Utilization of low quality roughages: effects of urea and protein supplements of differing solubility on digesta flows, intake and growth rate of cattle eating oaten chaff

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1. Expt 1. Five 150 kg steers with ruminal, abomasal and ileal cannulas were given 3000 g oaten chaff daily plus pelleted supplement with no added nitrogen (diet A) or 50 g N/d as urea (diet B), casein (diet C), casein and formaldehyde-treated casein (HCHO-casein) (50:50 w/w; diet D) and HCHO-casein (diet E), in a 5 × 5 Latin square design. The basal diet and supplement were fed in eight equal increments at intervals of 3 h. Proportions of dry matter and organic matter digested in the stomach and whole tract were greater for diets B, C, D and E than for diet A. Total volatile fatty acid levels in the rumen and the proportion of acetic acid were lower, and the proportion of propionic acid higher on diet A than on the other diets. Rumen ammonia levels were lower on diets A, D and E than on diets B and C. N flows at the abomasum, ileum and rectum were lower on diet A than on the other diets; abomasal flows and apparent intestinal absorptions of amino acids were higher on diets D and E than on diets A, B and C. Efficiencies of bacterial protein synthesis were 15, 15, 14, 13 and 12 g bacterial N/kg OM truly digested in the stomach on diets A, B, C, D and E respectively.

2. Expt 2. Forty 300 kg steers were fed oaten chaff *ad lib.* plus twice the amount of the same pelleted supplements as in Expt 1. Intake of oaten chaff was 23 % higher with N supplements (diets B, C, D and E) than without (diet A). Live-weight gains were 356, 798, 843, 842 and 805 g/d on diets A, B, C, D and E respectively.

3. It was concluded that efficiency of bacterial protein synthesis was not limited by the supply of peptides and amino acids in the rumen, and that increases in amino acid availability in the intestines from feeding HCHO-casein did not increase food intake or live-weight gain.

Peptides and amino acids contribute 200–400 mg/g nitrogen incorporated into microbial cells in the rumen (Pilgrim *et al.* 1970; Nolan & Leng, 1972; Nolan *et al.* 1976) and in vitro studies indicated that the optimum value for non-protein-N to amino acid-N for microbial growth was 75:25 (Maeng *et al.* 1976). When casein replaced approximately half the N supplied by urea on a virtually protein-free diet fed to sheep, Hume (1970) found that there was an increased flow of microbial protein from the stomach.

These observations indicate that microbial growth in the rumen of animals eating a low protein diet may be restricted by the supply of peptides and amino acids, with a concomitant reduction in the rate of cellulose digestion in the rumen. Since the rate of digestion and retention time in the rumen are major determinants of the voluntary food intake of low quality roughages (Thornton & Minson, 1973) it is possible that their intake may be limited by the supply of peptides and free amino acids to rumen micro-organisms.

We investigated this possiblity by supplying iso-nitrogenous amounts of urea, soluble protein (casein) or digestible by-pass protein (HCHO-casein) to steers fed oaten chaff.

EXPERIMENTAL

Expt 1

Animals and management. Five Friesian steer calves were fitted with simple cannulas in the rumen, abomasum and terminal ileum at 3 months of age. At 8 months of age and

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Table 1. *Expts 1 and 2. Components (g/kg air dry) and chemical composition (g/kg dry matter (DM)) of five pelleted supplements fed with oaten chaff to cattle together with daily amounts fed in the respective experiments*

	Supplements				
			Casein : formaldehyde-treated casein		
	Control	Urea	100:0	50:50	0:100
Diet ...	A	B	C	D	E
Components					
Molasses	406	361	406	378	350
Oaten chaff	197	175	197	183	180
Maize flour	373	332	—	—	—
Casein	—	—	373	174	—
Formaldehyde-treated casein	—	—	—	243	450
Urea	—	112	—	—	—
Calcium stearate	12	10	12	11	10
DM	919	910	930	925	920
Chemical composition					
Nitrogen	5·7	64·1	66·7	71·1	72·4
Ash	188	158	176	175	154
α-glucose polymers	435	462	54	51	53
Lignin	23	19	24	22	22
Amounts fed daily (g air dry)					
Expt 1	336	378	336	360	387
Expt 2	670	754	670	720	776

live weight 150 kg they were tethered in individual cages fitted with automatic feeders which delivered one-eighth portions of the daily feed at three hourly intervals.

Calves were allocated to the five dietary treatments according to a 5 × 5 Latin square design.

