The nutritive value of silages

Energy metabolism in sheep receiving diets of grass silage or grass silage and barley

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1. Two calorimetric experiments were conducted to study the utilization of energy in sheep given diets of grass silage or grass silage and barley. Three silages were investigated. One was made from first-harvest grass in the spring (S) and the others from regrowth cut either early or late in the autumn (E and L respectively). All were of perennial ryegrass (*Lolium perenne*) and preserved with formic acid. Each silage was given at two levels of feeding, the lower providing approximately a maintenance energy intake. The S and L silages were also given supplemented with barley.

2. The digestibilities of organic matter, cellulose and energy in the silages were high. Measured at maintenance, digestible energy (DE) contents (MJ/kg dry matter (DM)) were 11·83, 14·67 and 12·90 for S, E and L respectively. The DE contents of the S and E silages were depressed at the higher level of feeding but the effect was offset by changes in the energy losses as methane and urine. Metabolizable energy (ME) contents (MJ/kg DM) for the three silages, S, E and L were respectively 9·88, 12·54 and 10·73 at the low level of feeding and 9·91, 11·99 and 11·08 at the high level of feeding. The mean ME content of barley calculated by difference was 13·76 MJ/kg DM.

3. The mean efficiencies of utilization of ME for maintenance (k_m) for the S, E and L silages were 0·69, 0·71 and 0·68 respectively. Corresponding values for fattening (k_f) were 0·21, 0·57 and 0·59. Excepting the k_f for the S silage which was low, observed efficiencies were in broad agreement with those predicted by the equations of the Agricultural Research Council (1965). Similar agreement was obtained with all diets consisting of silage and barley.

Although grass silage is used throughout the United Kingdom for the winter feeding of cattle and sheep, the factors influencing its nutritive value are poorly understood (see Wilkins, 1974). This led to the work reported here which was designed to provide calorimetric information about energy metabolism in sheep receiving diets of silage alone or silage and barley. Three perennial ryegrass (*Lolium perenne*) silages were studied. One was made from first-harvest grass in the spring and the others from regrowth either early or late in autumn.

A preliminary report of some of the results has been given elsewhere (Thomas, Kelly, Chamberlain & Macdonald, 1976).

EXPERIMENTAL

Animals and management

A total of ten Finnish Landrace × Dorset Horn wether sheep were used. They were 2 or 3 years old and weighed 50–55 kg. The four animals used in Expt 1 were rumen-cannulated. The animals were held in metabolism cages in a small animal house during experimental periods except during the determination of respiratory exchange. Food was given in four equal meals each day at 07.00, 12.00, 17.00 and 22.00 hours. Water and mineralized salt blocks were freely available.

Foods

The three silages used were prepared from swards of perennial ryegrass, of the S_{23} and S_{24} varieties. The grass used in Expt 1 was cut in mid-May, 1974, allowed to wilt overnight and harvested with a precision chop forage harvester (New Holland Ltd, Aylesbury, Bucks.).

The material was ensiled in a 300 t wedge silo with the addition of formic acid solution (850 g formic acid/l) at a rate of 2·3 l/t fresh grass. The batches of grass used in Expt 2 were cut from a regrowth sward on 5 August and 17 September 1975. In each instance, a single-chop forage harvester (A. Kidd Ltd, Devizes, Wiltshire) was used and the grass ensiled directly into a 3 t polyethylene bag silo. As in Expt 1, formic acid was added at a rate of 2·3 l/t.

Before each experiment, the silage needed for the whole of the experimental period was removed from the silo and hand-mixed with a fork. The silage was then placed in sealed polyethylene bags containing approximately 50 kg of material and stored at $-5°$. Portions of the silage were taken from the cold-store each week. These were allowed to thaw and weighed into meal portions which were stored at $-5°$ until required. Sub-samples of silage were taken for analysis at each weekly weighing.

The barley used was rolled before feeding. In each experiment a single batch of grain was used for the whole experimental period.

Experimental designs and procedures

Expt 1. The main part of the experiment was conducted according to a 4×4 Latin square design involving four sheep and four dietary treatments. The four diets were (on a fresh weight basis) 2500 g/d of silage, 3500 g/d of silage, 2500 g/d of silage plus 200 g/d of barley and 2500 g/d of silage plus 400 g/d of barley. The diet consisting of the low level of silage alone was intended to meet the animals metabolizable energy (ME) requirement for maintenance and the other treatments to achieve positive energy balances. Each experimental period was 23 d and faeces and urine were collected during 8 d of the last 9 d. For 3 d of the period of collection (days 3, 4 and 5), determinations were made of respiratory exchange. On the final day of each period, samples of rumen fluid were taken at 10.00, 12.00, 14.00 and 16.00 hours.

Following the completion of the Latin square, the fasting metabolism of each animal was determined by the procedure described by Blaxter (1962).

