The degradation of different protein supplements in the rumen of steers and the effects of these supplements on carbohydrate digestion

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1. Four steers with simple rumen and abomasal cannulas were given diets consisting of ground and pelleted alkali-treated straw, rolled barley and tapioca supplemented with urea (diet U) or containing single-cell protein (diet SCP), maize–gluten meal (diet MGM) or rapeseed meal (diet RSM) in place of some of the tapioca. The isoenergetic diets were given in a 4×4 Latin square design in eight feeds/d at 3-h intervals and provided sufficient metabolizable energy to support a growth rate of approximately 0·5 kg/d. Chromic oxide and polyethylene glycol were given as markers and appropriate samples taken from the rumen and abomasum. Flows (g/d) at the abomasum of organic matter and nitrogenous and carbohydrate constituents were calculated.

2. Rumen ammonia levels were similar with all three protein supplements at about 9 mmol/l, which was significantly lower ($P < 0.05$) than that in animals on diet U (16 mmol/l). Rumen liquid outflow rates (/h) were 0·099, 0·139, 0·125 and 0·160 for diets U, SCP, MGM and RSM respectively; the difference between diet U and diet RSM was significant ($P < 0.05$). Corresponding values for Cr_2O_3 outflow rates were 0·027, 0·032, 0·027 and 0·030/h respectively, which did not differ significantly from each other.

3. RNA, ^{35}S and diaminopimelic acid (DAP) were used as microbial markers. Efficiencies of microbial-N (MN) synthesis, expressed as g MN/kg apparently digestible organic matter, truly digestible organic matter or carbohydrate fermented, were generally not significantly affected by the diet and averaged 29, 22 and 29 respectively based on mean RNA and ^{35}S markers. Corresponding values derived from DAP of 22, 16 and 21 g MN/kg respectively were all significantly ($P < 0.001$) lower. Using ^{35}S as microbial marker, MN flows at the abomasum as a proportion of non-ammonia-nitrogen flow were 0·78, 0·64, 0·51 and 0·78 for diets U, SCP, MGM and RSM respectively. Derived true rumen degradability values (g/g intake) of the total dietary N were 0·91, 0·79, 0·69 and 0·90 for diets U, SCP, MGM and RSM respectively. Protein supplement degradabilities for single-cell protein, maize-gluten meal and rapeseed meal were 0·73, 0·51 and 0·98 respectively.

4. Mouth-to-abomasum digestibility coefficients of the main neutral-sugar components of dietary polysaccharides were 0·68, 0·63 and 0·61 for arabinose, xylose and cellulose-glucose on diet U. These values were generally significantly increased with protein supplementation, but to different extents depending on the source. Maximum digestibility values of 0·81, 0·79 and 0·76 were obtained for arabinose, xylose and cellulose-glucose with diet RSM. Starch-glucose digestibility was high (0·90) on all diets and unaffected by supplementation.

Protein-rationing systems for ruminants have been put forward in a number of countries over recent years (Kaufmann, 1977; Verite *et al.* 1979; Agricultural Research Council, 1980, 1984; Madsen, 1985). Of prime importance in all these systems is the need to predict microbial protein synthesis and the extent of degradation of feed protein with different diets. Part of the purpose of the present study was to examine how these factors varied when diets based on proteins of widely different kinds were used. However, another matter of great importance is the influence of dietary nitrogen on carbohydrate digestion in the rumen. There is evidence that not only the amount of available N but also the form of the N influences carbohydrate and particularly fibre digestion in the rumen (McAllan & Smith, 1983*b*; McAllan & Griffith, 1987).

The present experiments were undertaken to determine the in vivo degradability of three markedly different proteins (with respect to nature, solubility, etc.) derived using different microbial markers and to study the effects of those proteins on the digestion of dietary carbohydrates.

METHODS
Animals, diets and collection of digesta

The experiment was designed as a 4×4 Latin square. Four Friesian steers were used, each fitted with simple rumen and abomasal cannulas at approximately 13–15 weeks of age as described by Smith & McAllan (1970). Approximately 10–12 weeks were allowed for recovery from surgery, and at the beginning and end of the experiment respectively the animals had mean weights (kg) of 142 (SE 2·8) and 190 (SE 2·5) and mean ages (weeks) of 25·2 (SE 0·6) and 38·0 (SE 0·6).

The animals were given four isoenergetic diets calculated to support a growth rate of approximately 0·5 kg/d (Ministry of Agriculture, Fisheries and Food, 1975). The diets consisted of approximately equal portions of ground and pelleted alkali-treated straw (Viton; BOCM Silcock Ltd, Basingstoke) and a concentrate mixture of rolled barley and tapioca. One diet was supplemented with urea and casein (diet U) and the other three diets had part of the tapioca replaced with a single-cell protein isolate (diet SCP), maize–gluten meal (CPC, Manchester; diet MGM) or Canadian rapeseed meal (Tower, 00; J. Bibby Agriculture Ltd, Fromingham; diet RSM). Details of the diets and intakes are given in Table 1. All diets contained a mineral–vitamin supplement. The straw and barley provided about 38 % of the total-N on all diets. Urea contributed 49 % of total-N intake for diet U and 10 % for the other diets. The protein supplements all supplied 60 % of the total-N intakes of their respective diets. Diets were given in eight equal portions at three-hourly intervals by automatic feeding devices.