Diets and feeding procedure. The basal diet was oaten chaff containing (g/kg dry matter (DM)) 7·7 N, 228 α-glucose polymers, 51 ash, 47 lignin. The composition of the five pelleted supplements is given in Table 1. HCHO-casein was sprayed at the rate of 100 g formalin solution containing 15 g HCHO/kg casein DM, a concentration of HCHO selected to render the casein completely insoluble in the rumen and partly digestible in the intestines (Hemsley *et al.* 1973). The digestibility of the treated casein was measured in two sheep on constant daily intakes of oaten chaff by introducing 200 g/d over 5 d into the rumen cannulas. Casein digestibility, calculated from the change in faecal N excretion before and after introducing the casein, was 0·80. The daily intakes of diets C, D and E were designed to provide 250 g digestible casein/d. Diet B contained an iso-nitrogenous amount of urea and diets A and B contained maize flour to make them iso-energetic with the other diets. The amounts fed provided equal daily intakes of molasses, oaten chaff and calcium stearate from the five diets (Table 1).

In a preliminary period the five calves were given the basal diet *ad lib.* with pellets of diet A in eight equal portions at 3 h intervals. Consistent intakes of 3 kg oaten chaff/d were recorded for all calves and intakes were kept at this level throughout the experiment. Each dietary period comprised a 2-week adjustment period followed by marker infusion over 8 d on the last 3 d of which digesta collections were made.

Marker infusions and digesta collections. Markers used were the ^{51}Cr complex of ethylene-diaminetetra-acetic acid (^{51}Cr-EDTA) (Downes & McDonald, 1964) and ^{103}Ru-labelled tris-(1,10-phenanthroline)-ruthenium (ii) chloride (^{103}Ru-P) (Tan *et al.* 1971) and lignin. ^{51}Cr-EDTA and ^{103}Ru-P were infused into the rumen at the rate of 113 μCi ^{51}Cr and

23 μc ^{103}Ru daily. Abomasal digesta samples were collected three times daily (0·5, 1·0 and 1·5 h respectively after three successive meals) and bulked; faecal samples were taken at the same times from the most recently voided material. Ileal digesta samples were collected once daily for 3 d and bulked. Digesta samples were stored at $-10°$.

After each collection period, marker infusion was discontinued and eight samples of rumen digesta were taken during the following 30 h. Half of each sample was acidified to pH 2 and all samples were stored at $-10°$.

Radioactivity measurements, chemical analyses and digesta flow calculations. Sub-samples of abomasal and ileal digesta were centrifuged at 2400 *g* for 20 min to obtain liquid-rich fractions and sub-samples of faeces were blended with water to form slurries. These slurry samples and samples of liquid-rich fractions and of total abomasal and ileal digesta were assayed for ^{51}Cr and ^{103}Ru in a scintillation spectrometer (Model 3320; Packard Instruments Pty Ltd, Sydney).

Total digesta and faecal samples were analysed for DM, by drying to constant weight at 80°, OM by ashing at 550° overnight. Sub-samples of total digesta were freeze-dried before analysis for N by a micro-Kjeldahl technique, lignin by the method of Van Soest (1963), α-glucose polymers by the method of Macrae & Armstrong (1968). Total digesta samples were also analyzed for amino acids, including 2,6-diaminopimelic acid, by ion-exchange chromatography using an amino acid Auto-Analyzer TSM (Technicon Instrument Corporation, Tarry Town, New York, USA) following 24 h hydrolysis in 6 M-hydrochloric acid at 136° using norleucine and guanidine as internal standards; corrections were made for losses of amino acids known to occur during this hydrolysis procedure. Liquid-rich digesta fractions were analyzed for DM, OM, N and α-glucose polymers.

Flows of abomasal, ileal and faecal digesta were calculated from the concentrations of two markers, using equations (1)–(3) from Faichney (1975). ^{51}Cr-EDTA was used as the liquid marker throughout. The solid phase marker was ^{103}Ru-P in the first two periods and lignin in the remaining three periods; the change of marker was necessitated by the unexpectedly low specific radioactivity of ^{103}Ru in one batch of the marker. Comparisons between ^{103}Ru and lignin as solid phase markers were made with digesta samples from three animals. OM flow rates (g/d) in the three animals, based on ^{103}Ru and lignin respectively, were 1937 and 2008, 1756 and 1686, 1565 and 1672 for abomasal flows, 1536 and 1468, 1170 and 1162, 1283 and 1330 for ileal flows, and 1553 and 1512, 1149 and 1068, 1046 and 1000 for faecal flows.

Volatile fatty acids (VFA) in acidified rumen samples were determined by gas–liquid chromatography (F & M Scientific 402; Hewlett Packard Australia Pty Ltd, Sydney) using 3-methyl *n*-valeric acid as an internal standard. Ammonia in acidified rumen samples was determined by the method of Chaney & Marbach (1962). Non-acidified rumen samples were assayed for ^{103}Ru and ^{51}Cr specific radioactivities and mean retention times were calculated as the reciprocal of disappearance rate constants for the two markers.

Bacterial N. The proportion of abomasal N present as bacterial N was calculated as mg DAPA/g N in digesta × g N/mg DAPA in bacterial samples. Bacterial samples isolated from rumen fluid samples collected from each of the five steers at the end of the last collection period contained 55·9, 61·2, 53·7, 40·9 and 53·7 mg DAPA/g N on diets A, B, C, D and E respectively; a mean value of 53·1 mg DAPA/g N was used throughout.