Expt 2. The experiment was carried out in two parts: (*a*) according to a 2×2 Latin square design replicated three times and involving six animals and two dietary treatments namely the early-cut autumn silage given at a level of 2500 g/d or at a level of 3500 g/d (on a fresh weight basis); (*b*) according to a 3×3 Latin square design replicated twice and involving the six animals used in Expt 2(*a*) and three dietary treatments. The three diets were (on a fresh weight basis) 3000 g/d of silage, 4000 g/d of silage and 3000 g/d of silage plus 400 g/d of barley. In each part of the experiment the diet consisting of the low level of silage alone was intended to meet the animals ME requirements for maintenance and the other treatments to achieve positive energy balances. In both parts of the experiment periods were 21 d and on the final 8 d, urine and faeces were collected. For 3 d of this period (days 3, 4 and 5) measurements were made of respiratory exchange.

Following the completion of Expt 2(*a*) the fasting metabolism of each animal was determined by the procedure described by Blaxter (1962). The animals were fully realimented before being introduced to Expt 2(*b*).

Calorimetric procedures

Oxygen consumption and carbon dioxide and methane production were determined using two closed-circuit respiration chambers of the type described by Wainman & Blaxter (1958). The animals, which were trained to the procedures, were harnessed for the collection of urine as described by Wainman & Paterson (1963) and for faeces as described by Fishwick (1973). They were introduced to the chamber and allowed to become accustomed to the instrument for 24 h before measurements of respiratory exchange were begun. Measurements of gas consumption and production were made over three consecutive 24 h periods.

Heat production was calculated from the gaseous exchange and urine nitrogen loss using the factors of Brouwer (1965) and energy retention was calculated as the difference between ME intake and heat production.

Analytical methods

Samples of food, faeces, urine and rumen fluid were analysed where appropriate for dry matter (DM) by drying to constant weight at 60° in a forced draught oven or by the toluene distillation method of Dewar & MacDonald (1961). Crude protein (N × 6·25) was determined by a Kjeldahl method and true protein in a similar way after precipitation with tannic acid. Ammonia-N was determined using the method of Sweetsur (1971), and short-chain fatty acids by the method of Cottyn & Boucque (1968). Other analyses were for water-soluble carbohydrates (Somogyi, 1945), ash (Fertilizer and Feedingstuffs Regulations, 1960) and cellulose (Crampton & Maynard, 1938). Lactic acid was determined using L(+) and D(−)-lactic dehydrogenase (*EC* 1.1.1.27 and *EC* 1.1.1.28) (Sigma Chemicals, 1974). Energy values were measured using an adiabatic bomb calorimeter, silage and urine being ignited on polyethylene (Nijkamp, 1965; McDonald, Henderson & Ralton, 1973).

The gaseous composition of the atmosphere in the respiration chamber in Expt 1 was determined by gas–liquid chromatography. The gases were measured using a Katharometer detector (Gow-Mac Instruments, Shannon, Ireland) after the separation of CO_2 on a column of Poropak Q (Waters Associates Inc, Massachusetts, USA) and O_2, N_2 and methane on a column of molecular sieve 5A (J.J's Chromatography Ltd, Kings Lynn, Norfolk). In Expt 2 gas analysis was carried out with a gas analyser (Cambridge Instrument Co. Ltd, London) using a paramagnetic detector for O_2 and Katharometer detectors for CO_2 and methane.

Statistical analysis

The results of Expt 1, Expt 2(*a*) and Expt 2(*b*) were analysed by analysis of variance. In Expt 1, one animal refused small but variable amounts of food when offered the silage at the high level. Where indicated, results were analysed using a missing plot value for this animal. Comparisons between the early-cut and late-cut autumn silages in Expt 2(*a*) and (*b*) were confounded by time effects. However, the animals and methods used throughout the experiment were constant and time effects within both parts of the experiment and in Expt 1 were small and non-significant. Comparisons have therefore been made between Expt 2(*a*) and (*b*) using a paired *t* test on the assumption that time effects were negligible.

RESULTS

The composition of the diets

The chemical composition of the dietary ingredients are shown in Table 1. All three silages were well preserved with low pH values and low concentrations of ammonia-N and butyric acid. The concentration of total lactic acid and the proportion of lactic acid present as the D(−) isomer was notably higher in the spring silage than in the autumn silages. The gross energy (GE) contents of all three silages were similar and slightly higher than that of the barleys. The late-cut autumn silage had a lower total N and soluble carbohydrate content and a higher cellulose content than the early-cut material.