Each experimental period lasted for 20 d (except for period 1 where, for technical reasons, the period lasted 30 d) at the end of which time the diets were changed. In each period from day 7 (day 17, period 1), 25 ml of a solution containing 7·2 g polyethylene glycol (molecular weight 4000; PEG), 61·5 μCi^{35}S and 115 mg sodium sulphate was added to each of the eight equal portions of feed daily. Paper impregnated with chromic oxide was also mixed in with the diet at a rate of approximately 1·8 g $Cr_2 O_3$/kg dry matter (DM). On day 13 (day 23, period 1) samples (approximately 300 g) of abomasal digesta were taken immediately before and after a feed and mid-way between two feeds. Individual samples were homogenized, combined on an equal weight basis, homogenized once again and stored for subsequent analysis. On day 15 (day 25, period 1) samples of rumen digesta (approximately 400 g) were taken immediately before and after a feed and mid-way between two feeds and combined. Samples of mixed rumen protozoa and mixed rumen bacteria were separated as described by Cockburn & Williams (1984) and Smith & McAllan (1974) respectively. A subsample of the isolated bacterial and protozoal fractions (about 25 g) was removed and extracted immediately for RNA estimation (McAllan & Smith, 1969) and the remainder stored for subsequent analysis. From day 16 (day 26, period 1) marker additions (PEG, ^{35}S and Cr_2O_3) were stopped and small rumen samples (approximately 50 ml) were taken at frequent intervals over the following 72 h and stored for the estimation of marker disappearance rate. Throughout each experimental period small samples of rumen digesta were taken every 2 or 3 d and ciliate protozoa numbers were estimated according to Warner (1962).

In each period samples of rumen digesta (about 400 g) were taken from one steer at the start of the marker infusion about 1·5 h after a feed and then daily at the same time for the 7 d of ^{35}S feeding. Rumen bacteria were separated as described previously and used to confirm establishment of equilibration of ^{35}S uptake. On other days (days 16–18), small samples of rumen digesta were removed just before a feed and 0·5, 1·5 and 2·5 h after a feed. pH was measured immediately and the sample was then acidified and used to determine ammonia concentrations.

Table 1. *Amounts of the main dietary components (kg dry matter/d), nitrogen (g/d) and metabolizable energy (ME, MJ/d) given to steers weighing 135–149 kg**

Diet… Component	U	SCP	MGM	RSM
Alkali-treated straw pellets	1·17	1·17	1·17	1·17
Rolled barley	1·01	1·01	1·01	1·01
Tapioca	0·39	0·10	0·14	—
Maize–gluten meal	—	—	0·32	—
Single-cell protein	—	0·30	—	—
Rapeseed meal	—	—	—	0·56
Urea	0·074	0·015	0·015	0·015
Casein	0·058	0·034	0·034	0·034
Nitrogen	70·8	74·2	74·2	74·6
Estimated† rumen-degradable N (RDN)	64·1	58·3	52·5	66·8
ME‡	28·8	29·1	28·7	30·9
RDN:ME	2·23	2·00	1·83	2·16

* These amounts were increased by 5% for each succeeding 15 kg weight range.

† Estimated from degradabilities obtained in the present experiment by using ^{35}S (see Table 4).

‡ Calculated from energy values (MJ/kg) for individual components of: alkali-treated straw 7·3, rolled barley 12·9, tapioca 15·0, casein 15·0, maize–gluten meal 15·0, single-cell protein 15·1, rapeseed meal 10·9.

Analytical and calculations

DM, organic matter (OM) and α–ϵ–diaminopimelic acid (DAP) were determined as described by Smith *et al.* (1978). Total-N (TN) and ammonia-N were estimated by the procedures of Smith & McAllan (1970) and Merry (1980) respectively. Urea-N was estimated colorimetrically by the procedure of Technicon Instruments Corporation (1967). Cr_2O_3 was determined according to Williams & Smith (1974) and PEG by the method of Smith & McAllan (1971). Monosaccharide components of feedstuffs, bacteria and digesta samples were analysed as described by McAllan & Smith (1974). RNA was estimated as described by McAllan & Smith (1969) and ^{35}S by the method of Mathers & Miller (1980).