Expt 2

Animals and management. Forty Hereford steers aged approximately 12 months, and weighing an average of 288 kg live weight were used. Animals were housed in individual stalls with sawdust bedding and water available *ad lib.* They were allocated to five diets

on a live-weight basis, using restricted randomization. Live weights were recorded weekly over 57 d, and live-weight changes estimated by regressions of live weight *v.* time.

Diets and feeding procedure. The basal diet was the same oaten chaff as fed in Expt 1. It was offered *ad lib.*, accumulated food refusals being removed twice weekly. The five pelleted supplements were also the same as fed in Expt 1, although larger amounts were fed (Table 1) because of the higher roughage intakes of the larger animals. Daily portions of pellets were placed on top of the oaten chaff each morning, and generally eaten within 1 h.

RESULTS

Expt 1

Animals remained in good health throughout the experiment, and consumed all food offered. Digesta flows of OM at the duodenum, ileum and rectum were similar for the four diets given N supplements (diets B, C, D and E) (Table 2). On the control diet (A), faecal flows of OM were significantly greater than on the other diets; ileal and faecal flows followed the same trend but were less consistent as indicated by the larger standard errors. The proportion of abomasal flows of OM digested in the intestines tended to be lower on diets supplying soluble N supplements (diets B and C). Abomasal flow of α-glucose polymers was lower ($P < 0{\cdot}10$) on the control diet than on the four N diets (diets B, C, D, E). Abomasal, ileal and faecal flows of N were significantly lower on the control diet than on diets B, C, D and E.

Abomasal N flows were 9–16 % higher than N intakes on diets B, C, D and E but 70 % higher on diet A, which indicated substantial N re-cycling on diet A (Table 3). The amount of N apparently digested in the intestines did not differ significantly between diets. Abomasal flow of bacterial N and efficiency of bacterial N synthesis did not differ between diets; bacterial N as a proportion of total abomasal N was significantly lower on diets D and E.

Abomasal flows of most amino acids were substantially higher on diets D and E than on diets A, B and C, particularly proline and glutamic acid (Table 4). On diets B and C, abomasal flows of amino acids were slightly higher than those on diet A. Amino acid analyses on ileal digesta samples were carried out on single pooled samples for each diet. For this reason, apparent intestinal absorptions (abomasal minus ileal flows) were not analysed statistically. Apparent intestinal absorptions of indispensable amino acids were similar on diets A, B and C, which were substantially lower than those on diets D and E. Apparent intestinal absorptions of dispensable amino acids were lowest on diets B and C, higher on diet A and highest on diets D and E. The proportion of abomasal flow apparently absorbed was higher with indispensable than with dispensable amino acids on all treatments.

VFA concentrations in rumen fluid from diets B, C, D and E were significantly higher than on diet A and this was associated with lower proportions of acetic and higher proportions of propionic acid on diet A (Table 5). Rumen ammonia levels on diets A, D and E were substantially lower than those on diets B and C (Table 5). Retention times for ^{103}Ru-P and ^{51}Cr EDTA did not differ between treatments.

Expt 2

Animals remained in good health throughout the experiment, and readily consumed the pelleted supplements. Intakes of oaten chaff were similar for the four diets fed with N supplements (diets B, C, D and E) (Table 6), these intakes being an average of 23 % higher than on the control diet (diet A). Live-weight gains also were similar on diets B, C, D and E, and more than double that on diet A.

Calculated intakes of metabolizable energy (ME) were used to predict live-weight gains

Table 2. *Expt* 1. *Intake and digestion of organic matter* (OM) *and α-glucose polymers in steers eating oaten chaff plus supplements**

	Supplements						Statistical significance of differences between means	
			Casein : HCHO-casein					
Diet* ...	Control A	Urea B	100:0 C	50:50 D	0:100 E	SEM	*F*†	*P*
OM								
Intake (g/d)	3137	3182	3141	3161	3191	—	—	—
Abomasal flow (g/d)	1889	1639	1644	1759	1655	109·9	—	NS
Ileal flow (g/d)	1364	1373	1267	1285	1234	82·6	—	NS
Faecal flow (g/d)	1521	1292	1312	1190	1134	37·3	A > (BCDE)	0·01
Apparently digested (g/d) in								
Stomach	1248	1543	1497	1402	1536	104·6	A < (BCDE)	0·05
Small intestines	525	266	377	474	421	75·5	—	NS
Tract	1616	1890	1829	1971	2003	50·3	A < (BCDE)	0·01
Proportion intake apparently digested in								
stomach	0·40	0·48	0·48	0·44	0·48	0·033	A < (BCDE)	0·05
Tract	0·52	0·59	0·58	0·63	0·62	0·016	A < (BCDE)	0·001
Proportion of abomasal flow apparently digested in								
Small intestines	0·28	0·16	0·23	0·27	0·25	0·040	—	NS
α-glucose polymers								
Intake (g/d)	829	858	701	702	704	—	—	—
Abomasal flow (g/d)	29	42	40	55	47	7·0	A < (BCDE)	0·10
Apparently digested in stomach (g/d)	800	816	661	647	657	7·0	(AB) > (CDE)	0·01

NS, Not significant.
* For details, see Table 1.
† Where groups of treatment letters are shown in parentheses statistical tests were for combined means.

