 - 0.0169X, r = 0.72, \text{residual SD } 0.140,$$

indicating that when forage N content exceeded 25.5 g/kg, net losses of N between the diet and the duodenum would occur, whilst at a concentration of 40 g N/kg OM in the forage, NAN flow to the small intestine would be only 0.75 of N intake. Equally, at a concentration of 22.1 g N/kg OM as found in diet R3, duodenal NAN flow would be expected to be 1.06 times N intake, a finding in broad agreement with the values presented in Fig. 3. Support for this overall summary of the results is provided by the earlier studies of Hogan & Weston (1970) and more recently by Beever & Siddons (1986).

*Consequences for N utilization*

The results of the present study have confirmed earlier observations by Beever *et al.* (1985, 1986*a, b*) and others that in animals given high-quality forages, a significant proportion of the ingested N may fail to reach the small intestine as NAN and is probably absorbed from the rumen as ammonia. This effect appears to be due to imbalance in the rumen between degraded N and degraded energy supply (Beever *et al.* 1986*a*). It has been demonstrated (Black *et al.* 1979; Barry *et al.* 1982; Thomson *et al.* 1984) that animals consuming fresh forages, particularly grass, will respond, in terms of increased animal performance, to protein supplements but it may ultimately be possible to achieve similar effects by manipulation of forage composition through the introduction of plant tannins (Thomson *et al.* 1971; Barry & Manley, 1984) or the use of protein protection agents (Beever *et al.* 1987) or energy-rich supplements.

*Conclusions*

It is evident from the present study that the infusion and sampling apparatus described by Evans *et al.* (1981*a, b*) provided realistic estimates of nutrient flow to the small intestine of grazing ruminants. It is also evident that current knowledge on the nutritive value of fresh forages is limited. Whilst the findings obtained in the present study are useful, further research to investigate the effect of factors such as forage species, variety, sward management and animal-plant interactions on the nutritive value of fresh forage is urgently required. What currently remains uncertain is whether fresh forages can be evaluated by cutting and feeding to housed animals and the results applied with confidence to animals grazing pasture. Given the findings of the present study, it is reasonable to conclude at this stage that both options may be of value in evaluating grazed forage, and to disregard the usefulness of direct measurements of nutrient supply in grazing animals would be premature.

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## REFERENCES

- Agricultural Research Council (1980). *The Nutrient Requirements of Ruminant Livestock* Farnham Royal: Commonwealth Agricultural Bureaux.
- Baker, R. D. (1978). *Proceedings of the British Grassland Society* 119-126.
- Baker, R. D., Alvarez, F. & Ledu, Y. L. P. (1981). *Grass and Forage Science* 36, 189-199.
- Barry, T. N. & Manley, T. W. (1984). *British Journal of Nutrition* 51, 493-504.
- Barry, N. N., Manley, S. R., Davis, S. R. & Redekopp, C. (1982). In *Forage Protein in Ruminant Animal Production*, Occasional Publication of the British Society of Animal Production no. 6, pp. 146-148 [D. J. Thomson, D. E. Beever and R. G. Gunn, editors]. Thames Ditton: British Society of Animal Production.
- Bartlett, M. S. (1937). *Proceedings of the Royal Society* 160, 262-282.
- Beever, D. E., Cammell, S. B. & Wallace, A. S. (1974). *Proceedings of The Nutrition Society* 33, 73A.
- Beever, D. E., Dhanoa, M. S., Losada, H. R., Evans, R. T., Cammell, S. B. & France, J. (1986*a*). *British Journal of Nutrition* 56, 439-454.
- Beever, D. E., Losada, H. R., Cammell, S. B., Evans, R. T. & Haines, M. J. (1986*b*). *British Journal of Nutrition* 56, 209-225.

- Beever, D. E., Losada, H. R., Gale, D. L., Spooner, M. C. & Dhanoa, M. S. (1987). *British Journal of Nutrition* **57**, 57–67.
- Beever, D. E. & Siddons, R. C. (1986). In *Control of Digestion and Metabolism in Ruminants. Proceedings of VIth International Symposium on Ruminant Physiology*, pp. 479–497 [L. P. Milligan, W. L. Grovum and A. Dobson, editors]. Englewood Cliffs, New Jersey: Prentice Hall.
- Beever, D. E., Terry, R. A., Cammell, S. B. & Wallace, A. S. (1978). *Journal of Agricultural Science, Cambridge* **90**, 463–470.
- Beever, D. E., Thomson, D. J. & Cammell, S. B. (1976). *Journal of Agricultural Science, Cambridge* **86**, 443–452.
- Beever, D. E., Thomson, D. J., Ulyatt, M. J., Cammell, S. B. & Spooner, M. C. (1985). *British Journal of Nutrition* **54**, 763–775.
- Black, J. L., Dawes, S. T., Colebrook, W. F. & James, K. J. (1979). *Proceedings of the Nutrition Society, Australia* **4**, 126.
- Corbett, J. L., Furnival, E. P., Inskip, M. W., Perez, C. J. & Pickering, F. S. (1976). *Proceedings of the Australian Society of Animal Production* **11**, 329–332.
- Cruikshank, G. J., Poppi, D. P. & Sykes, A. R. (1985). *Proceedings of the New Zealand Society of Animal Production* **45**, 113–116.
- Evans, C. C., MacRae, J. C. & Wilson, S. (1977). *Journal of Agricultural Science, Cambridge* **89**, 17–22.
- Evans, R. T., Hayes, D. G. & Beever, D. E. (1981a). *Laboratory Practice* **30**, 591–593.
- Evans, R. T., Skelton, K. V. & Beever, D. E. (1981b). *Laboratory Practice* **30**, 997–1000.
- Faichney, G. J. (1975). In *Digestion and Metabolism in the Ruminant*, pp. 277–291. [I. W. McDonald and A. C. I. Warner, editors]. Armidale, Australia: University of New England Publishing Unit.
- Hogan, J. P. & Weston, R. H. (1970). In *Physiology of Digestion and Metabolism in the Ruminant*, pp. 474–485 [A. T. Phillipson, editor]. Newcastle upon Tyne: Oriel Press.
- Kotb, A. R. & Luckey, T. D. (1972). *Nutrition Abstracts and Reviews* **42**, 813–845.
- Langlands, J. P. (1975). In *Digestion and Metabolism in the Ruminant*, pp. 320–332. [I. W. McDonald and A. C. I. Warner, editors]. Armidale, Australia: University of New England Publishing Unit.
- MacRae, J. C. & Ulyatt, M. J. (1974). *Journal of Agricultural Science, Cambridge* **82**, 309–319.
- Thomson, D. J., Beever, D. E., Haines, M. J., Cammell, S. B., Evans, R. T., Dhanoa, M. S. & Austin, A. R. (1984). *Journal of Dairy Research* **52**, 17–31.
- Thomson, D. J., Beever, D. E., Harrison, D. G., Hill, I. W. & Osbourn, D. F. (1971). *Proceedings of the Nutrition Society* **30**, 14A.
- Ulyatt, M. J. & Egan, A. R. (1979). *Journal of Agricultural Science, Cambridge* **92**, 605–616.
- Ulyatt, M. J. & MacRae, J. C. (1974). *Journal of Agricultural Science, Cambridge* **82**, 295–307.
- Verite, R., Remond, B. & Journet, M. (1984). *Canadian Journal of Animal Science* **64**, Suppl., 328–329.