Daily flows of digesta constituents were calculated using the dual-phase-marker procedure of McAllan & Smith (1983*b*); average Cr_2O_3:PEG values in abomasal digesta were 0·195 (SE 0·0127) (*n* 16) which were 1·187 (SE 0·067) (*n* 16) of the ratios in the feeds. Marker (^{35}S, RNA and DAP):N values were established in microbial samples from each steer on each diet and the proportion of non-NH$_3$-N (NAN) in abomasal digesta which was of microbial origin was estimated according to McAllan & Smith (1983*a*). Apparently digested organic matter (ADOM) was measured as the difference between OM intake and OM flow at the abomasum (g/d). Truly digested organic matter (TDOM) was measured as ADOM + microbial organic matter (MOM) flow. MOM flow was estimated as:

$$\text{MOM flow at abomasum} = \frac{\text{OM}}{\text{N}} \text{ (bacterial isolates)} \times \text{MN flow at the abomasum,}$$

OM:N ratios were established in bacterial isolates from each steer on each diet, MN is microbial-N.

Degradability of protein in total diets was estimated as:

$$\frac{\text{Total NAN flow} - \text{MN flow} + \text{endogenous-N (EN) flow,}}{\cdot \text{ total NAN intake}}$$

Table 2. *Mean ciliate protozoa numbers, ammonia-nitrogen concentration (mmol/l) and pH values in rumen digesta from steers receiving diets containing different protein supplements*, together with fractional outflow rates of polyethylene glycol (PEG) and chromic oxide from the rumen over 24 and 48 h respectively. Also shown are* $^{35}S:N$ *values (disintegrations/min (dpm) per mg N) in samples of mixed bacteria and mixed protozoa isolated from rumen digesta taken 9 d after the start of infusion of* ^{35}S *into the rumen*

(Results are mean values with their standard errors for four steers unless otherwise stated)

Diet...	U		SCP		MGM		RSM	
	Mean	SE	Mean	SE	Mean	SE	Mean	SE
Protozoa ($\times 10^5$/ml)‡	16·4	1·03	18·4	1·47	16·2	1·42	16·3	3·18
NH_3-N (mmol/l)	16·4	2·57	9·2	1·46	10·6	2·42	8·0	0·95
pH	6·58	0·035	6·40	0·036	6·40	0·045	6·57	0·057
Fractional outflow (/h): PEG	0·10	0·018	0·14	0·030	0·13	0·034	0·16	0·019
Cr_2O_3	0·03	0·002	0·03	0·003	0·03	0·005	0·03	0·004
$^{35}S:N$ (dpm/mg) ($\times 10^{-3}$): Bacteria	8·8	0·52	6·6	0·32	6·8	0·30	4·5	0·50
Protozoa	10·7	0·35	6·4	0·49	5·7	0·99	4·5	0·41

U, Straw + barley + urea + casein; SCP, straw + barley + urea + casein + single-cell protein; M, straw + barley + urea + casein + maize–gluten meal; RSM, straw + barley + urea + casein + rapeseed meal.

* For details, see Table 1.

† Only three values.

where an EN flow of 0·032 g/kg live weight per d was assumed (Ørskov & MacLeod, 1983).

In estimating the degradability of the individual protein supplements it was assumed that all the casein supplementary protein and urea-N were degraded in the rumen. Thus surviving feed-N at the abomasum of steers receiving diet U was assumed to derive entirely from the straw and rolled barley components (S + BNAN), the degradability of which was calculated as described previously but where the denominator was straw + barley NAN intake. It was then assumed that the straw + barley protein degradability would not be altered in the other diets thus:

$$\text{supplemented protein degradability} =$$

$$\frac{\text{total NAN flow} - [\text{S} + \text{BNAN intake} \times (1 - \text{S} + \text{BNAN degradability}) + \text{MN flow} + \text{EN flow}]}{\text{supplement protein NAN intake}}$$

The statistical analysis used was standard analysis of variance appropriate to the Latin square design (Cochran & Cox, 1962) taking account of variation between calves, periods and rations. The significance of differences between ration means was tested using standard errors based on the error squares with 6 df. Supplemental protein degradability values which did not fit the Latin square model were compared by paired t test (Snedecor & Cochran, 1972) as were some comparisons of values between markers.

RESULTS

Rumen characteristics

Numbers of ciliate protozoa in rumen digesta remained fairly constant throughout the experiment (Table 2). Numbers in digesta from animals receiving single-cell protein (diet SCP) tended to be greater than those receiving the other diets but the difference was not significant. The incorporation of ^{35}S into rumen bacterial and protozoal protein had

Table 3. *Mean amounts (g/d) of dietary nitrogen consumed and digesta-N components flow at the abomasum of steers receiving diets containing different protein supplements**

(Results are mean values with their standard errors for four steers)

Diet...	U		SCP		MGM		RSM	
	Mean	SE	Mean	SE	Mean	SE	Mean	SE
Total-N intake (g/d)	76·2	2·45	81·0	3·1	81·8	3·17	80·9	2·89
Abomasal flows (g/d)								
Ammonia-N	2·26	0·81	2·50	0·34	2·56	0·63	3·00	0·42
Non-ammonia-N	54·8	5·0	61·8	1·8	67·5	3·4	59·8	5·2
RNA-N	3·87	0·23	4·11	0·40	3·23	0·34	4·26	0·53
DAP-N	0·142	0·020	0·117	0·007	0·103	0·016	0·117	0·016
Microbial-N: RNA†	41·1	2·4	43·7	4·2	34·3	3·6	45·3	5·6
^{35}S	42·5	1·7	39·5	1·8	34·2	2·1	47·3	3·7
DAP	35·9	5·1	29·5	1·7	25·9	4·0	29·6	3·5

U, Straw + barley + urea + casein; SCP, straw + barley + urea + casein + single-cell protein; MGM, straw + barley + urea + casein + maize–gluten meal; RSM, straw + barley + urea + casein + rapeseed meal; DAP–N, α–ϵ-diaminopimelic acid-N.