Table 3. *Expt* 1. *Intake and digestion of N, abomasal flows of bacterial N and efficiencies of bacterial synthesis in steers eating oaten chaff plus supplements**

	Supplements						Statistical significance of differences between means	
			Casein:HCHO-casein					
	Control	Urea	100:0	50:50	0:100	SEM	*F*†	*P*
Diet* ...	A	B	C	D	E			
Total N								
Intake (g/d)	24·9	47·2	45·4	48·6	51·0	—	—	—
Abomasal flow (g/d)	42·1	52·8	50·0	56·1	57·9	4·17	A < (BCDE)	0·05
Ileal flow (g/d)	11·0	15·0	18·7	19·8	21·7	1·18	A < (BCDE)	0·001
Faecal flow (g/d)	12·1	14·4	16·4	17·1	18·5	0·72	A < (BCDE)	0·001
Abomasal flow ÷ intake	1·70	1·12	1·09	1·16	1·14	—	—	—
Apparently digested in small intestines (g/d)	31·1	37·8	31·3	36·3	36·2	4·38	—	NS
Proportion of abomasal flow apparently digested in small intestines	0·74	0·72	0·62	0·65	0·63	0·033	(AB) > (CDE)	0·01
Bacterial N								
Abomasal flow (g/d)	27·0	31·8	29·1	24·4	24·0	3·82	—	NS
Proportion total abomasal N flow	0·65	0·60	0·58	0·45	0·42	0·065	(ABC) > (DE)	0·05
Bacterial N/kg OM apparently digested in stomach	21·6	20·6	19·4	17·4	15·6	3·42	—	NS
Bacterial N/kg OM truly digested in stomach‡	15·4	14·9	14·2	13·1	12·1	—	—	—

OM, organic matter; NS, not significant.
* For details, see Table 1.
† Where groups of treatment letters are shown in parentheses statistical tests were for combined means.
‡ Equation no. 6 of Czerkawski (1978).

(Ministry of Agriculture, Fisheries and Food, 1975) which corresponded closely with actual gains.

DISCUSSION

Expt 1

Bacterial protein synthesis

Efficiency of bacterial protein synthesis was determined when iso-nitrogenous supplements of urea, soluble protein (casein) or digestible by-pass protein (HCHO-casein) were fed at three-hourly intervals. It was assumed that casein would increase the available pool of amino acids and peptides in the rumen. When solutions of casein were introduced into the rumen of steers, protein half-lives were found to be 5·6–21·5 min (Mangan, 1972) and 65 min (Broderick, 1978). Rates of proteolysis would have been slower in our experiment because the casein was fed as dry pellets, and solubilization of casein in McDougall's (1948) buffer at 38° takes approximately 12 min (R. G. Redman & R. C. Kellaway, unpublished results). Thus, it is reasonable to assume that the supply of amino acids and peptides in the rumen was indeed greater when casein was fed than when urea was fed. However, efficiencies of bacterial protein synthesis, as indicated by abomasal flows of bacterial N/kg OM truly digested in the stomach, were similar on the control, urea and casein diets (15·4, 14·9 and 14·2 respectively). It appears that the supply of amino acids and peptides was not limiting bacterial protein synthesis on this diet. If 60 % of bacterial N on the urea diet were derived from ammonia-N, 12·7 g N/d would have been supplied from amino acids and peptides arising from proteolysis of dietary protein and endogenous secretions. Of N recycled to the gut anterior to the duodenum 68–77 % was found to be endogenous protein rather than urea (Macrae *et al.* 1977). Measurements of endogenous protein in abomasal secretions (Harrop, 1974) indicate that this pathway would account for most of the endogenous protein recycled anterior to the duodenum, leaving little for secretion into the rumen. Thus, it appears that most of the amino acids and peptides in the rumen arise from dietary rather than endogenous sources. Dietary protein intake on the urea treatment was approximately 20 g/d of which 12·7 g/d may well have undergone proteolysis in the rumen. On semi-synthetic diets which were virtually protein-free, replacement of 100 mg/g urea N with casein N had no effect on efficiency of microbial N synthesis (Ben-Ghedalia *et al.* 1978) whereas replacement of 480 mg/g urea N with casein N increased abomasal flow of protein by 13 % (Hume, 1970). This may represent an upper limit to in vivo responses to soluble dietary proteins in terms of microbial protein synthesis.