The intake of DM *and apparent digestibility of* DM, *organic matter and cellulose*

The results for the intake of DM and the digestibility coefficients of dietary constituents are given in Tables 2 and 3. For all three silages the digestibility of DM and organic matter was high. At the low levels of feeding the digestible organic matter contents of the dry matter

Table 1. *The chemical composition of the dietary ingredients*

Expt ...	1		2(*a*)	2(*b*)	
	Silage	Barley	Silage	Silage	Barley
Dry matter (DM) (g/kg)*	303	637	238	189	846
Organic matter (g/kg DM)	911	972	878	906	976
Gross energy (MJ/kg DM)	18·70	18·40	18·83	19·15	18·36
Total nitrogen (g/kg DM)	19·0	15·8	29·2	22·6	16·1
Non-protein-N (g/kg total N)	477	—	477	507	—
Ammonia-N (g/kg total N)	81	—	34	94	—
Cellulose (g/kg DM)	290	65	239	316	62
pH	3·70	—	4·14	4·04	—
Total lactic acid (g/kg DM)	116	—	41	44	—
L(+)-lactic acid (mg/g total lactic acid)	580	—	742	691	—
D(−)-lactic acid (mg/g total lactic acid)	420	—	258	309	—
Water-soluble carbohydrates (g/kg DM)	89	—	96	77	—
Acetic acid (g/kg DM)	23	—	5·5	9·1	—
Butyric acid (g/kg DM)	0·4	—	1·3	1·3	—

* Values for silage obtained by the method of Dewar & MacDonald (1961).

Table 2. *Expt* 1. *The dry matter* (DM) *intake, apparent digestibility of* DM, *organic matter and cellulose and the digestible organic matter content of the dietary* DM *in sheep receiving diets of silage and silage and barley*

(Mean values for four animals)

Diet† (g fresh wt/d)		DM intake (g/d)	Digestibility			Digestible organic matter in DM (g/kg)
Silage	Barely		DM	Organic matter	Cellulose	
2500	—	752	0·715	0·732	0·746	667
3500	—‡	1031	0·704	0·722	0·738	659
2500	200	917	0·726	0·744	0·702	686
2500	400	1081	0·745	0·763	0·699	710
SE of the difference between two means§			0·0071	0·0064	0·0095	6·1
Statistical significance of treatment differences			**	**	*	**

† For details, see p. 206.
‡ Values for three animals only.
§ For comparisons with the high level of silage feeding multiply SE by 1·15. Statistical significance by *F* test: * $P < 0.05$; ** $P < 0.01$.

were 667 g/kg for the spring silage and 705 and 652 g/kg respectively for the early and late-cut autumn silages. For the spring and early-cut autumn silages there was a reduction in the digestibility of dry matter and organic matter at the high level of feeding but the effect was not apparent with the late-cut autumn silage. With both spring and autumn silages the digestibility of cellulose was depressed when the diet was supplemented with barley.

At corresponding levels of DM intake, all differences in digestibility between the early and late-cut silages were statistically significant ($P < 0.01$) as were the differences in the digestible organic matter content of the DM.

The digestion and utilization of dietary N

The results for the intake and metabolism of N as shown in Tables 4 and 5. The proportion of dietary N digested was high for all diets and effects due to the level of feeding or to

Table 3. *Expt 2. The dry matter* (DM) *intake, apparent digestibility of* DM, *organic matter and cellulose and the digestible organic matter content of the dietary* DM *in sheep receiving diets of silage and silage and barley*

(Mean values for six animals)

Expt no.	Diet† (g fresh wt/d) Silage	Barley	DM intake (g/d)	Digestibility DM	Organic matter	Cellulose	Digestible organic matter in DM (g/kg)
	2500	—	644	0·753	0·804	0·841	705
	3500	—	878	0·734	0·784	0·818	688
2(*a*)	SE of the difference between two means			0·0055	0·0055	0·0021	4·8
	Statistical significance of treatment differences			*	*	***	*
	3000	—	568	0·673	0·712	0·796	652
	4000	—	745	0·683	0·724	0·798	658
	3000	400	896	0·732	0·768	0·740	717
2(*b*)	SE of the difference between two means			0·0081	0·0064	0·0063	5·8
	Statistical significance of treatment differences			**	**	***	**

Statistical significance by *F* test: * $P < 0{\cdot}05$; ** $P < 0{\cdot}01$; *** $P < 0{\cdot}001$.
† For details, see p. 206.

supplementation of the diets with barley were relatively small. For sheep given silage at the low levels of feeding a large proportion of the digested N was lost in the urine so that only a small proportion of the N was retained; in Expt 2(*b*) animals on this treatment were in negative N balance. N retention was increased when the silages were given at the higher levels of feeding and for the spring and late-autumn silages when barley supplements were given.

Excepting the faecal loss at the low levels of feeding, at corresponding levels of feeding all differences between the early- and late-cut autumn silages were statistically significant ($P < 0{\cdot}05$).

