A method of circulating and sampling the rumen contents of sheep fed on ground, pelleted foods

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Considerable difficulty and uncertainty can be associated with sampling through cannulas from the reticulo-rumen of the sheep. The contents are by no means homogeneous, and for some purposes relatively large samples have to be withdrawn from various places within the organ and mixed in an external container before a subsample is taken for analysis and the unused portion returned to the rumen. In addition to the uncertainties of this conventional method of sampling, it is a rather cumbrous procedure and rumen contents may be lost from the open cannula if the animal bleats or moves rapidly.

Mixing of materials added to the rumen is a slow process. J. G. Gordon (1960, personal communication) has studied the concentration of polyethylene glycol (PEG) in samples withdrawn as described above and has found it to fluctuate widely for several hours after introduction of the marker. The rumen contents are in fact a relatively unhomogeneous and slowly mixing pool. The early stages of experiments that depend on tracing the concentration or specific activity of materials added to the rumen have therefore an inherent uncertainty. Examples of experiments particularly sensitive to this uncertainty are those involving measurement by isotope dilution of short-chain fatty acid production (Sheppard, Forbes & Johnson, 1959; Gray, Jones & Pilgrim, 1960).

Finely ground, pelleted diets are becoming of increasing importance in the nutrition of ruminants and, being stable and easily stored and handled, they are useful for certain types of experimental work. Although theoretically with such diets a small sample can be made representative of the whole, their effect in decreasing rumen motility can increase the difficulties of experiments when sampling from the rumen is involved. The rumen contents remain heterogeneous on these diets, the proportion of dry matter being higher at the surface than in the lower part of the rumen. We thought, however, that with these diets continuous artificial mixing of the rumen contents could be achieved and that added materials would be rapidly mixed with the rumen contents.

This paper describes the construction and use of a pump which withdraws rumen contents from a more or less central position in the rumen, forces them through an external circuit containing a sampling device before returning them to the liquid surface in the rumen, and thus gives rapid mixing of rumen contents. Experiments

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are described which were designed to test the effects of this method of circulating the rumen contents on the metabolism of food in the rumen and the efficiency with which the system mixed PEG with the rumen contents.

EXPERIMENTAL

Animals and their treatment. Three mature Greyface wethers of about 120 lb live weight were fitted, by the method of Ash (1957), with rumen cannulas of 2 in. internal diameter.

The animals were given 500 g of pelleted diet at 7.30 a.m. and again at 7.30 p.m. The rations were completely consumed within 15 min of feeding. Two diets were used: grass cubes consisting entirely of dried-grass meal, and maize cubes consisting of 50 % dried-grass meal, 44 % ground flaked maize, 5 % linseed meal, 0·5 % steamed bone flour and 0·5 % salt, with 1 g cobalt chloride hexahydrate added to each 1000 lb. The results of some analyses of these diets are given in Table 1. Two animals received the grass-cube diet. The third animal was kept throughout on maize cubes. Access to water, salt and mineral licks was provided at all times. During sampling periods the animals were held in a stall.

Table 1. *Chemical composition of the pelleted diets, expressed as a percentage of the dry matter*

Component	Grass cubes	Maize cubes
Ether extract (ash-free)	4·25	2·29
'Normal acid fibre'	37·75	25·10
Crude protein (N × 6·25)	16·38	15·75
Dialysable nitrogenous material (N × 5·98)	2·40	1·06
Ash	9·86	6·59

Analytical methods. Nitogen was measured by the macro-Kjeldahl technique. 'Normal acid fibre' was determined by the method of Walker & Hepburn (1955). Total volatile acid and ammonia determinations were made on cell-free supernatant rumen fluid prepared as follows. Immediately after withdrawal, samples of rumen contents were centrifuged for 5 min at 2000 ***g***. A measured portion of the supernatant fluid was treated with 0·1 vol. 10 N-H_2SO_4 and centrifuged at 20 000 ***g*** for 20 min. The mean value with its standard error for recovery of added volatile fatty acids was 99·7 ± 0·2 % (four observations). Fatty acids were titrated in the steam distillate; ammonia, after micro-diffusion into boric acid, was titrated with hydrochloric acid (Conway & O'Malley, 1942). Chromatography of the volatile acids was by the method of James & Martin (1952). pH was determined directly by glass electrode in samples immediately after withdrawal.