The concentration of ammonia in the rumen which promotes maximal bacterial protein synthesis is approximately 4·1 mol/l (Mercer & Annison, 1976; Okorie *et al.* 1977). Ammonia concentrations were lower than this on the control diet (2·1 mol/l) and on the treated-casein diets (2·4 and 2·2 mol/l on diets D and E respectively). On the control diet the low concentration of rumen ammonia was associated with a lower proportion of DM intake digested in the stomach (Table 2), a lower concentration of total VFA in the rumen and a lower value for acetic:propionic acid (Table 5) than on the other diets. Despite the lower rate of fermentation on the control diet, efficiency of bacterial growth was no lower than on other diets. DM intake was controlled at a similar level on all diets in Expt 1. The more rapid rate of fermentation on all but the control diet would have facilitated higher DM intakes on these diets as indeed was found in Expt 2 (Table 6). Under these conditions it is possible that fractional outflow rates from the rumen would have been higher, recycling of microbial N lower, and therefore net efficiencies of microbial protein synthesis higher.

In contrast to the control diet, low levels of rumen ammonia on the treated casein diets did not reduce the extent of OM digestion. Higher levels of rumen ammonia on the urea

Table 4. *Expt* 1. *Abomasal flows (F) (g/d) and apparent intestinal absorption (IA) (g/d) of amino acids in steers eating oaten chaff plus supplements**

	Supplements											Statistical signicance of differences between means‡	
					Casein:HCHO-casein								
					100:0		50:50		0:100		SEM	*F*†	*P*
Diet* ...	A		B		C		D		E				
	F	IA	F	IA	F	IA	F	IA	F	IA			
Indispensable amino acids													
Lysine	16·5	12·7	18·5	14·3	18·1	15·5	19·8	12·8	22·5	16·7	1·63	NS	—
Histidine	5·5	4·1	5·6	4·0	5·4	4·5	7·4	4·2	8·6	5·7	0·70		0·01
Arginine	13·9	10·6	14·1	11·1	11·5	9·7	16·5	10·0	15·5	11·0	1·21		0·05
Threonine	11·8	9·6	10·5	7·2	11·6	7·9	16·8	13·5	14·0	9·2	1·42		0·01
Valine	9·9	7·5	11·5	8·1	11·8	6·8	14·5	10·8	17·5	11·9	1·27		0·01
Methionine	7·2	6·2	7·0	5·7	6·8	5·2	8·4	7·0	8·6	6·9	0·85	(DE) > (ABC)	0·10
Isoleucine	9·4	7·6	11·2	8·4	11·5	8·7	14·2	10·7	15·0	10·6	1·34		0·01
Leucine	16·2	12·5	17·1	12·0	18·5	13·1	23·1	17·1	26·3	19·1	2·28		0·01
Phenylalanine	10·7	7·5	11·5	7·4	10·6	4·7	15·5	11·3	19·3	14·3	1·85		0·01
Total	101·1	78·3	107·0	78·2	105·8	76·1	136·2	97·4	147·3	105·4	9·97		0·01
IA ÷ F	0·77		0·73		0·72		0·72		0·72				
Dispensable amino acids													
Aspartic	22·6	19·4	19·9	13·2	23·7	16·9	24·1	18·3	22·6	14·2	3·18	NS	—
Serine	10·1	8·4	10·8	8·0	13·0	9·7	16·2	13·2	16·2	11·9	1·41		0·01
Glutamic acid	28·4	19·6	28·7	14·8	31·8	17·7	46·6	32·0	47·8	28·7	4·22	(DE) > (ABC)	0·001
Proline	10·1	6·5	11·1	4·5	11·4	4·7	20·2	13·0	24·5	14·8	2·02		0·001
Glycine	12·9	6·3	13·9	4·5	12·7	1·7	15·6	5·7	11·6	2·0	2·46	NS	—
Alanine	15·7	11·6	15·7	9·2	16·4	8·9	18·9	12·5	17·1	10·3	1·92	NS	—
Cystine	0·2	0·2	1·4	1·4	1·3	1·3	1·8	1·8	0·2	0·2		NS	—
Tyrosine	8·2	6·5	10·2	7·2	10·8	6·1	14·2	11·0	13·7	9·9	1·36	(DE) > (ABC)	0·01
Total	108·2	78·5	111·7	62·8	121·1	67·0	157·6	107·5	153·7	92·0	14·25	(DE) > (ABC)	0·01
IA ÷ F	0·73		0·56		0·55		0·68		0·60		0·559	—	—
Total amino acids	209·3	156·8	218·7	141·0	226·9	143·1	293·8	204·9	301·0	197·4	22·40	(DE) > (ABC)	0·01
IA ÷ F	0·75		0·64		0·63		0·70		0·66				

NS, not significant.
* For details, see Table 1.
† Where groups of treatment letters are shown in parentheses statistical tests were for combined means.
‡ Relate to abomasal flows only.