The digestion and utilization of dietary energy

Results for the intakes of GE, losses of energy in faeces, urine and methane and retentions of energy are given in Tables 6 and 7. As for the digestibility of DM and organic matter, the digestibility of energy in the silages was high. The digestible energy (DE) contents of the DM varied from 11·4–14·7 MJ/kg and were closely related to the dietary digestible organic matter and N contents. For the combined results of determinations with diets of silage alone DE (MJ/kg DOM) = 12·99 + 0·270 N (g/kg DM) (r 0·89). For the spring and the early-cut autumn silages the digestibility of energy was reduced at the high level of feeding but a reduction did not occur with the late-cut autumn silage. In animals receiving unsupplemented silages urine energy losses varied from 3·5–5·0% of the GE intake and tended to be lower at high than at low levels of feeding. Supplementation of silages with barley reduced the proportion of GE lost in urine. Methane energy losses varied between treatments from approximately 5·9–7·8% of the GE intake. There was a tendency for the proportion of methane to be less when silages were given at high levels of feeding than when they were given at low levels of feeding.

For the silages, the ME content of the GE varied from 56·0–66·6%, corresponding to a range of ME contents in the DE of 83·1–86·2%. The ME contents of the DM ranged from

Table 4. *Expt* 1. *The intake, urine and faecal losses and retention of nitrogen (N) (g/d) in sheep receiving diets of silage and silage and barley*

(Mean values for four animals)

Diet† (g fresh wt/d)							
Silage	Barley	Intake	Faecal loss	Urine loss	Retention	Digested N (g/kg N intake)	Retained N (g/kg N digested)
2500	—	14·32	4·81	7·44	2·06	664	213
3500	—‡	19·64	6·57	10·19	2·88	665	213
2500	200	17·06	6·29	8·08	2·69	631	248
3500	400	19·87	6·50	8·27	5·02	672	377
SE of the differences between two means§		—	0·43	0·22	0·33	27·0	21·1
Statistical significance of treatment difference			•	**	**	NS	***

NS, not significant.
† For details, see p. 206.
‡ Values for three animals only.
§ For comparisons with the high level of silage feeding multiply SE by 1·15. Statistical significance by *F* test: * $P < 0.05$; ** $P < 0.01$; *** $P < 0.001$.

Table 5. *Expt* 2. *The intake, urine and faecal losses and retention of nitrogen (N) (g/d) in sheep receiving diets of silage and silage and barley*

(Mean values for six animals)

Expt no.	Diet† (g fresh wt/d)							
	Silage	Barley	Intake	Faecal loss	Urine loss	Retention	Digested N (g/kg N intake)	Retained N (g/kg N digested)
2(*a*)	2500	—	18·82	4·86	10·93	3·09	745	221
	3500	—	25·68	7·01	15·02	3·63	727	194
	SE of the difference between two means		—	0·11	0·54	0·49	5·4	28·5
	Statistical significance of treatment differences			***	**	NS	*	NS
2(*b*)	3000	—	12·84	4·76	8·17	−0·08	630	−13
	4000	—	16·92	6·41	9·12	1·39	621	134
	3000	400	18·05	6·52	8·77	2·76	638	239
	SE of the difference between two means		—	0·40	0·27	0·49	22·8	46·5
	Statistical significance of treatment differences			NS	NS	*	NS	•

Statistical significance by *F* test: NS, not significant. * $P < 0.05$; ** $P < 0.01$; *** $P < 0.001$.
† For details, see p. 206.

9·80–12·54 MJ/kg and were closely related to the DOM contents of the diet; ME (MJ/kg DOM) = 10·66 + 0·244 N (g/kg DM) (*r* 0·93).

At corresponding levels of feeding there were significant ($P < 0.01$) differences between the early-cut and late-cut autumn silages in the amount (%) of GE lost in the faeces and at the high level of feeding in the amount (%) lost in the urine. Differences between the silages in the metabolizability of energy were significant ($P < 0.01$) but differences in the values for ME:DE were significant ($P < 0.05$) only at the low level of feeding.

For all three silages the low level of feeding resulted in an average net energy retention close to zero (Tables 6 and 7). In Expt 2(*a*) the high level of feeding gave a positive energy

Table 6. *Expt* 1. *The intake of gross energy* (GE), *losses of energy in faeces, urine and methane, the digestible* (DE) *and metabolizable energy* (ME) *contents of the diet, the intake of* ME *and the energy retained in sheep receiving diets of silage and silage and barley*

(Mean values for four animals)