PEG was determined in weighed samples of rumen contents. The samples (about 15 g) were extracted five times with 30 ml water and the combined supernatant liquids were made up to 250 ml for analysis by the method of Hydén (1956).

Samples for the determination of dry-matter percentage were dried at 105° for 48 h.

Construction of the pump. Two main requirements have to be met in the design of a pump suitable for the circulation of rumen contents; first it must be gentle in action with no grinding effect and, secondly, it must present no projections or irregularities that might lead to silting and blockage. A pump, working on the Dale–Schuster principle (Dale & Schuster, 1927–8), in which the moving parts consist of a tubular rubber membrane and rubber flutter valves and in which the diameter of channel throughout the external circuit is almost uniform, was constructed. This pump, illustrated in Fig. 1 and Pl. 1, was found to have a flow rate of from 1 to 2 l./min. The diaphragm (*A*) is cut from a $1\frac{1}{2}$ in. bicycle tyre inner tube. The diaphragm is

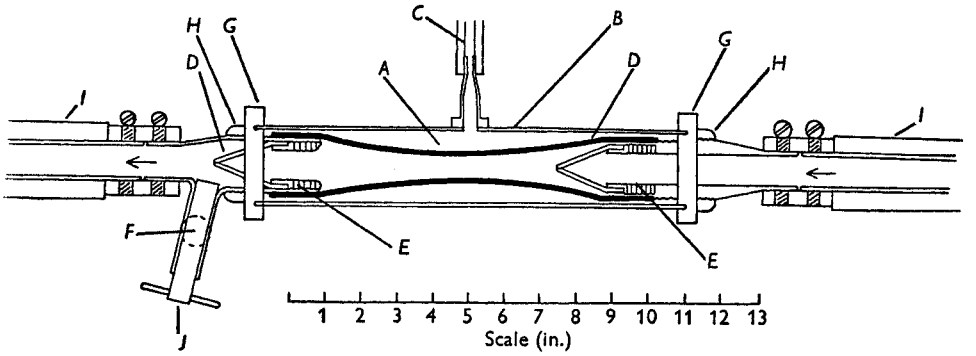

Fig. 1. Diagram of the construction of the rumen perfusion pump. The arrows show the direction of flow of the rumen contents. *A*, rubber diaphragm; *B*, air chamber; *C*, side arm to bellows; *D*, flutter valves; *E*, valve holders; *F*, sampling spigot; *G*, end plates; *H*, locking nuts; *I*, polyurethane foam insulation; *J*, spigot plunger.

held in the air chamber (*B*), a cylindrical Perspex box with a side arm (*C*), which is connected by rubber tubing to the bellows. The flutter valves (*D*) are cut from the type used in civilian respirators. They are bound with thin wire to the valve holders (*E*), which are of Tufnol and screw into the ends of the pump. The rest of the components are machined in Perspex. The sampling spigot (*F*) is machined with a slight taper, inserted into a machined hole and fixed with Perspex cement. The end plates (*G*) of the air chamber screw over the ends of the pump and are held by locking nuts (*H*). The cylindrical part of the air chamber recesses into rubber cushions sunk in the end plates. The end plates are held together by 3/16 in. steel rods threaded to take 2BA nuts. The withdrawal and return pipes (also of Perspex) are brought round in smooth curves and inserted through a rubber bung which fits the aperture of the rumen cannula. The withdrawal pipe is joined to a length of polyvinylchloride tubing, the end of which is cut obliquely. When the pump is in position this tube reaches a more or less central position towards the caudal end of the rumen. The return pipe ends just inside the cannula. The distance from the inside of the cannula to the end of the withdrawal pipe is about 6 in. The portions of those pipes that are outside the animal are covered with two layers of $\frac{1}{2}$ in. thick polyurethane foam (*I*) to minimize heat loss. Fig. 2 shows the position of the pipes when the pump is attached to the animal. The diaphragm is alternately compressed and distended by air pressure from a set of bellows of the type used in a rabbit respiratory pump (C. F. Palmer Ltd,

London). The bellows are actuated by a reciprocating arm (stroke 2·75 in.) driven by an electric motor (0·166 h.p.; 1420 rev/min) geared down 1:40 and giving 35·5 strokes/min. The bellows and motor are shown in Pl. 1.