Table 5. *Expt* 1. *Concentrations of volatile fatty acids (VFA) and ammonia in rumen fluid, and mean retention times of* ^{103}*Ru-P and* ^{51}*Cr-EDTA in the rumen of steers eating oaten chaff plus supplements**

	Supplements						Statistical significance of differences between means	
			Casein:HCHO-casein					
	Control	Urea	100:0	50:50	0:100	SEM	*F*†	*P*
Diet* ...	A	B	C	D	E			
Total VFA (mM/l)	61	79	78	78	73	5·1	A < (BCDE)	0·05
VFA proportions								
Acetic	0·600	0·661	0·674	0·671	0·638	0·0132	A < (BCDE)	0·01
Propionic	0·260	0·197	0·158	0·169	0·206	0·0158	A > (BCDE)	0·01
iso-butyric	0·031	0·027	0·026	0·031	0·031	—	—	NS
n-butyric	0·080	0·089	0·106	0·090	0·093	—	—	NS
iso-valeric	0·018	0·013	0·021	0·019	0·027	—	—	NS
n-valeric	0·011	0·013	0·016	0·010	0·017	—	—	NS
Ammonia (mmol/l)	2·0	5·2	5·2	2·4	2·2	0·80	(ADE) < (BC)	0·05
Mean retention times (h)								
^{103}Ru-P	33·0	31·4	23·5	38·3	25·0	7·04	—	NS
^{51}Cr EDTA	21·3	22·0	20·8	28·3	19·4	3·45	—	NS

NS, not significant.
* For details, see Table 1.
† Where groups of treatment letters are shown in parentheses statistical tests were for combined means.

Table 6. *Expt* 2. *Measured dry matter intakes (DMI), calculated intakes of metabolizable energy* (ME) *and net absorption of amino acid nitrogen (AAN), predicted and actual live-weight gains in* 40 *Hereford heifers eating oaten chaff plus supplements**

	Supplements						Statistical significance of differences between means	
			Casein:HCHO-casein					
	Control	Urea	100:0	50:50	0:100	SEM	*F*†	*P*
Diet* ...	A	B	C	D	E			
DMI (kg/d)								
Pellets	0·62	0·69	0·62	0·67	0·72	—	—	—
Oaten chaff	5·51	6·72	6·70	6·96	6·69	0·218	A < (BCDE)	0·001
Total	6·12	7·41	7·32	7·63	7·41	—	—	—
Total (g/kg live wt$^{0·75}$)	69·0	83·3	83·7	88·0	82·9	2·52	A < (BCDE)	0·001
Calculated intakes and absorptions (/d)								
ME‡ (MJ)	48·7	68·2	65·1	72·6	71·7	—	—	—
Total N	50·9	105·4	101·1	110·0	113·3	—	—	—
N intake/MJ ME	1·05	1·55	1·55	1·52	1·58	—	—	—
AAN (g/d)§	36·1	38·9	39·3	58·3	54·2	—	—	—
g AAN/MJ ME	0·74	0·57	0·60	0·80	0·76	—	—	—
MJ ME/kg DMI	8·0	9·2	8·9	9·5	9·7	—	—	—
Predicted live-wt gain (g/d)‖	319	786	709	887	884	—	—	—
Actual live-wt gain (g/d)	356	798	843	842	805	68·7	A < (BCDE)	0·001

NS, not significant.
* For details, see Table 1.
† Where groups of treatment letters are shown in parentheses statistical tests were for combined means.
‡ Digestible organic matter in dry matter (Table 2) × 0·156 × DMI.
§ Apparent absorption AAN/kg DMI (Tables 4 and 2) × organic matter intake × 0·75 (utilization factor; Roy *et al.* 1977).
‖ Ministry of Agriculture, Fisheries and Food (1975).

and casein diets (5·2 and 5·2 mol/l respectively) did not facilitate greater OM digestion in the stomach than on the treated casein diets.

The range of 12–15 g bacterial N/kg OM truly digested in the stomach reported here is lower than the mean of 20 g bacterial N/kg OM for thirty experiments, summarized by Czerkawski (1978), in which microbial N was measured in terms of DAPA. However, it corresponds with efficiencies determined on similar low quality roughages (Jackson *et al.* 1971; Kropp *et al.* 1977). Efficiencies calculated by the same method as we used (Czerkawski, 1978) were 12–14 g bacterial N/kg OM truly digested in the rumen of steers eating low-quality grass (Kropp *et al.* 1977), and these were associated with low dilution rates in the rumen (2·1–2·2 %/h). A major factor which affects efficiency of microbial growth is rumen dilution rate, and the mean rate of 4·5 %/h in the present experiment (reciprocal of ^{15}Cr EDTA retention time in Table 5) is considerably lower than rates of 8–10 %/h which appear to be optimal (Owen & Isaacson, 1977). A possible reason for the relatively low dilution rates in our experiment was that the diet was fed as chaffed particles 10 mm long. Much less mastication and rumination, and therefore saliva production, could have occurred with this material than with long roughages (Balch & Campling, 1965).