Diet† (g fresh wt/d)		GE intake	Faecal energy		Urine energy		Methane energy		DE	ME		DE in dietary DM	ME in dietary DM	ME intake	Energy retained
Silage	Barley	(MJ/d)	MJ/d	% GE	MJ/d	% GE	MJ/d	% GE	(% GE)	(% GE)	ME:DE	(MJ/kg)	(MJ/kg)	(MJ/d)	(MJ/d)
2500	—	12·85	3·96	30·8	0·69	5·1	0·77	6·0	69·2	57·8	0·836	11·83	9·88	7·43	−0·03
3500	—‡	17·77	5·83	33·0	0·59	3·4	1·03	5·9	67·0	57·8	0·862	11·48	9·91	10·21	0·69
2500	200	15·95	4·56	28·6	0·50	3·2	1·07	6·7	71·4	61·5	0·862	12·42	10·71	9·82	1·28
2500	400	18·82	4·95	26·3	0·51	2·7	1·40	7·4	73·7	63·6	0·862	12·82	11·06	11·96	2·70
SE of the difference between two means§		—	0·25	1·16	0·08	0·42	0·05	0·24	1·66	1·40	0·0067	0·16	0·21	0·22	0·25
Statistical significance of treatment differences			**	**	**	**	***	**	•	•	*	***	**	***	***

† For details, see p. 206.
‡ Values for three animals only.
§ For comparisons with the high level of silage feeding multiply SE by 1·15. Statistical significance by F test: * $P < 0.05$; ** $P < 0.01$; *** $P < 0.001$.

Table 7. *Expt 2. The intake of gross energy* (GE), *losses of energy in faeces, urine and methane, the digestible* (DE) *and metabolizable energy* (ME) *contents of the diet, the* ME *intake and the energy retained in sheep receiving diets of silage and silage and barley*

(Mean values for six animals)

Expt no.	Diet† (g fresh wt/d)		GE intake (MJ/d)	Faecal energy		Urine energy		Methane energy		DE (% GE)	ME (% GE)	ME:DE	DE in dietary DM (MJ/kg)	ME in dietary DM (MJ/kg)	ME intake (MJ/d)	Energy retained (MJ/d)
	Silage	Barley		MJ/d	% GE	MJ/d	% GE	MJ/d	% GE							
2(*a*)	2500	—	12·12	2·68	22·1	0·55	4·5	0·82	6·8	77·9	66·6	0·855	14·67	12·54	8·07	0·87
	3500	—	16·54	4·20	25·4	0·74	4·5	1·06	6·4	74·6	63·6	0·853	14·04	11·99	10·52	2·26
	SE of the difference between two means		—	0·10	0·62	0·06	0·40	0·03	0·14	0·62	0·68	0·0060	0·12	0·13	0·11	0·11
	Statistical significance of treatment differences			***	**	*	NS	**	NS	***	*	NS	**	*	***	***
2(*b*)	3000	—	10·87	3·54	32·6	0·44	4·06	0·80	7·41	67·3	56·0	0·831	12·90	10·73	6·09	−0·33
	4000	—	14·33	4·52	31·5	0·51	3·58	1·02	7·07	68·4	57·8	0·844	13·12	11·08	8·28	1·07
	3000	400	16·89	4·48	26·5	0·47	2·81	1·31	7·77	73·5	62·9	0·856	13·86	11·86	10·63	2·48
	SE of the difference between two means		—	0·15	0·84	0·03	0·25	0·03	0·19	0·84	1·07	0·0067	0·16	0·21	0·13	0·22
	Statistical significance of treatment differences			**	*	NS	*	***	NS	*	*	NS	*	*	***	**

NS, not significant.
Statistical significance by *F* test: * $P < 0{\cdot}05$; ** $P < 0{\cdot}01$; *** $P < 0{\cdot}001$.
† For details, see p. 206.

Table 8. *Expts* 1 *and* 2. *The fasting metabolism* (*MJ/kg liveweight*$^{0.75}$) *of the ten sheep*

(Values in parentheses are mean values with their standard errors for the observed values expressed in kJ/d)

Expt 1				Expt 2			
Sheep no.	Fasting metabolism	Mean	SE	Sheep no.	Fasting metabolism	Mean	SE
201	0·248			208	0·238		
117	0·215	0·238	0·008	214	0·209	0·231	0·004
118	0·237	(4980)	(176)	204	0·262	(4422)	(219)
212	0·251			206	0·209		
				209	0·213		
				210	0·255		

balance of 2·26 MJ/d and in Expts 1 and 2(*b*) the corresponding balances were 0·69 and 1·07 MJ/d. The reason for the relatively low retention in Expt 2(*b*) was the low ME intake but in Expt 1 the ME intake was high and a large proportion of that energy was lost as heat. Supplementation of the spring and late-autumn silages with barley resulted in an increase in ME intake and energy retention.

Fasting metabolism

The mean values for fasting metabolism obtained in the two experiments were similar and the standard errors of the estimates were small (Table 8). The values were also close to the value of 0·228 kJ/kg live weight$^{0.75}$ indicated by the Agricultural Research Council (1965).