* For details, see Table 1.

† Calculated from measured RNA flows 'adjusted' ($\times 0\cdot85$) to correct for surviving feed RNA (Smith *et al.* 1978).

reached maximum levels by the fourth day after the start of infusing ^{35}S with all diets and remained essentially constant over the remainder of the infusion period. Relations (paired *t* test comparisons) found between ^{35}S and NAN in mixed rumen bacteria and protozoa showed that significantly ($P < 0\cdot05$) more ^{35}S was incorporated into both bacteria and protozoa in animals receiving the urea diet (diet U). Incorporation of ^{35}S into the microbes of animals receiving diets containing rapeseed meal (diet RSM) tended to be lower than in those animals receiving diets SCP or MGM, but the difference was only significant ($P < 0\cdot05$) between rumen bacteria from animals receiving diet MGM. Mean rumen pH values are also presented in Table 2. There was little between-feeding variation within diets (maximum spread 0·2 units) and no significant differences between values obtained with different diets. There was also little between feeding variation in rumen ammonia levels which remained above 5 mmol/l at all times, but in comparing mean values (Table 2) it was found that levels in rumen digesta from animals receiving diet U were significantly greater ($P < 0\cdot05$) than in digesta from animals receiving the other diets. PEG and Cr_2O_3 rumen outflow rates are also presented in Table 2. There were no significant differences between Cr_2O_3 rumen outflow rates on any diet. PEG rumen outflow rates tended to be lower with diet U compared with the other diets but this was only significant ($P < 0\cdot05$) compared with diet RSM.

Nitrogenous constituent flow at the abomasum and feed-protein degradability

Abomasal flows of various nitrogenous constituents are presented in Table 3. Urea-N was not detected in any digesta samples. There were no significant differences between diets on the amounts of NH_3-N passing the abomasum/d. NAN flows at the abomasum were, on average, about 0·75 g/g NAN intake. In paired *t* test comparisons, amounts of NAN flowing at the abomasum of steers receiving diets SCP, MGM or RSM were greater than for those receiving the diet U. These differences approached significance for diet SCP ($P < 0\cdot10$) and were significant ($P < 0\cdot01$) for diet MGM. RNA-N flows tended to be higher with diets SCP and RSM and lower with diet MGM compared with the basal diet but the

Table 4. *Mean estimates (g/g intake) of total feed-nitrogen, protein-supplement-N and basal-component-N degraded between mouth and abomasum of steers receiving diets containing different protein supplements*

(Results are presented for values derived using RNA, ^{35}S or DAP as microbial markers)

Microbial marker...	RNA		^{35}S		DAP	
	Mean	SE	Mean	SE	Mean	SE
Protein degradability†						
Complete feed						
Diet U	0·89	0·030	0·91	0·027	0·82	0·080
Diet SCP	0·84	0·048	0·79	0·012	0·66	0·012
Diet MGM	0·66	0·030	0·69	0·023	0·58	0·053
Diet RSM	0·88	0·047	0·90	0·031	0·68	0·022
Protein supplement‡						
Single-cell protein	0·88	0·027	0·73	0·050	0·64	0·025
Maize–gluten meal	0·55	0·013	0·51	0·053	0·47	0·099
Rapeseed meal	0·96	0·056	0·98	0·056	0·66	0·055
Basal ingredients§						
Straw–barley	0·71	0·055	0·76	0·054	0·54	0·076

U, Barley + straw + urea + casein; SCP, barley + straw + urea + casein + single-cell protein; MGM, barley + straw + urea + casein + maize–gluten meal; RSM, barley + straw + urea + casein + rapeseed meal; DAP, α–ε–diaminopimelic acid.

* For details, see Table 1.
† Corrected for an assumed endogenous-N secretion of 0·032 g/d per kg live weight.
‡ Corrected for surviving basal-feed-N at abomasum derived from each individual marker.
§ Assuming urea and casein are completely degraded in the rumen.

differences were not significant. No significant differences were found in the amounts of DAP-N passing the abomasum/d related to diet. MN flows calculated using RNA as a microbial marker were not significantly different ($P < 0.05$) between diets. When ^{35}S was used as microbial marker significantly lower MN flows ($P < 0.05$) were found for diet MGM compared with diets U and RSM, and for diet SCP compared with diet RSM. MN flow as a proportion of NAN flow (based on ^{35}S) was 0·53, 0·64, 0·78 and 0·78 for diets MGM, SCP, RSM and U respectively. There were no significant differences between MN flows with different diets calculated using DAP as a microbial marker, but these were significantly ($P < 0.001$) lower by about 25 % than flows calculated using either RNA or ^{35}S.