N utilization

Abomasal N flow was 1·7 times the N intake on the control diet (Table 3) which indicates substantial N recycling on this diet. Studies with ^{15}N-labelled urea indicated that only 8–18 % of urea recycled to the digestive tract was degraded in the rumen (Nolan & Leng, 1972; Nolan *et al.* 1976). Other studies with ^{14}C-labelled urea found that only 23–32 % of N added to digesta between the mouth and duodenum could be accounted for as recycled urea N (Macrae *et al.* 1977). It appears that the balance of N recycled anterior to the duodenum can be accounted for by abomasal secretions (Harrop, 1974). In our experiment, if it is assumed that 27 % of the N added to digesta between the mouth and abomasum was urea, this would have added 4·6 g urea-N/d to the rumen N pool, which is a relatively small proportion of the N intake of 25 g/d on the control diet. Indeed N was limiting the rate of fermentation on this diet as shown by the substantial increase in rumen fermentation when dietary N supplements were given.

N digestibility in the intestines was influenced by the microbial N:casein-N value. On diets A and B where bacterial N was 0·65 and 0·60 of abomasal N, apparent N digestibilities were 0·74 and 0·72 respectively, whereas on diets D and E where bacterial N was 0·45 and 0·42 of abomasal N, apparent N digestibilities were 0·65 and 0·63 respectively (Table 3). The relatively low digestibility of HCHO-treated casein in diets D and E was partly anticipated from the preliminary in vivo test which showed that the digestibility of treated casein between the rumen and rectum was 0·80. We attempted to compensate for this by feeding proportionately more N in diets D and E.

Mean abomasal flows and mean apparent intestinal absorptions of amino acids were 36 and 37 % higher respectively on diets D and E than on diets A, B and C. The proportions of glutamic acid and proline in abomasal digesta and apparently absorbed amino acids were greater on diets D and E than on diets A, B and C. This is related to the higher concentration of these amino acids in casein than in bacteria and confirms observations made by Faichney (1974), Sharma *et al.* (1974) and Williams & Smith (1976).

Expt 2

Production responses to N supplements

Intakes of oaten chaff with diets receiving N supplements were 21–26 % higher than on the control diet (Table 6). N intake/MJ ME was 1·05 on the control diet, which is con-

siderably less than the value of 1·25 adopted as a desirable value for rumen degradable N (RDN):ME intake (Roy *et al.* 1977). The consequences of low availability of RDN are reductions in digestibility and rate of passage (Campling *et al.* 1962). We found that in Expt 1 digestibility and rumen VFA levels were lowest on the control diet. It is likely that rate of passage was also lowest on this treatment and that this would have been a major factor limiting intake. N intake/MJ ME was in excess of 1·5 on all other treatments.

The substantial intake responses to urea in our experiment are similar to those reported by Campling *et al.* (1962) for oat straw. In our experiment 26 % of DM intake was α-glucose polymers of which one-third came from maize flour in the pellets and two-thirds from grain in the oaten chaff. It is possible that this highly-fermentable substrate enhanced efficiency of urea utilization as suggested by Egan (1975). However, Campling *et al.* (1962), Hemsley & Moir (1963) and Faichney (1965) all showed that addition of sucrose did not enhance urea utilization on straw diets. In contrast, Hennessy *et al.* (1978) found that intra-ruminal infusion of molasses (which avoids the palatability factor) increased considerably the effects of urea alone on digestibility and intakes of native pasture hay. Synergistic effects on intake of feeding highly fermentable substrates with urea are often confounded with palatability of the substrate. It would be useful to differentiate between effects of highly fermentable substrates on intake and rumen fermentation of low quality roughages supplemented with urea.

All N supplements fed in Expt 2 increased intake of oaten chaff to the same extent as urea. If abomasal flows of amino acids were indeed higher on HCHO-casein diets than on the other diets, as calculated in Table 6, there were no stimulatory effects on intake such as those reported by Egan (1975). The value for g digestible protein:MJ digestible energy (DE) below which duodenal infusion of casein increased voluntary intake in Egan's (1975) experiments with sheep was approximately 6·0. In Expt 2 it can be calculated from values in Table 6 that there were 5·7, 4·9, 5·8, 7·2 and 6·7 g digestible protein:MJ DE on diets A, B, C, D and E respectively. This may be interpreted to indicate that these animals should have increased their intake in response to the greater protein:energy on diets D and E. In fact they did not respond to the greater protein:energy, but did give a substantial response to urea-N which actually reduced the protein:energy ratio (4·9 on diet B). It is possible that the disparity between our results and those of Egan (1975) is due to species differences. The lowest voluntary intake per kg live-weight$^{0·75}$ in our experiment was about double that in Egan's (1975) experiments, which conforms with observations of Playne (1970) that voluntary intake of low quality roughages per unit metabolic size is much greater for cattle than for sheep.

Predicted live-weight gains were very similar to actual live-weight gains which indicates that live-weight responses to N supplements can be explained entirely in terms of intake. The calculated values for amino acid N absorbed:MJ ME intake were all well in excess of requirements given by Ørskov (1977), and differences between treatments did not give rise to differences in efficiencies of energy utilization.