Use of pump. The rubber bung through which the withdrawal and return pipes pass was inserted into the cannula and bound to it with adhesive tape. The weight of the pump was supported by adjustable guys from a framework over the holding stall. The bellows were then started and a small quantity of water (50 ml) was introduced through a side tube to wet the valves. This small quantity was considered negligible in relation to the volume of the rumen contents (about 7 l.). The pump was in operation

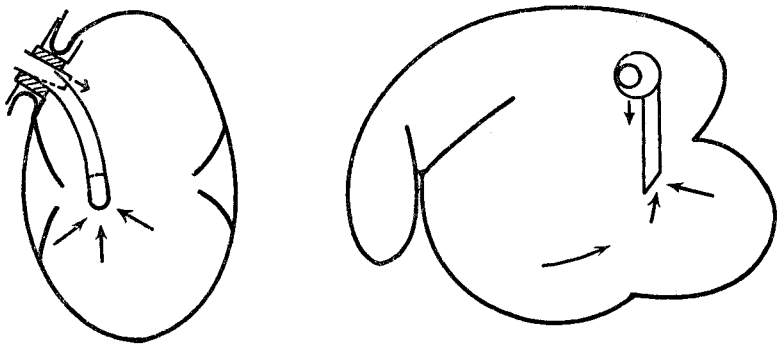

Fig. 2. Position within the rumen of the return and withdrawal pipes of the rumen perfusion pump. Arrows show the direction of flow of the contents. Left, rumen viewed from caudal end; right, reticulo-rumen viewed from left side.

for at least 30 min before the beginning of each experiment. Additions of PEG were made in 60 ml of water over a 3 min period by injection through a side tube situated on the delivery side of the pump. This was also the site used for constant injection. All material introduced here must pass through the rumen before coming round to the sampling spigot. Samples were withdrawn through the spigot by holding a tube under the orifice and rapidly working the plunger (*J*, Fig. 1). Later a small Perspex block was fixed on the side arm to hold a 10 ml open-ended syringe so that samples of more constant size could be withdrawn.

Conventional sampling. The end of a flexible tube of $\frac{3}{4}$ in. internal diameter was placed at the bottom of the anterior ventral sac. Aspiration was begun, and continued while the end of the tube was gradually moved up through the rumen contents to the surface. While the tube was being raised it was moved backwards, forwards and across the anterior part of the rumen. About 200 ml were collected in this way. By the same procedure of placing the end of the sampling tube at the bottom and gradually raising it to the surface 200 ml were withdrawn from the mid-rumen and a further 200 ml from the posterior end of the rumen. The three samples were collected in the same container. After shaking, subsamples were taken of suitable size for analysis.

Efficiency of mixing and sampling. A series of experiments was performed in which 5 g PEG were injected into the pump circuit over 3 min and samples of rumen contents were collected at 10 min intervals over a 1 h period from the time of injection.

Reproducibility of sampling for dry-matter determination. Preliminary experiments using the conventional sampling technique described above gave poor reproducibility of sampling for dry-matter determination on 10 g subsamples. With one of the animals fed on grass cubes, four experiments were carried out on different days in each of which three samples of 9 g wet weight were withdrawn from the pump at 1, 3, 5, 7, 9 and 11 h after feeding. Each time the first sample was withdrawn 5 min before the hour, the second sample at the hour, and the third sample 5 min after the hour. This procedure allowed a flow through the pump circuit of some 10–20 l. between the collection of the first and third samples. Three similar experiments were carried out on the animal fed on maize cubes. The object of these experiments was to test the between-replicate reproducibility of sampling from the pump for dry matter.