Feed-protein degradability values, between mouth and abomasum are presented in Table 4. Comparisons of total feed-N degradability within any one marker showed that diet MGM-N was significantly ($P < 0.05$) less degraded than N in the other three diets. Diet SCP-N degradability was also found to be significantly less ($P < 0.05$) compared with diets U and RSM with both ^{35}S and DAP as microbial markers but not with RNA. For the individual protein supplement N degradability (also shown in Table 4) values derived using ^{35}S for maize–gluten and rapeseed were not significantly different from those derived using RNA. Single-cell-protein-N degradability values derived using RNA however, were significantly ($P < 0.05$) higher than those using ^{35}S. Comparison of N degradability values obtained using different markers showed that apart from the single-cell-protein supplement, there were no significant differences between results obtained using either ^{35}S or RNA. However, apart from the value for complete feed-N degradability of diet U, all other N degradabilities derived using DAP were significantly lower than the corresponding values derived from either ^{35}S or RNA.

Table 5. *Mean microbial-nitrogen yields in terms of amounts of organic matter (OM)* *apparently (ADOM) or truly (TDOM) digested in the rumen or of total carbohydrate* *digested in the rumen of steers receiving diets containing different protein supplements**

(Results are mean values with their standard errors for four steers)

Diet...	U		SCP		MGM		RSM	
	Mean	SE	Mean	SE	Mean	SE	Mean	SE
Microbial-N yield (g/kg ADOM)								
Based on: RNA	29·4	2·2	32·5	3·8	25·1	3·2	30·5	2·8
^{35}S	31·5	2·0	29·3	2·0	24·7	0·9	32·0	2·4
DAP	26·0	4·4	21·9	1·9	19·1	3·3	19·8	1·9
Microbial-N yield (g/kg TDOM)†								
Based on: RNA	21·8	1·6	24·2	2·5	19·6	2·5	22·3	2·0
^{35}S	22·5	0·6	21·9	1·2	19·2	0·5	23·3	1·3
DAP	19·2	3·2	16·3	1·2	14·9	2·6	14·5	1·4
Microbial-N yield (g/kg carbohydrate fermented in the rumen)								
Based on: RNA	27·6	2·4	32·8	4·4	24·3	2·9	31·9	4·7
^{35}S	28·6	1·4	29·7	2·1	24·2	1·8	33·3	2·3
DAP	24·2	4·3	22·2	1·9	18·4	3·3	20·9	3·4

U, Straw + barley + urea + casein; SCP, straw + barley + urea + casein + single-cell protein; MGM, straw + barley + urea + casein + maize–gluten meal; RSM, straw + barley + urea + casein + rapeseed meal; DAP, α, ϵ-diaminopimelic acid.

* For details, see Table 1.

† Corrected for microbial OM contribution at the abomasum.

Estimates of total feed-N degradabilities calculated without correction for an EN contribution were 7·5, 6·6 and 8·8 % units less than the corrected values shown derived from ^{35}S, RNA and DAP respectively and the differences were all significant ($P < 0.001$). Uncorrected estimates of the basal-components (straw + barley) N degradability were 0·593, 0·545 and 0·371 derived using ^{35}S, RNA and DAP respectively. These were all significantly lower ($P < 0.001$) than the corrected values shown. Protein supplement degradability values uncorrected for an EN contribution were very similar to the corrected values shown.

Efficiency of microbial protein synthesis

OM intakes and the proportions of OM apparently (ADOM) or truly (TDOM) digested between mouth and abomasum are shown in Table 5. There were no significant differences in intakes or ADOM between diets. Values for TDOM on the other hand showed significantly greater digestion by paired t test with diet RSM compared with diet U ($P < 0.10$); diet SCP ($P < 0.05$) or diet MGM ($P < 0.02$). MN yields (Table 5), expressed in terms of ADOM were not significantly different when derived using either RNA or DAP as microbial marker. With ^{35}S, MN yields were significantly lower for diet MGM compared with diets U and SCP ($P < 0.10$) or diet RSM ($P < 0.05$). When efficiencies were expressed in terms of TDOM, no significant differences were found between diets using either RNA or DAP as microbial markers and with ^{35}S the only significant difference found ($P < 0.05$) was a lower efficiency with diet RSM compared with diet MGM. Table 5 also shows the efficiencies of MN synthesis in terms of carbohydrate fermented in the rumen. Values obtained for diet MGM were found to be significantly lower than those obtained for diets SCP and RSM with either RNA or ^{35}S as microbial markers. No other significant differences were found. Efficiencies derived using DAP as microbial marker were