Fermentation in the rumen

The results for the pH and concentrations of ammonia-N and total and individual short-chain fatty acids in the rumen obtained in Expt 1 showed postprandial depressions in rumen pH and increases in the concentrations of ammonia-N and total short-chain fatty acids. As in other experiments with silages of this type (Farhan & Thomas, 1977) there was a postprandial decrease in the proportion of acetic acid in the rumen and a compensatory increase in the proportion of propionic acid. However, postprandial variations in the composition of the rumen fluid were similar for all dietary treatments and the results are therefore summarized in Table 9 as mean values. For the diets consisting solely of silage rumen pH was on average approximately 7 but the pH was reduced with the inclusion of barley in the diet. Ammonia-N concentrations on all treatments were reasonably high although a slightly lower concentration was observed with the diet containing the high level of barley than with the other diets. The concentration of total short-chain fatty acids in the rumen increased in close correlation with the ME intake but where the additional ME was supplied as barley there were changes in the composition of the fatty acid mixture. These were characterized by a reduction in the proportion of acetic and propionic acids and a compensatory increase in the proportion of butyric acid.

DISCUSSION

The silages used in these experiments were prepared under carefully controlled conditions and were well preserved with good fermentation characteristics (see Table 1). But they were typical of many good-quality formic acid silages prepared on commercial farms in south-west Scotland. After their removal from the silo, the silages were stored at $-5°$. This procedure allowed mixing to improve the silages uniformity and eliminated changes in their composition due to oxidative deterioration. The possibility that cold storage of silage may influence its nutritive value cannot be excluded but any effects of cold storage are

Table 9. *Expt* 1. *The pH and concentration of ammonia-nitrogen and total and individual short-chain fatty acids (VFA) in the rumen fluid of sheep receiving diets of silage and silage and barley*

(Mean values for four animals. Value for each animal is a mean of four samples)

Diet† (g fresh wt/d)					Individual fatty acids (mmol/mol total VFA)					
Silage	Barley	pH	Ammonia-N (mg/l)	Total VFA (mmol/l)	Acetic acid	Propionic acid	Isobutyric acid	Butyric acid	Isovaleric acid	Valeric acid
2500	—	6·98	124	68·1	609	264	12	90	12	13
3500	—‡	7·02	119	86·2	605	269	11	90	11	15
2500	200	6·88	130	78·6	602	241	10	120	14	12
2500	400	6·75	93	94·8	585	227	10	153	12	13
SE of the difference between two means§		0·05	35	5·6	17	20	1	10	2	1
Statistical significance of treatment differences		**	NS	*	NS	NS	NS	**	NS	*

NS, not significant.
† For details, see p. 206.
‡ Values for three animals only.
§ For comparisons with the high level of silage feeding multiply SE by 1·15. Statistical significance by F test: * $P < 0·05$; ** $P < 0·01$.

probably small. In calorimetric studies, Ekern, Blaxter & Sawers (1965) showed that with fresh grass cold storage resulted in only slight changes in energy and N metabolism. More recently, MacRae, Campbell & Eadie (1975) have found that with high-quality grasses the solubility of the plant proteins is reduced by freezing and thawing and Beever, Cammell & Wallace (1974) have shown that this is linked with an increased uptake of amino acids from the small intestine. The effects on protein solubility are thought to be due to the rupture of the plant vacuolar membrane and the precipitation of the cytoplasmic proteins by the consequent decrease in pH. With silages, however, the cytoplasmic proteins have been exposed to low pH in the silo and any release of vacuolar contents on freezing and thawing is probably of little significance.

For the three silages studied the digestibility of DM, organic matter and cellulose was high (Table 2 and 3) and this was reflected in their high DE contents. At the maintenance level of feeding, the spring silage contained 11·83 MJ/kg DM and the autumn silages 14·67 and 12·90 MJ/kg DM (Tables 6 and 7). The digestibility of energy in the spring and early-cut autumn silages was reduced when the level of feeding was increased (Tables 6 and 7) but for the late-cut autumn silage the digestibility was unchanged. The reductions in digestibility are similar to those found with diets of dried grass (Armstrong, 1964; Alwash & Thomas, 1971) and probably reflect the effect of the level of feeding on both the rate of passage of digesta through the alimentary tract and on cellulolytic activity in the rumen. With dried grasses the effects of level of feeding on digestibility are more marked with ground and pelleted than with chopped grasses (Alwash & Thomas, 1971), but although the digestibility of silages is reduced when they are given in a finely chopped form (Thomas, Kelly & Wait, 1976) here the 'chop length' of the silage did not seem important to the effect of the level of feeding. The reduction with the single-chopped early-autumn silage was slightly greater than with the precision chopped spring silage.