Effects of thorough mixing on the concentration of various constituents of rumen contents. If the continuous circulation of the rumen contents materially altered the pattern of metabolic processes of the rumen one might expect alterations in the concentration of the metabolic products present in the organ. Comparisons were therefore made of the concentration of volatile acids and ammonia and of the pH of the rumen on days when the pump was in use throughout the day and on days when it was not.

On 4 days samples were drawn by the conventional method and on 4 days with the pump from a sheep fed on the grass-cube diet. Samples were drawn from the animal fed on the maize-cube diet on 5 days by the conventional method and on 5 days by the pump method. The samples were examined for pH, ammonia concentration, volatile fatty acid concentration and partition of the fatty acids between acetate, propionate and butyrate. It was hoped to alternate the experiments with the two sampling methods at regular intervals, but occasional large losses of rumen contents due to the sheep's bleating during sampling by the conventional method and a mechanical failure of the bellows in the middle of the series of experiments made it impossible. The animals were mature and had been on the fixed diet for at least 6 months before the beginning of the experiments; consequently any trends with time would be expected to be negligible.

Effects of thorough mixing on the digestibility of the diet. The pump was used with two sheep in alternate weeks for 8 h/day for 6 days of the week, and collections of faeces and urine were made twice daily. These collections were combined, subsampled and analysed at the end of each 3-day period. The collection periods were from 7 a.m. on Monday to 7 a.m. on Thursday, and from 7 a.m. on Thursday to 7 a.m. on Sunday. The periods with the pump were from 9 a.m. to 5 p.m. The animals were moved from the metabolism crates to their pens on the 7th day for cleaning and so that the metabolism crates could be scrubbed. One animal (receiving the maize-cube diet) became cage-weary after 5 weeks and was rested. The other animal on grass cubes remained contented and was kept in the metabolism crate for a total of 9 weeks with the sequence *UPUPUPU*, where *U* denotes a week when the pump was not used and *P* a week when it was. With the animal on the maize-cube diet the sequence was *UUPUP*.

RESULTS

Efficiency of mixing. The results of four experiments in which PEG was added to the rumen and samples were withdrawn at intervals from the pump are given in Fig. 3. It will be seen that all values from 30 min onwards lie very close to the straight lines that have been drawn through them and that consequently mixing was complete by that time. The earlier values do not depart so far from the straight lines as to suggest