Table 6. *Carbohydrate composition of feed components (g/kg dry matter); amounts of neutral sugars released by hydrolysis with 0·5 M-sulphuric acid (ribose, rhamnose, mannose, arbinose, xylose and α-linked glucose (starch) and 13 M-sulphuric acid (β-linked glucose (cellulose))*

(Results are mean values with their standard errors for four samples)

Component	Rhamnose		Ribose		Mannose		Arabinose		Galactose		Xylose		α-Linked glucose (starch)		β-Linked glucose (cellulose)	
	Mean	SE	Mean	SE	Mean	SE	Mean	SE	Mean	SE	Mean	SE	Mean	SE	Mean	SE
Barley	—	—	1·80	0·05	3·24	0·31	30·7	0·48	24·1	0·12	57·8	1·3	542	14	60·0	0·6
Straw pellets	—	—	3·89	0·42	2·57	0·17	29·6	1·12	9·31	0·90	165	7·1	26·1	0·5	379	4·0
Single-cell protein	4·26	0·07	17·0	1·21	—	—	—	—	3·00	0·10	—	—	68·4	0·9	—	—
Maize–gluten meal	—	—	—	—	2·08	0·41	5·84	0·35	—	—	5·95	0·06	184·4	15·1	7·95	0·35
Rapeseed meal	2·47	0·34	2·82	0·02	2·55	0·07	60·2	0·20	35·6	0·8	22·4	0·20	80·1	1·5	82·5	1·70

Table 7. *Daily intakes (g/d), abomasal flows (g/d) and apparent rumen digestibilities (g/g intake) of the major dietary carbohydrates together with mean amounts of organic matter (OM) consumed (kg/d), flowing to the abomasum (kg/d) and the proportions of OM intake apparently (ADOM) and truly (TDOM) digested between mouth and abomasum of steers given diets containing different protein supplements**

(Results are mean values with their standard errors for four steers)

Diet...	U		SCP		MGM		RSM	
Component	Mean	SE	Mean	SE	Mean	SE	Mean	SE
OM								
Intake	2·68	0·12	2·65	0·13	2·66	0·14	2·77	0·14
Flow at abomasum	1·27	0·08	1·29	0·02	1·28	0·09	1·29	0·08
ADOM	0·53	0·019	0·52	0·029	0·52	0·022	0·54	0·03
TDOM†	0·71	0·028	0·69	0·017	0·67	0·018	0·73	0·018
Galactose								
Intake	38	1·2	39	1·2	41	1·8	59	1·9
Flow at abomasum	10	1·6	9	2·4	8	0·5	10	0·3
Proportion digested in the rumen	0·73	0·04	0·77	0·06	0·80	0·02	0·83	0·01
Arabinose								
Intake	71	2·2	71	2·2	73	2·3	107	3·4
Flow at abomasum	23	0·6	21	2·5	16	0·9	20	0·9
Proportion digested in the rumen	0·68	0·00	0·70	0·04	0·78	0·01	0·81	0·01
Xylose								
Intake	272	8·4	272	8·4	274	8·5	285	8·8
Flow at abomasum	101	3·7	89	5·9	66	1·7	60	2·3
Proportion digested in the rumen	0·63	0·09	0·67	0·02	0·76	0·01	0·79	0·01
Cellulose–glucose								
Intake	544	16·7	544	17·1	547	16·8	593	18·3
Flow at abomasum	210	15·0	182	7·6	138	5·2	145	15·0
Proportion digested in the rumen	0·61	0·02	0·67	0·02	0·75	0·00	0·76	0·02
Starch–glucose								
Intake	1002	31·3	779	24·4	783	25·3	673	21·6
Flow at abomasum	101	10·5	76	1·9	79	6·0	68	11·6
Proportion digested in the rumen	0·90	0·01	0·90	0·00	0·90	0·01	0·90	0·02

U, Barley + straw + urea + casein; SCP, barley + straw + urea + casein + single-cell protein; MGM, barley + straw + urea + casein + maize–gluten meal; RSM, barley + straw + urea + casein + rapeseed meal.

† Corrected for microbial OM flow at the abomasum.

* For details, see Table 1.

significantly ($P < 0.001$) lower than those obtained with either ^{35}S or RNA however they were expressed.