Methane losses with diets solely of silage ranged from 5·9 to 7·4% of the GE intake and urine losses from 3·4 to 5·1% (Tables 6 and 7). Both tended to be proportionally lower at the high than at the low level of feeding offsetting the effects of level of feeding on DE. The ME content of the DE varied from 83·1 to 86·3% a range higher than that suggested for the computation of ME (Agricultural Research Council, 1965; Ministry of Agriculture, Fisheries and Food, 1975) but consistent with some other published values for silages (Agricultural Research Council, 1965). Calculated on a per kg of DM basis the ME contents varied from moderately good levels of approximately 9·9 and 10·7 MJ/kg DM for the spring and late-cut autumn silages to an excellent 12·5 MJ/kg DM for the early-cut autumn silage. These values compare well with those recently determined at the Rowett Research Institute for sixteen Aberdeenshire silages prepared on commercial farms from mixed grass swards, most of which contained Timothy (*Phleum pratense*) and Ryegrass (Department of Agriculture and Fisheries for Scotland, 1975). The results for the autumn silages also indicate that where the source of herbage, ensilage technique and fermentation in the silo is constant the maturity of the harvested grass has a similar importance in determining the ME content for silage as it does for dried grass (Armstrong, 1964).

Supplementation of the spring and late-cut autumn silages with barley reduced the digestibility of cellulose in the diet (Tables 2 and 3) and there were associated reductions in rumen pH (Table 9). These effects are broadly similar to those reported with diets of dried grass and barley (Ørskov & Fraser, 1975). The barley supplements increased the ME content of the diets and reduced urine energy losses (Tables 6 and 7). Calculated by difference, the average ME content of barley was $13{\cdot}76 \pm 0{\cdot}29$ MJ/kg DM (Expt 1 and 2, n 4). This is approximately 5·8% higher than the mean value derived in experiments at Rostock (Hoffman, Schiemann & Nehring, 1963) and the Rowett Institute (Department of Agriculture and Fisheries for Scotland, 1975), but it is within the range published by the latter centre for

individual barleys and is close to the 13·7 MJ/kg DM indicated by the Ministry of Agriculture, Fisheries and Food (1975).

Several authors have reported that with diets consisting solely of silage, levels of ammonia in the rumen and the proportion of dietary N lost in the urine are characteristically high. Supplementation of these silages with cereals leads to a reduced ammonia concentration and improved N retention (see Wilkins, 1974). With the present experiments the energy available from the diets consisting solely of silage given at a low level was restricted and only small amounts of N were retained (Tables 4 and 5). All treatments providing additional ME resulted in increased N retention. But additional energy in the form of barley rather than silage improved the efficiency of utilization of digested N. There is evidence from other experiments that this effect is linked with an increased uptake of amino acids from the small intestine (Thomas *et al.* 1976), but it should be noted that in Expt 1 the average rumen ammonia concentration was depressed by barley only at the high level of supplementation (Table 9).

There are special problems in measuring the efficiency of utilization of the ME of silages. The materials are heterogenous in nature which must increase the errors of the calorimetric determinations and because of the limitations on their voluntary intake it is difficult to achieve large positive energy retentions to optimize the accuracy of estimation of the net energy for fattening. In the present experiments estimates of energy utilization were made over a comparatively narrow range of retentions mainly at or above zero energy balance. The values for energy retention (ER) with each of the silages scaled by dividing by the fasting metabolism of the sheep used in the determination and plotted against the ME intakes (MEI) scaled in the same way, as suggested by Blaxter (1973), indicated that the efficiency of utilization of energy differed markedly between the silages. For the early-cut autumn silage ER = 0·7369 MEI − 0·0437 MEI^2 − 1 ($P < 0·001$) and for the late-cut autumn silage ER = 0·6647 MEI − 0·007 MEI^2 − 1 ($P < 0·001$) but for the spring silage the relationship was more curvilinear, ER = 1·0160 MEI − 0·2354 MEI^2 − 1 ($P < 0·001$), indicating a poor efficiency of utilization of the energy of the spring silage for tissue deposition. Assuming two linear relationships between energy retention and energy intake, one above and one below maintenance, the partial efficiencies of utilization of ME for maintenance (k_m) and fattening (k_f) can be calculated by the method of Blaxter & Wainman (1964). Using the mean values obtained for fasting metabolism and for energy retention at each level of feeding (Tables 6, 7 and 8) the results indicate k_m values of 0·666, 0·679 and 0·673 for the spring, early-cut autumn and late-cut autumn silages respectively and corresponding k_f values of 0·220, 0·563 and 0·625. These values represent estimates for which the effects of errors in the determination of fasting metabolism or energy retention in individual animals are minimized. In contrast, calculations made using the results for individual animals are markedly affected by errors in any single determination but standard errors for the values of k_m and k_f can be calculated. Using the results for individual sheep the k_m and k_f for the spring silage were calculated as 0·690 ± 0·0185 (n 4) and 0·205 ± 0·06102 (0·1431 ± 0·00630 not allowing for the missing value). Corresponding values for the early-cut autumn silage were 0·707 ± 0·0049 (n 6) and 0·565 ± 0·0617. For the late-cut autumn silage k_m was 0·677 ± 0·0174 but k_f could be estimated less satisfactorily. For two sheep the energy retention at the high level of feeding was very small and thus there was only a small difference in ME intake between the calculated zero energy balance point and the high level of feeding. For these animals k_f was anomalously high. Using the remaining four values k_f was 0·593 ± 0·1246 (n 4).