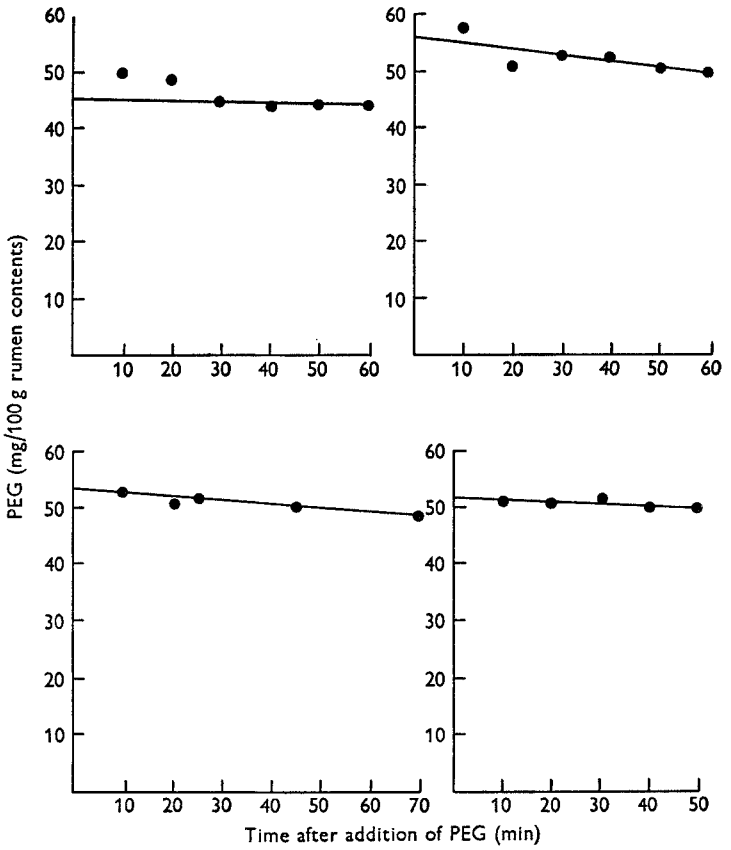

Fig. 3. Concentration in four experiments of polyethylene glycol in small samples of rumen contents of a sheep, withdrawn from the rumen perfusion pump after the addition of the reference substance. The straight lines are drawn by eye through the later points.

that sampling at these times would give rise to large errors. The assumption is being made that over a short time the disappearance curve of the soluble marker can be closely approximated to a straight line.

Reproducibility of sampling for dry-matter determination. The results of the checks on replication of dry-matter determinations on samples of rumen contents drawn with the pump are given in Table 2. The standard deviation between replicates was derived from four sets of triplicate determinations for each value given in Table 2 and varied from 0·16 g dry matter/100 g wet weight 1 h after feeding down to 0·06 g

dry matter/100 g wet weight at the end of a feeding cycle in the experiments using one of the sheep fed on grass cubes. The range of the standard deviation between replicates of the samples withdrawn from the animal fed on maize cubes was from 0·06 to 0·15 g dry matter/100 g wet weight with a less consistent pattern with time after feeding. The fact that the mean value of the dry-matter percentage was the

Table 2. *Reproducibility of sampling for dry-matter determination from the rumen of the sheep by the pump method. Mean values for the percentage of dry matter in the rumen contents with standard deviations between replicates and between days*

(Values are derived from four sets of triplicate analyses for the animal fed on grass cubes and three sets of triplicate analyses for the animal fed on maize cubes)

Time after feeding (h)	Mean value	SD between replicates	SD between days
	Grass-cube diet		
1	9·6	0·16	1·0
3	8·9	0·10	0·9
5	8·2	0·09	1·0
7	7·7	0·07	0·9
9	7·2	0·06	0·8
11	6·9	0·06	0·8
	Maize-cube diet		
1	12·4	0·14	1·7
3	12·4	0·09	1·7
5	11·7	0·14	1·4
7	11·2	0·15	1·3
9	10·7	0·12	1·1
11	10·3	0·06	1·1

same 1 h after feeding and 3 h after feeding for samples from the animal fed on maize cubes is due to one of the difficulties that may occur with pelleted diets. The pellets are eaten very rapidly and can be swallowed whole or only partly chewed. We have on occasion found quantities of pellets that were completely dry inside. Under these circumstances it is impossible to make a 9 g sample representative of the whole since the pellets weigh about 2·5 g each. There is a high probability of drawing a sample in which the proportion of dry matter is less than that of the rumen as a whole and a low probability of drawing a sample with a dry-matter content much greater than that of the rumen as a whole. The probability of obtaining a sample of 9 g wet weight containing a pellet, even when half of the pellets have not disintegrated but are circulating freely, is of the order of 1 in 10. Occasionally a blockage of the pump has occurred when a pellet has lodged in the valves. The problem of disintegration of the pellets can be overcome by using pellets loosely compacted or by offering the feed gradually so that the pellets are chewed.

The material sampled with the pump is, of course, withdrawn from a central position towards the caudal end of the rumen (see Fig. 2). This material must by the nature of the position of the return pipe be replaced by material from the surrounding region and not directly from the material that has been returned to the rumen, so that

sampling from the pump at successive time intervals is effectively sampling different portions of the rumen contents. Material drawn from the originally thinner regions of the rumen contents has to pass through the originally thicker upper region before it can return to the sampling point. The reproducibility of the dry-matter samples taken at intervals when a flow of 5–20 l. has gone through the pump is therefore fairly good evidence of attainment of an even distribution of dry matter in the rumen.