Carbohydrate disappearance between mouth and abomasum

The carbohydrate composition of the different feed components are given in Table 6. Appreciable amounts of starch–glucose were found in the maize–gluten meal (184 g/kg DM) and considerable amounts of structural sugars in the rapeseed meal (about 200 g/kg DM). Daily intakes, abomasal flows and apparent rumen digestibilities of the main dietary carbohydrates are presented in Table 7. Of the other minor sugars measured but not

reported in detail, intakes of mannose were similar with all diets at about 7·5 g/d, of which approximately 0·60 disappeared between mouth and abomasum. Ribose intakes of about 12 g/d were found with diet SCP and about 7 g/d with the other diets, all of which had an apparent digestibility of about 0·60. Intakes of rhamnose with all diets were less than 1 g/d and there were net gains of 4–6 g/d between mouth and abomasum. Intakes of galactose, arabinose, xylose and cellulose–glucose were similar between diets, U, SCP and MGM but were all higher with diet RSM. There were no significant differences in abomasal flows of galactose but rumen digestibility was greater with the protein-supplemented diets compared with diet U, but the difference was only significant for diet RSM. Flows of arabinose at the abomasum of steers receiving diet MGM were lower than those with the other diets. Rumen digestibilities were significantly greater ($P < 0.05$) with diets RSM and MGM compared with diets U or SCP. There were significant differences in the amounts of xylose and cellulose passing the abomasum with different diets. Diet RSM supported the highest rumen degradabilities of xylose and cellulose and these were significantly greater ($P < 0.05$) than those obtained with either diets U or SCP. There were no significant differences between diets MGM or RSM but digestibilities with diet MGM were significantly greater than those with diets U or SCP.

There were expected differences between starch intake of the basal diet compared with the other diets as a result of replacement of some tapioca with protein supplement and this was reflected in abomasal flows. However, mouth to abomasal digestibilities of starch were similar with all diets.

DISCUSSION

The use of chronically fistulated animals is the most realistic means of studying synthetic and degradative processes in the rumen under conditions approaching those found in practice. These types of experiment are extremely intensive and very difficult to perform with both physical and practical limitations on the number of animals which can be used in any one experiment. A Latin square design was deemed to be the best choice in the present experiments because of the small numbers of animals used. The limitations of this design with respect to degrees of freedom, different randomizations, etc. are fully recognized.

In the present experiments, no urea was detected in any abomasal digesta. It seems certain also that the casein in the basal diet (diet U) would have been effectively degraded before digesta left the rumen (McDonald, 1954; Williams & Smith, 1976), although it would transiently have provided amino acids and peptides in addition to those coming from microbial turnover. Thus it is reasonable to assume that virtually all the non-microbial NAN entering the duodenum of animals receiving diet U came from straw pellets and barley plus endogenous material and that the NAN additional to this amount passing the duodenum of animals receiving protein-supplemented diets was of supplement origin. Calculation in this way of undegraded supplement N is not dependent on any estimate of EN although it does assume that this remains constant between diets. On the other hand degradability values for whole diets calculated simply by deducting an estimate of MN from the total NAN flow at the duodenum or abomasum are based on an assumption that EN is negligible. This is generally recognized to be untrue but correction depends on using an assessed value for EN. In the values given in Table 4 we have assumed a value of 32 mg/kg live weight per d (Ørskov & MacLeod, 1983) and, taking this into account, EN had a substantial effect on calculated degradability; on average it increased values for whole diets by 7% points and values for residual basal constituents by about 15% points. Other experiments have indicated that EN for cattle is variable and may be greater than this (Ørskov et al. 1986) and other workers have sometimes calculated degradability values

using assumed EN values more than twice those used in Table 4 (Amaning-Kwarteng *et al.* 1986). It appears that values calculated by this method should be viewed with caution.

Problems in the estimation of MN flows by different procedures and their use in deriving protein degradabilities have been pointed out (MacRae & Reeds, 1980). Our results using DAP as a marker were always significantly lower than those derived using the other markers (Table 4). This was presumably mainly a result of protozoal-N contribution at the abomasum. Values derived using RNA as microbial marker were very similar to those using ^{35}S except for diet SCP. This was probably because single-cell protein had a very high RNA content (RNA–N:TN was 0·13) and some of this RNA survived the rumen to be attributed to rumen microbial RNA. This would have resulted in an overestimation of single-cell protein degradability.

A number of workers have reported degradability values for maize–gluten meal. Thus Stern *et al.* (1983) used lactating dairy cows which were given, in four meals/d, a series of diets composed of approximately equal proportions of concentrates and roughages to which various levels of maize–gluten meal had been added. Degradability of the maize–gluten meal was calculated by regression of duodenal amino acids on amino acid intake and showed that 43 % of the maize–gluten meal was degraded in the rumen. Other experiments with growing steers receiving roughage–concentrate diets (40:60, w/w) supplemented with maize–gluten meal and using RNA to measure MN indicated the percentage of maize–gluten meal intake degraded in the rumen to be 39 (Zinn *et al.* 1981), while a recent study with steers receiving roughage–concentrate (20:80, w/w) diets (Garrett *et al.* 1987) showed a value of 58. Our own value for the percentage of maize–gluten meal degraded in the rumen of 51 was consistent with these estimates.