The k_m values, in all instances, are in reasonable agreement with those calculated as suggested by the Agricultural Research Council (ARC, 1965) or by Blaxter (1973) using equations derived from diets not including silage, (Table 10), and this confirms the findings of Ekern & Sundstøl (1973) (Table 10). For the autumn silages the k_f values are also in

Table 10. *The partial efficiences of utilization of the metabolizable energy* (ME) *of silage diets for maintenance* (k_m) *and fattening* (k_f) *determined in Expts* 1 *and* 2(*a*) *and* (*b*) *and in published experiments and the values calculated by the equations of the Agricultural Research Council* (1965) *and Blaxter* (1973)

Diet and details of silage	Determined		Calculated*		Calculated†		Source of results
	k_m	k_f	k_m	k_f	k_m	k_f	
Spring, first cut perennial ryegrass (*Lolium perenne*), wilted and ensiled with formic acid	69	21	72	50	68	46	Expt 1
Early-autumn, regrowth perennial ryegrass, direct cut and ensiled with formic acid	71	57	75	57	70	53	Expt 2(*a*)
Late-autumn, regrowth perennial ryegrass, direct cut and ensiled with formic acid	68	59	71	48	67	44	Expt 2(*b*)
Spring, wilted	66	40	71	48	67	44	Ekern & Sundstøl (1973)
Spring, first harvest, wilted	—	49	—	49	—	45	Smith, Wainman & Dewey (1977)
Spring, first harvest, wilted	—	54	—	47	—	50	Smith, Wainman & Dewey (1977)
Summer or autumn third harvest, wilted	—	31	—	48	—	44	Smith, Wainman & Dewey (1977)
Spring, first cut silage plus barley 82:18, w/w‡	—	43	—	53	—	49	Expt 1
Spring, first cut silage plus barley 70:30, w/w‡	—	52	—	55	—	50	Expt 1
Late-autumn, regrowth silage plus barley 63:37 w/w‡	—	54	—	54	—	50	Expt 2(*b*)
Mean difference between predicted and actual values with SE			+3·2 ±0·66	+5·3 ±3·37	−1·0 ±0·63	+1·8 ±3·20	

* Calculated from equation of Agricultural Research Council (1965): k_m = 0·3 ME/GE+54·6; k_f = 0·81 ME/GE+3·0, where GE is gross energy.

† Calculated from equations of Blaxter (1973): k_m = 0·207 ME/GE+55·9; k_f = 0·78 ME/GE+0·6.

‡ Proportions of silage:barley on a dry matter basis given in parentheses; k_m value for determination of k_f has been assumed to be that given by the calculation of Agricultural Research Council (1965).

broad agreement with those derived by the Agricultural Research Council equation (Table 10). But for the spring silage the determined k_f was unusually low (Table 10). Previous studies by Ekern & Sundstøl (1973) have given k_f values slightly lower than predicted by the Agricultural Research Council (1965) method, but agreement with the values predicted by the equations of Blaxter (1973) is fairly good (Table 10). More recently Smith, Wainman & Dewey (1977) have reported good agreement between determined and predicted values for two first harvest silages (Table 10), but they also observed an unusually low k_f with a silage made from third harvest grass (Table 10). Their low value could, however, be explained by the maturity of the grass. With dried forages of given metabolizability, k_f is lower for aftermaths than for first harvest materials (Blaxter, 1973). However, this effect cannot explain the results of Expt 1. It is possible that the low k_f of the spring silage is due to an imbalance in the mixture of nutrients absorbed from the digestive tract but if this is so the imbalance is readily corrected by a barley supplement since the k_f values for diets containing barley, agree well with those derived from prediction equations (Table 10). However, to account for the high heat increment any imbalance with the high level of silage would have to be severe and there is no evidence from the composition of rumen fluid (Table 9) or from methane production (Table 6) of abnormalities in digestion, at least in the rumen. An alternative and equally consistent suggestion is that the poor utilization of the spring silage is due to impairment of the animals metabolism by products formed during fermentation in the silo. These would have their most serious effects at high levels of silage intake and would cause the relationship between energy retention and energy intake to be markedly curvilinear.

The mechanism by which energy metabolism could be impaired and the agent(s) responsible are matters of conjecture. However, a notable difference between the spring and autumn silages was in their lactic acid content, particularly in their content of D(−)-lactic acid (Table 1). This acid is known to be slowly utilized in the tissues (Dunlop & Hammond, 1965; MacKenzie, 1967) and if it is absorbed in significant amounts from silage diets its presence in the circulation might have important metabolic consequences.

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