Effects of thorough mixing on the concentration of various constituents of rumen contents. The comparisons of the mean values for pH, ammonia and total volatile fatty acids are given in Tables 3, 4 and 5. The percentage composition of the volatile acids present are given in Table 6. The results for each type of sampling are, for the animal fed on grass cubes, the means of four experiments.

Table 3. *Comparisons of the pH of sheep rumen contents withdrawn by the conventional method and by the pump technique*

(Mean values with their standard errors for four experiments with each sampling method for the animal fed on grass cubes and for five experiments with each sampling method for the animal fed on maize cubes)

Time after feeding (h)	Animal fed on grass cubes		Animal fed on maize cubes	
	Conventional method	Pump	Conventional method	Pump
0·15	6·03 ± 0·04	6·00 ± 0·05	5·87 ± 0·08	5·80 ± 0·09
0·45	6·04 ± 0·05	6·02 ± 0·02	5·66 ± 0·08	5·67 ± 0·07
1·30	6·07 ± 0·07	6·08 ± 0·03	5·60 ± 0·07	5·56 ± 0·06
2·30	6·13 ± 0·12	6·13 ± 0·03	5·51 ± 0·07	5·57 ± 0·06
3·30	6·22 ± 0·10	6·20 ± 0·03	5·53 ± 0·08	5·61 ± 0·13
4·30	6·24 ± 0·10	6·29 ± 0·03	5·62 ± 0·13	5·68 ± 0·17
5·30	6·42 ± 0·10	6·35 ± 0·03	5·67 ± 0·12	5·75 ± 0·19
6·30	6·47 ± 0·09	6·41 ± 0·04	5·74 ± 0·10	5·85 ± 0·21
7·30	6·59 ± 0·10	6·47 ± 0·04	5·81 ± 0·11	5·95 ± 0·21
8·30	6·66 ± 0·08	6·58 ± 0·03	6·00 ± 0·13	6·06 ± 0·27
9·00	6·68 ± 0·07	6·63 ± 0·03	6·03 ± 0·13	6·10 ± 0·21

Table 4. *Comparisons of the concentration of ammonia (m-equiv./l.) in the liquid of samples of sheep rumen contents withdrawn by the conventional method and by the pump technique*

(Mean values with their standard errors for four experiments with each sampling method for the animal fed on grass cubes and for five experiments with each sampling method for the animal fed on maize cubes)

Time after feeding (h)	Animal fed on grass cubes		Animal fed on maize cubes	
	Conventional method	Pump	Conventional method	Pump
0·30	13·2 ± 1·9	14·3 ± 0·5	18·4 ± 0·9	16·1 ± 1·5
1·30	8·6 ± 1·9	11·0 ± 0·8	14·3 ± 2·8	12·6 ± 1·9
3·30	1·3 ± 0·8	2·0 ± 0·5	9·6 ± 3·0	9·2 ± 2·1
5·30	0·4 ± 0·2	1·3 ± 0·4	7·0 ± 1·4	9·2 ± 2·0
7·30	2·1 ± 0·6	4·0 ± 0·9	7·3 ± 0·5	9·9 ± 2·1
9·30	4·3 ± 0·7	7·5 ± 1·5	9·9 ± 0·9	11·3 ± 2·4

Effects of thorough mixing on the digestibility of the diet. The results of the digestibility trials in which faecal organic matter, 'normal acid fibre' and N and also urinary N were determined are given in Table 7. The values listed are the mean values for 3-day periods. The results are for eight periods when the pump was not used and six periods when the pump was used for 8 h/day for the animal fed on the diet of grass cubes.