Very little information obtained in vivo appears to exist concerning the degradability in the rumen of single-cell protein or rapeseed meal. Hagemeister & Kaufmann (1974) in experiments with milking cows obtained degradability values of 60 and 58 % for rapeseed meal and single-cell protein respectively. However, these values were derived using DAP as a 'microbial' marker and were from animals in which duodenal digesta were collected posterior to the entry of the bile and pancreatic ducts, so that EN estimates would have been elevated. Both these factors would have resulted in underestimation of dietary protein degradability. More recently Rooke *et al.* (1983), in experiments with heifers receiving silage-based diets given twice daily and using ^{35}S as microbial marker, obtained a mean degradability value for rapeseed-meal-supplement protein of 77 %. The only other information available for these supplements appears to be that based on the 'in rumen' porous synthetic fibre (psf) bag method. For single-cell protein, predicted values ranged from 46 to 72 % depending on the liquid fractional outflow rate assumed (Teller *et al.* 1985). A number of studies have been made for rapeseed meal using the psf bag method and have given values ranging from 68 to 87 % (Agricultural Research Council, 1984; Madsen & Hvelplund, 1985; Ørskov *et al.* 1981) with values again being markedly influenced by assumed outflow rates (Ørskov *et al.* 1981).

Thus, while our ^{35}S-based degradability value for single-cell protein of 73 % is not inconsistent with the small amount of published information, our value of 98 % for rapeseed meal is very high. The reason for this is not clear, particularly as the liquid outflow rates for rapeseed meal in the present experiments were relatively high, about 0·16/h. It seems unlikely that the frequent feeding used in the present experiments could have been responsible (Robinson & Sniffen, 1985) and presumably the high degradability was simply a characteristic of the particular sample of rapeseed meal used. Certainly rapeseed meal solubility can vary considerably with preparation procedure (Laycock & Miller, 1981).

The mean efficiency of microbial protein synthesis obtained in these experiments of approximately 30 g MN kg ADOM (rumen; derived from RNA and ^{35}S) was similar to

that reported as a mean for a wide variety of diets (Agricultural Research Council, 1980) and found in a number of more recent studies (e.g. Meyer *et al.* 1986). There appeared to be a tendency for reduced efficiency with diet MGM as reported also by Garrett *et al.* (1987), but this was not generally significant. Efficiency values in the present work were appreciably higher than those (14–20) reported previously for steers receiving similar diets based on ground and pelleted alkali-treated straw and rolled barley with a variety of protein supplements (McAllan *et al.* 1986). It was suggested for the earlier work that the low efficiencies observed may have been causally related to the inclusion of sodium hydroxide-treated straw in the diets, but this was not supported by the present findings. The apparent conflict may have been because in the present experiments diets were given eight times daily compared with only twice daily in the earlier work and it has been shown that increased frequency of feeding may result in increased efficiency (McAllan *et al.* 1987; Tamminga, 1979, 1982). Although, as for the values quoted previously, ADOM (rumen) is commonly used as a measure of energy supply when assessing the efficiency of microbial protein synthesis (e.g. Agricultural Research Council, 1980, 1984), some workers have suggested that carbohydrate digestion in the rumen might form a more appropriate basis for the calculation (Madsen, 1985). The results given in Table 4 indicated, however, that estimates of ADOM (rumen) corresponded closely to those of total carbohydrate disappearance. This would not necessarily apply under all conditions but the finding suggests that for commonly used ruminant diets it would make little practical difference in a feeding system which of these bases were used.

Digestibility values between mouth and abomasum obtained for fibre fractions of the diets used in the present experiments (0·63–0·83 for hemicellulose sugars and 0·61–0·76 for cellulose) were broadly in the range found in earlier experiments with growing steers given diets based on alkali-treated straw and barley and supplemented with protein (McAllan & Griffith, 1987). A number of previous studies both in vitro (Vestergaard-Thomsen, 1985) and in vivo (McAllan & Smith, 1983*b*; McAllan & Griffith, 1987) have demonstrated that fibre digestion in the rumen varied with the kind, as well as the amount, of dietary protein and this was shown also in the present work. In the earlier studies diets containing a rapidly degraded dietary N supplement generally supported less fibre digestion than those containing more resistant dietary protein, and it was postulated that one reason for the difference may have been that a slow release of peptides or amino acids was beneficial to cellulolytic and hemicellulolytic activity.

This hypothesis received support in the present study when diet SCP was compared with diets U and MGM which contained more and less rapidly degradable 'protein' respectively. Xylose, arabinose and β-linked (cellulose) glucose rumen digestibilities were all ranked in the order diets MGM > SCP > U with many of the differences being significant (Table 7). The hypothesis is not, however, supported when diet RSM is considered. This supplement was found to be degraded as completely as the 'protein' in diet U and yet, in diets containing it, fibre digestion was little different to that seen with diets containing the resistant maize–gluten-meal supplement. Thus the results provide confirmation that different protein sources facilitate fibre digestion to different extents, but go against the ideas either that this was due to differences in NH_3 release (NH_3 concentration was highest with diet U) or simply to differences in degradability. However, it has been shown in vitro (Vestergaard-Thomsen, 1985) that cellulolytic microbes may need a supply of long-chain peptides for optimal growth and differences between protein sources in sustaining fibre digestion may reflect differences in the pattern of breakdown of these proteins in the rumen.

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