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REFERENCES

Balch, C. C. & Campling, R. C. (1965). In *Physiology of Digestion and Metabolism in the Ruminant*, p. 108 [R. W. Dougherty, editor]. London: Butterworths.
Ben-Ghedalia, D., McMeniman, N. P. & Armstrong, D. G. (1978). *Br. J. Nutr.* **39**, 37.
Broderick, G. A. (1978). *J. Nutr.* **108**, 181.

Campling, R. C., Freer, M. & Balch, C. C. (1962). *Br. J. Nutr.* **16**, 115.
Chaney, A. L. & Marbach, E. P. (1962). *Clin. Chem.* **8**, 130.
Czerkawski, J. W. (1978). *J. Dairy Sci.* **61**, 261.
Downes, A. M. & McDonald, I. W. (1964). *Br. J. Nutr.* **18**, 153.
Egan, A. G. (1975). *Rev. Rur. Sci.* **2**, 135.
Faichney, G. J. (1965). *Aust. J. agric. Res.* **16**, 159.
Faichney, G. J. (1974). *Aust. J. agric. Res.* **25**, 583.
Faichney, G. J. (1975). In *Digestion and Metabolism in the Ruminant*, p. 277 [I. W. McDonald and A. C. I. Warner, editors]. Armidale: University of New England Printing Unit.
Harrop, C. J. F. (1974). *J. agric. Sci., Camb.* **83**, 249.
Hemsley, J. A. & Moir, R. J. (1963). *Aust. J. agric. Res.* **14**, 509.
Hemsley, J. A., Reis, P. J. & Downes, A. M. (1973). *Aust. J. biol. Sci.* **26**, 961.
Hennessy, D. W., Nolan, J. V., Norton, B. W., Ball, F. M. & Leng, R. A. (1978). *Aust. J. exp. Agric. Anim. Husb.* **18**, 477.
Hume, I. D. (1970). *Aust. J. agric. Res.* **21**, 305.
Jackson, P., Rook, J. A. F. & Towers, K. G. (1971). *Proc. Nutr. Soc.* **30**, 1A.
Kropp, J. R., Johnson, R. R., Males, J. R. & Owen, F. N. (1977). *J. Anim. Sci.* **46**, 844.
McDougall, E. I. (1948). *Biochem. J.* **43**, 99.
Macrae, J. C. & Armstrong, D. G. (1968). *J. Sci. Fd Agric.* **19**, 578.
Macrae, J. C., Wilson, S., Milne, J. A. & Spence, A. M. (1977). *Proc. Nutr. Soc.* **36**, 77A.
Maeng, W. J., Van Nevet, C. J., Baldwin, R. L. & Morris, J. G. (1976). *J. Dairy Sci.* **59**, 68.
Mangan, J. L. (1972). *Br. J. Nutr.* **27**, 261.
Mercer, J. R. & Annison, E. F. (1976). In *Protein Metabolism and Nutrition*, p. 397 [D. J. A. Cole, K. N. Boorman, P. J. Buttery, D. Lewis, R. J. Neale and H. Swan, editors]. London: Butterworths.
Ministry of Agriculture, Fisheries and Food (1975). *Tech. Bull. Min. Agric. Fish. Fd* no. 33.
Nolan, J. V. & Leng, R. A. (1972). *Br. J. Nutr.* **27**, 177.
Nolan, J. V., Norton, B. W. & Leng, R. A. (1976). *Br. J. Nutr.* **35**, 127.
Okorie, A. V., Buttery, P. J. & Lewis, D. (1977). *Proc. Nutr. Soc.* **36**, 38A.
Ørskov, E. R. (1977). *Wld Rev. Nutr. Diet.* **26**, 225.
Owen, F. N. & Isaacson, H. R. (1977). *Fedn Proc. Fedn Am. Socs exp. Biol.* **36**, 198.
Pilgrim, A. F., Gray, F. V., Weller, R. A. & Belling, C. B. (1970). *Br. J. Nutr.* **24**, 589.
Playne, M. J. (1970). *Proc. Aust. Soc. Anim. Prod.* **8**, 511.
Roy, J. H. B., Balch, C. C., Miller, E. L., Ørskov, E. R. & Smith, R. H. (1977). *Proc. 2nd int. Symp. Protein Metabolism and Nutrition, The Netherlands*, p. 126.
Sharma, H. R., Ingalls, J. R. & Parker, R. J. (1974). *Can. J. Anim. Sci.* **54**, 305.
Tan, N. H., Weston, R. H. & Hogan, J. P. (1971). *Int. J. appl. Radiat. Isot.* **22**, 301.
Thornton, R. F. & Minson, D. J. (1973). *Aust. J. agric. Res.* **24**, 889.
Van Soest, P. J. (1963). *J. Ass. off. agric. Chem.* **46**, 829.
Williams, A. P. & Smith, R. H. (1976). *Br. J. Nutr.* **36**, 199.

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