Table 5. *Comparison of the concentration of the total volatile fatty acids (m-equiv./l.) in the liquid of samples of sheep rumen contents withdrawn by the conventional method and by the pump technique*

(Mean values with their standard errors for four experiments with each sampling method for the animal fed on grass cubes and for five experiments with each sampling method for the animal fed on maize cubes)

Time after feeding (h)	Animal fed on grass cubes		Animal fed on maize cubes	
	Conventional method	Pump	Conventional method	Pump
0·30	120 ± 4	112 ± 4	97 ± 9	103 ± 6
1·30	119 ± 6	115 ± 4	118 ± 8	122 ± 5
3·30	96 ± 4	98 ± 3	120 ± 9	121 ± 9
5·30	93 ± 5	87 ± 3	109 ± 7	113 ± 11
7·30	78 ± 7	76 ± 2	99 ± 6	99 ± 10
9·30	67 ± 7	68 ± 2	88 ± 6	88 ± 11

Table 6. *Comparisons of the percentage composition of the volatile acids of sheep rumen contents withdrawn by the conventional method and by the pump technique*

(Mean values with their standard errors for four experiments of each type for the animal fed on grass cubes and for five experiments of each type for the animal fed on maize cubes)

Time after feeding (h)	Conventional method			Pump		
	Acetate	Propionate	Butyrate	Acetate	Propionate	Butyrate
	Animal fed on grass cubes					
0·30	62·6 ± 1·5	27·9 ± 0·8	9·1 ± 1·0	60·9 ± 0·4	28·6 ± 0·5	8·7 ± 0·2
1·30	63·0 ± 0·7	25·9 ± 1·3	10·2 ± 1·2	63·7 ± 1·3	27·2 ± 1·2	8·3 ± 0·3
3·30	65·0 ± 1·4	22·4 ± 1·8	11·8 ± 1·0	65·8 ± 0·8	23·7 ± 0·6	10·0 ± 0·3
5·30	69·4 ± 1·1	18·8 ± 1·1	10·7 ± 0·5	70·2 ± 0·3	19·2 ± 0·3	10·1 ± 0·3
7·30	69·9 ± 1·2	18·6 ± 1·6	11·0 ± 0·3	68·0 ± 1·5	20·9 ± 1·5	10·0 ± 0·6
9·30	69·7 ± 0·6	17·7 ± 0·8	11·1 ± 0·4	69·6 ± 1·1	17·6 ± 0·5	12·0 ± 1·4
	Animal fed on maize cubes					
0·30	64·3 ± 1·7	19·7 ± 1·8	14·4 ± 0·3	64·7 ± 0·5	20·6 ± 1·3	13·5 ± 1·1
1·30	63·3 ± 1·8	21·0 ± 1·9	14·2 ± 1·1	63·5 ± 1·1	21·1 ± 1·2	14·1 ± 1·2
3·30	64·2 ± 0·7	20·1 ± 0·6	14·1 ± 1·5	63·5 ± 0·7	20·7 ± 0·7	14·5 ± 1·6
5·30	65·4 ± 2·1	19·3 ± 2·0	13·4 ± 1·7	64·5 ± 1·7	19·7 ± 0·8	14·7 ± 1·2
7·30	65·0 ± 1·4	18·6 ± 1·5	14·4 ± 1·5	65·4 ± 1·1	18·5 ± 1·0	14·5 ± 1·2
9·30	65·1 ± 1·4	18·3 ± 1·3	14·6 ± 1·0	65·8 ± 1·2	17·7 ± 0·8	14·2 ± 1·4

The values listed for the animal fed on the diet of maize cubes are the means of the results for six periods when the pump was not used and four periods when the pump was in use for 8 h/day.

No difference between treatments was found in the digestibility of organic matter or fibre but there appeared to be trends of divergence in the distribution of N between faeces and urine. The divergences were, however, in opposite directions in the two animals.

Table 7. *Effects of continuous mixing of the rumen contents of sheep by pumping on digestibility of the components of the diet*

(Mean values with their standard errors determined for a series of 3-day collection periods. Figures in parentheses are the numbers of 3-day periods)

	Without pump	With pump
Grass-cube diet		
Organic matter, digestibility coefficient (%)	70·0 ± 0·8 (8)	70·6 ± 0·6 (6)
Normal acid fibre, digestibility coefficient (%)	59·1 ± 2·2 (8)	61·9 ± 1·1 (6)
Ratio, faecal N:dietary N (%)	30·3 ± 0·9 (8)	31·1 ± 0·5 (6)
Ratio, urinary N:dietary N (%)	51·6 ± 1·9 (8)	55·4 ± 1·4 (6)
Maize-cube diet		
Organic matter, digestibility coefficient (%)	84·1 ± 0·4 (6)	83·3 ± 1·5 (4)
Normal acid fibre, digestibility coefficient (%)	73·1 ± 0·7 (6)	72·3 ± 2·8 (4)
Ratio, faecal N:dietary N (%)	25·8 ± 0·6 (6)	27·4 ± 1·8 (4)
Ratio, urinary N:dietary N (%)	54·8 ± 3·2 (6)	47·7 ± 1·5 (4)

DISCUSSION

The method described here is a simple, convenient and rapid means of obtaining small but representative samples from the rumen of the sheep. Solutions added to the rumen under the conditions described are rapidly mixed with the rumen liquid. The method is therefore well suited to the determination of rumen volume by dilution methods. The samples being truly representative are suitable also for short-term in vitro incubations of the type proposed by Carroll & Hungate (1954) and Stewart, Stewart & Schultz (1958).

Questions arise, however, as to how far the extreme artificial mixing imposed on the rumen affects the physical state of the animal, the rates of metabolism in the rumen and the rates of passage of digesta along the gastro-intestinal tract. Animals that have been used regularly with the apparatus once or twice a week for periods of several hours have remained in excellent health over the last year. The apparatus caused no obvious distress when in place and did not restrict the movement of the animal except that it was held in a standing position in a stall. The weight of the apparatus is largely supported by the guys and does not appear to cause any discomfort.

It would be surprising if the change from a system in the rumen in which there are gradients of pH, rH, and concentration of metabolites to a more uniform distribution did not result in some changes of metabolism, but the experiments comparing the concentration of components in the rumen indicated that these changes were not very large. Ammonia was the only component investigated to show any appreciable difference in concentration between sampling methods. It is impossible to say which of the many processes that might influence the ammonia concentration, such as salivary secretion, rate of proteolysis, rate of deamination of amino acids or rate of incorporation of ammonia into other metabolites, were affected by the alteration in sampling method. It would perhaps not be amiss to point out that the conventional method of sampling does to some extent affect physiological normality in that it involves

a considerable loss of the normal rumen gas and its replacement by air. The need for a certain degree of caution, however, in applying the new method to studies of N metabolism is probably indicated.

It might, at first sight, be expected that, while the pump is in use and some 500–600 ml of the rumen contents are outside the animal in the pump circuit, the rates of absorption of substances from the rumen would be decreased. The absorption rate, however, is more a function of the area of rumen wall effectively wet by the rumen contents than of the volume of rumen contents. It would appear from the results obtained in this work that the greater agitation of the rumen contents during the experiments with the pump in place effectively compensated for the smaller volume of rumen contents. The fall in the liquid level would in any event be in a region where the rumen wall is not heavily papillated and so has a relatively small effective surface area.

The results of the digestibility trial did not show any significant differences in digestibility of organic matter or fibre, which indicates that there was little interference with the normal processes of digestion and that the method should be useful in the study of ruminant digestion.

SUMMARY

1. A pump that can circulate the rumen contents of sheep fitted with a large-bore rumen cannula was built and is described. The apparatus is designed so that solutions may be added to a continuously flowing stream of rumen contents and so be rapidly mixed with the contents. It is so designed that samples representative of the whole contents can be easily withdrawn from the pump circuit. The method is applicable only when the food of the sheep is ground to a fairly fine consistency.

2. Experiments were carried out to test the efficiency of the apparatus in mixing added polyethylene glycol with the rumen contents.

3. The effects of the apparatus on the concentration of various metabolites in the rumen were examined.

4. The effect of the apparatus on the overall digestibility of the diet was investigated.

5. The apparatus appeared to have little effect on metabolism in the rumen or on digestibility, except possibly on the metabolism of nitrogen.

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EXPLANATION OF PLATE

Pl. 1. The rumen perfusion pump in position on the sheep. The bellows and motor are shown (right) and an injection apparatus (lower centre).

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